Impact of pre and post emergence herbicide application on soil microbial population assessment in direct wet seeded rice

Abstract

Direct wet seeded rice is eminent due to low energy and cost required for the cultivation. The only disadvantage is difficulty to manage weeds due to which farmers rely mostly on herbicides for the effective management. Herbicides are bioactive compounds which will affect the soil microflora by its ecotoxicological behaviour. Hence, in this study herbicides such as pendimethalin, pretilachlor + pyrazosulfuron ethyl (tank mix) as pre-emergence and bispyribac sodium + ethoxy sulfuron (tank mix), penoxsulam + cyhalofop butyl (ready mix), trifamone + ethoxy sulfuron (ready mix), bispyribac sodium as post emergence herbicides in their regular dosage were evaluated for their impact on soil microbial population viz; bacteria, fungi and actinomycetes. The results revealed that these herbicides affected the microbial population during the initial days but later the population was regained at 55 DAS which indicates the reduction in toxicity level of the applied herbicides.

Keywords: pre- emergence herbicides, post- emergence herbicides, direct wet seeded rice, microbial count, herbicidal degradation

1. INTRODUCTION

In India, direct seeded rice is gaining popularity due to its less consumption of energy, early maturity, less methane emission, improved soil health, faster and easier planting (Kumar and Ladha, 2011). The major disadvantage of this system is difficulty to maintain weeds which makes herbicide application necessary (Abd El- Naby *et al.,* 2017).

Herbicide application leaves a large portion of the residual chemicals in the top layer of the soil (0-15 cm), where most of the microbial activity takes place (Alexander, 1977). According to Das and Debnath (2006), the accumulation of chemicals in the soil might affect the soil microbial population. Since microbes are scavengers or heterotrophs in soil, which decomposes herbicides and derive energy, carbon, and other nutrients from the soil for efficient metabolism and vigorous growth. This increases biomass of microbes and transforms the plant nutrients in soil (Nongthombam *et al.*, 2009).

Herbicide residues may have adverse effect on plants, animals and human beings. According to World Health Organization (WHO), “any substance or mixture of substances in food for man or animals resulting from the use of a pesticide and includes any specified derivatives, such as degradation and conversion products, metabolites, reaction products, and impurities that are considered to be of toxicological significance” is defined as herbicide or pesticide residue (Sondhia, 2014). Application of pendimethalin in field peas and chickpea showed a half life of 11.23-19.83 days (Sondhia, 2013).

Incidences of acute herbicide self poisoning is common in Asia due to butachlor, fluchloralin, paraquat, 2,4-D, pendimethalin, glyphosate etc. (Senarathna *et al*., 2009). Nair *et al.* (2005), reported that 2,4- D causes higher DNA damage and chromosomal aberrations in human lymphocytes. Similarly, herbicides such as butachlor has some genotoxic effects which is harmful for fishes as it alters the chromosome number and affects the cellular activities (Yadav *et al*., 2013). The herbicide residues are accumulated in the tissues of fish which leads to biomagnification by food chain (Tilak *et al.*, 2007).

On the other hand, some research evidences exhibit that herbicides show stimulating effect on growth and metabolism of non-symbiotic nitrogen fixing bacteria (Das *et al*., 2012) and phosphate solubilizing microbes (Das and Debnath, 2006). At this juncture, this study is necessary to identify whether herbicide application suppress or enhance/ stimulate the soil microbial population.

To determine the impact of herbicide application on soil microflora, it is imperative to know the count of microbial population at different time of application period. The objective of the present study is to investigate the changes in microbial population by the effect of two pre-emergence herbicides pendimethalin and tank combination of pretilachlor + pyrazosulfuron and the effect of post emergence herbicides such tank mix (bispyribac sodium + ethoxy sulfuron), ready mix (penoxsulam + cyhalofop butyl), ready mix (triafamone + ethoxysulfuron), bispyribac sodium. These treatments were compared with the non herbicidal treated plots such as hand weeded plot, weed free plot and weedy check.

2. Materials and methods

Field experiment has been conducted at wetland farm, Tamil Nadu Agricultural University, Coimbatore during Summer, 2022, to study the efficient herbicidal treatment in wet direct seeded rice. The experiment comprised of 10 weed control treatments viz; pendimethalin 1 kg ha^{-1} at 3 DAS *fb* tank mix of bispyribac sodium 25 g ha^{-1} + Ethoxy sulfuron 18g ha^{-1} on 20 DAS (T_1); Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Penoxulam + Cyhalofop butyl 135 g ha^{-1} on 20 DAS (Ready mix) (T_2); Pendimethalin 1 kg ha^{-1} on 3 DAS *fb* Triafamone + Ethoxy sulfuron 66.5 g ha^{-1} on 20 DAS (Ready mix) (T_3); Pretilachlor 600 g ha^{-1} + Pyrazosulfuron ethyl 15 g ha^{-1} on 3 DAS (Tank mix) + HW on 40-45 DAS (T_4); Pendimethalin 1 kg ha^{-1} on 3 DAS + HW on 40-45 DAS (T_5); Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25 g ha^{-1} on 25 DAS + HW on 45 DAS (T_6); Triafamone + Ethoxy sulfuron 66.5 g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS (T_7); HW twice (20 and 40 DAS) (T_8); Weed free control (T_9); Weedy check (Unweeded control) (T_{10}).

Microbial population count of bacteria, fungi and actinomycetes was done at 15 DAS, 35 DAS and 55 DAS.The microbial count was determined by the serial dilution and pour plating method. Bacterial population was estimated by nutrient agar method (Kawser, 2016), fungal population was cultured on rose bengal agar (Martin, 1950), actinomycetes population was enumerated using Kenknight culture media (Dinesh *et al.,* 2017). The composition of media is given in Annexure 1. After incubation period, the colonies were counted and the number of viable bacteria, fungi and actinobacteria [expressed as colony forming units (cfu)] per gram dry weight of soil was estimated by considering the soil dilutions.

The original values were log transformed and the statistical analysis for different treatments was done by using OP Stat software. The least significance difference (LSD) at 5% level of significance was used for analysis of variance for randomised block design (Cochran and Cox 1957).

Annexure 1: Composition of culture media

S.No.	Culture name	Composition
1.	Nutrient agar medium	1 g glucose 3 g beef extract 5 g peptone 20 g agar 5 g NaCl 1000 mL distilled water pH adjusted to 7
2.	Rose Bengal medium	10 g glucose 5 g peptone 1 g KH_2PO_4 0.05 g $MgSO_4.7H_2O$ 0.033 g rose bengal Streptomycin 30 mg 15 g agar 1000 mL distilled water pH adjusted to 5.5
3.	Kenknight culture media	1 g dextrose 0.1 g KH_2PO_4 0.1 g $NaNO_3$ 0.1 g KCl 15 g agar 1000 mL distilled water pH adjusted to 7.0-7.2

3. Results and discussion

After the application of pre- emergence herbicides (pendimethalin and tank mix of pretilachlor + pyrazosulfuron ethyl) the bacterial count decreased drastically as compared to non herbicide applied plots at 15 DAS. The bacterial count in pendimethalin applied plots recorded 1.25×10^5 cfu/g of soil and for pretilachlor + pyrazosulfuron ethyl with 1.26×10^5 cfu. Similarly, for fungi and actinomycetes the count decreased which coincides with the findings of Latha and Gopal (2010), where bacterial population decreased after 15 days of herbicide application and fungal and actinomycetes population was also less than the control. But, after the application of post emergence herbicides at 35 DAS there was a slight increase in the fungal population. The bacterial and actinomycetes count was decreased in pendimethalin *fb* tank mix of bispyribac sodium + ethoxy sulfuron, the fungal population of pretilachlor + pyrazosulfuron ethyl regained population after 20 days of herbicide application which is supported by Simerjeet *et al.* (2014), that pyrazosulfuron ethyl recovered the population of fungi at 30 DAS. After the application of post emergence herbicides at 35 DAS there was decrease in the microbial population and then at 55 DAS it was found that bacterial and actinomycetes population (Table1,2) of all the treatments regained treatment Pendimethalin 1kg ha^{-1} *fb* Bispyribac sodium 25g ha^{-1} + Ethoxy sulfuron18g ha^{-1} (tank mix) [T_1], Pretilachlor 600g ha^{-1} + Pyrazosulfuron ethyl 15g ha^{-1} on 3 DAS (Tank mix) + HW on 40-45 DAS [T_4], Pendimethalin 1 kg ha^{-1} on 3 DAS + HW on 40-45 DAS [T_5], Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} on 25 DAS + HW on 45 DAS [T_6], Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS [T_7], but the fungal population decreased (Table 2). This was in confirmation with the findings of Sourav *et al.* (2014) who reported that bacterial and actinomycetes count proliferates with increase in oxidizable organic C as these organisms participates in herbicide utilization and degradation as compared to the fungal propagules.

Similarly, the bacterial, fungal and actinomycetes count was lower for pendimethalin *fb* cyhalofop butyl + penoxsulam (pre mix) initially, but the count was regained after 55 DAS (Table 1, 2, 3) which coincide with the findings of Ramalakshmi *et al.* (2017) that herbicides have some toxic effect just after the application but afterwards these microbes participate in degradation process by which degraded organic herbicides provide carbon rich substrates to increase the microbial population in soil and is greater than the control.

Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS [T_7] shows decrease in bacterial, fungal and actinomycetes count after 15 days of post emergence herbicides application but after 55 DAS it regained its population and was on par with the control. This is in confirmation with the findings of Gnanchitra *et al.* (2018) who reported that growth of rhizobium population was affected initially due to the residual impact of imidacloprid but after that it attained normal growth rate.

4. Conclusion

There will be a gradual regaining of the soil microbial population at the later stages of the crop growth due to herbicide application. Initially the count will be lower as the microbes will face the adverse effect of herbicides but substantially the population has been increased due to co metabolization of herbicides which is retained in soil as oxidisable carbon (Sourav *et al.,* 2014). Hence, it could be concluded that, in general the bacterial and actinomycetes population gets decreased due to herbicide application but when the toxicity level of the herbicides applied reduces the population slowly starts increasing, whereas the fungal population increases during herbicide degradation and later it decreases. Therefore, the toxicity of herbicides affects all the microbial population at the initial stage. Later at the reduction in the toxicity level the microbial population slightly increases due to the available carbon obtained after degradation.

Table 1: Effect of pre and post emergence herbicides on bacterial population in direct wet seeded rice

Treatments		Bacterial count X 10^5 cfu		
		15 DAS	35 DAS	55 DAS
T_1	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} + Ethoxy sulfuron18g ha^{-1} on 20 DAS (Tank mix)	17.8 (1.25)	12.5 (1.10)	25.5 (1.41)
T_2	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Penoxulam + Cyhalofop butyl 135g ha^{-1} on 20 DAS (Ready mix)	17.3 (1.23)	11.6 (1.06)	29.5 (1.47)
T_3	Pendimethalin 1 kg ha^{-1} on 3 DAS *fb* Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix)	16.3 (1.21)	10.5 (1.02)	27.3 (1.44)
T_4	Pretilachlor 600g ha^{-1} + Pyrazosulfuron ethyl 15g ha^{-1} on 3 DAS (Tank mix) + HW on 40-45 DAS	18.4 (1.26)	25.1 (1.40)	37.7 (1.58)
T_5	Pendimethalin 1 kg ha^{-1} on 3 DAS + HW on 40-45 DAS	16.5 (1.21)	26.5 (1.42)	35.2 (1.55)
T_6	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} on 25 DAS + HW on 45 DAS	16.9 (1.22)	13.4 (1.13)	33.5 (1.53)
T_7	Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS	34.1 (1.53)	18.4 (1.26)	36.4 (1.56)
T_8	HW twice (20 and 40 DAS)	34.2 (1.53)	34.9 (1.54)	37.4 (1.57)
T_9	Weed free control	33.9 (1.53)	35.9 (1.55)	35.3 (1.55)
T_{10}	Weedy check (Unweeded control)	34.3 (1.53)	35.3 (1.55)	36.5 (1.56)
C.D		0.018	0.015	0.014
S.E		0.008	0.007	0.007

(Values in parentheses are log_{10} transformed)

Table 2: Effect of pre and post emergence herbicides on fungal population in direct wet seeded rice

Treatments		Fungal count X 10^4 cfu		
		15 DAS	35 DAS	55 DAS
T_1	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} + Ethoxy sulfuron18g ha^{-1} on 20 DAS (Tank mix)	16.67 (1.26)	45.67 (1.66)	21.13 (1.32)
T_2	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Penoxulam + Cyhalofop butyl 135g ha^{-1} on 20 DAS (Ready mix)	17.00 (1.24)	36.00 (1.54)	21.30 (1.33)
T_3	Pendimethalin 1 kg ha^{-1} on 3 DAS *fb* Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix)	16.70 (1.25)	37.23 (1.57)	19.87 (1.30)
T_4	Pretilachlor 600g ha^{-1} + Pyrazosulfuron ethyl 15g ha^{-1} on 3 DAS (Tank mix) + HW on 40-45 DAS	18.50 (1.22)	52.09 (1.73)	27.17 (1.43)
T_5	Pendimethalin 1 kg ha^{-1} on 3 DAS + HW on 40-45 DAS	17.10 (1.26)	49.20 (1.69)	26.67 (1.42)
T_6	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} on 25 DAS + HW on 45 DAS	17.80 (1.26)	44.60 (1.65)	20.47 (1.31)
T_7	Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS	56.97 (1.76)	22.43 (1.35)	46.03 (1.66)
T_8	HW twice (20 and 40 DAS)	56.37 (1.75)	56.23 (1.46)	58.87 (1.44)
T_9	Weed free control	56.17 (1.75)	57.00 (1.75)	58.00 (1.77)
T_{10}	Weedy check (Unweeded control)	56.83 (1.76)	57.13 (1.77)	57.93 (1.77)
C.D		0.035	0.028	0.043
S.E		0.017	0.013	0.020

(Values in parentheses are log_{10} transformed)

Table 3: Effect of pre and post emergence herbicides on actinomycetes population in direct wet seeded rice

Treatments		Actinomycetes count X 10^3 cfu		
		15 DAS	35 DAS	55 DAS
T_1	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} + Ethoxy sulfuron18g ha^{-1} on 20 DAS (Tank mix)	12.33 (1.09)	7.35 (0.86)	14.23 (1.15)
T_2	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Penoxulam + Cyhalofop butyl 135g ha^{-1} on 20 DAS (Ready mix)	12.43 (1.09)	7.43 (0.87)	14.10 (1.15)
T_3	Pendimethalin 1 kg ha^{-1} on 3 DAS *fb* Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix)	12.43 (1.09)	6.65 (0.82)	13.63 (1.13)
T_4	Pretilachlor 600g ha^{-1} + Pyrazosulfuron ethyl 15g ha^{-1} on 3 DAS (Tank mix) + HW on 40-45 DAS	13.27 (1.12)	7.93 (0.90)	14.67 (1.16)
T_5	Pendimethalin 1 kg ha^{-1} on 3 DAS + HW on 40-45 DAS	12.33 (1.09)	10.33 (1.01)	14.43 (1.15)
T_6	Pendimethalin 1kg ha^{-1} on 3 DAS *fb* Bispyribac sodium 25g ha^{-1} on 25 DAS + HW on 45 DAS	12.50 (1.09)	6.93 (0.84)	13.87 (1.14)
T_7	Triafamone + Ethoxy sulfuron 66.5g ha^{-1} on 20 DAS (Ready mix) + HW on 40-45 DAS	15.60 (1.19)	6.97 (0.84)	13.97 (1.14)
T_8	HW twice (20 and 40 DAS)	15.27 (1.18)	15.00 (1.17)	15.50 (1.19)
T_9	Weed free control	15.33 (1.18)	15.43 (1.19)	15.47 (1.19)
T_{10}	Weedy check (Unweeded control)	15.33 (1.18)	14.93 (1.17)	15.50 (1.19)
C.D		0.018	0.028	0.017
S.E		0.008	0.013	0.008

(Values in parentheses are log_{10} transformed)

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