Short Research

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Studies on Phytotoxicity of Herbicides and Herbicide Mixtures and its effect on Yield of Direct-Sown Rice (*Oryza sativa* L.)

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Abstract

The present investigation was carried out at Institute ResearchFarmofICAR-NRRI, Cuttack (Odisha) during wet season of 2016 to study the phytotoxicity of a new herbicide molecule XR-848 Benzyl Ester along with its mixture with Cyhalofop Butyl in different doses and its effect on yield of direct-sown rice. The experiment was laid out in Randomized Block Design (RBD) with three replications and nine treatments viz. four herbicide mixtures(T_1-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 120(20+100)g ha^{-1}, T_2-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 150(25+125)g ha^{-1}, T_3-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 180(30+150)g ha^{-1} andT_4-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 360(60+300)g ha^{-1}), three alone herbicides(T_5-XR-848 Benzyl Ester 2.5% EC (w/v) @ 25g ha^{-1}, T_6-XR-848 Benzyl Ester 2.5% EC (w/v) @ 30g ha^{-1} andT_7-Bispyribac-Na 10% SC @ 30g ha^{-1}), one weed free (T_8)and an untreated weedy check (T_9).Under phytotoxicity analysis, visual observations (Phyotoxic score), chlorophyll content and SPAD meter readings at 10, 20 and 30 days after treatment (DAT) of herbicides were taken, in which the T_4proved to be the most phytotoxic causing a reduction of 29.41% in grain yield which was at par with the untreated weedy check. Whereas T_2 showed a minimal phyotoxicity at initial stage with a quick recovery within 30 DAT of herbicide and was proved to be the best herbicide mixture yielding at par with the weed free (T_8).

Keywords: Herbicide, herbicide mixture, phytotoxicity, yield, direct-sown rice

1. Introduction

Rice (*Oryza sativa* L.) is the principal source of food and income for the people of South-East Asia. India is the second largest producer of rice next to China with an annual growing area 45Mha and production 90 Mha which contributs nearly 45 per cent of total food grain production in the country (Singh et al., 2013). Direct-sown rice (DSR) is becoming more popular than the traditional methods of rice cropping (Kumar et al., 2016a,b; Kumar et al., 2015a,b; Singh et al., 2017). But the DSR is very prone to weed infestation as the weeds get ample opportunity to grow simultaneously with rice, right from the germination to maturity (Roy et al., 2011). In DSR, chemical weed control is getting a growing acceptance among the farmers as the traditional methods of weed control are non-economic as well as labour-intensive (Chatterjee et al., 2016). However, negative impacts on environment like weed flora shift and herbicide resistance are the major disadvantages of long term use of herbicides with same mode of action. This situation needs to evaluate different aspects of new molecules of herbicide along with its mixtures with well established herbicides. Post emergence application of different herbicides may lead initial injury up to 30% in rice (Thapa, 2012), such as leaf chlorosis and growth stunting during 7 to 14 days after application which disappears shortly (Rahman, 2016). Hence an attempt was made to evaluate a new molecule of herbicide i.e. XR-848 Benzyle Ester and its mixture with a well-established herbicide from aryloxyphenoxypropionate group i.e. Cyhalofop-butyl for their phytotoxicity and effect on yield in wet DSR.

2. Materials and Methods

The experiment was conducted at Institute Research Farm of ICAR-National Rice Research Institute, Cuttack (Odisha) (20°272''102.2' N, 85°562''92.2' E; 24 m above mean sea level) during wet season of 2016. The experiment soil was sandy clay loam with pH 7.8 with low available N (215.4 kg ha^{-1}), medium available P (48 kg ha^{-1}), high available P (322.8 kg ha^{-1}) and medium organic carbon (0.52%). The experiment was laid out in randomized complete block design (RCBD) with three replications. The gross and net plot size were 6.0 m×5.0 m=30 m^2 and 5.1 m×4.0 m=20.4 m^2,respectively in which sprouted seeds of Rice (var. *Naveen*, Indica type) were directly sown manually with a seed rate of 80 kg ha^{-1}. There were nine treatments consisting of well established herbicide (T_7-Bispyribac-Na 10% SC at 30 g ha^{-1}), new herbicide at two doses (T_5-XR-848 Benzyl Ester 2.5% EC (w/v) at 25g ha^{-1} and T_6-XR-848 Benzyl Ester 2.5% EC (w/v) at 30g ha^{-1}) and four herbicide mixtures of different doses (**T_1**-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) at 120(20+100) g ha^{-1}, T_2-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) at 150(25+125)g ha^{-1}, T_3-XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) at 180(30+150)g ha^{-1}, T_4-XR-848 Benzyl Ester+CyhalofopButyl 12% EC (w/v) at 360(60+300)g ha^{-1}) along with T_8-Weed Free and untreated T_9-Weedy check. The new herbicides XR-848 Benzyl Ester 2.5% EC (w/v) and formulated herbicide mixture XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) were obtained from Daw AgroSciences India Pvt Ltd. All the chemical treatments were applied at 2–3 leaf stage of appearance of weeds. The untreated weedy check was kept undisturbed during the entire cropping period. Recommended fertilizer application ($N:P_2O_5:K_2O$::100:60:40 kg ha^{-1}) was followed, with full dose of P and K application as basal during final land preparation and the N fertilizer application in four equal splits at 15, 30, 45 and 60 days after sowing.

2.1. Phytotoxicity

In order to measure phytotoxicity, three methods viz. visual observation (phytotoxic score), chlorophyll content analysis and SPAD chlorophyll meter readings were taken at 10, 20 and 30 days after treatment (DAT) of the above mentioned post emergence herbicides.

2.1.1. Visual observations(Phytotoxic score)

Visual observations on phytotoxicity parameters were recorded at 10,20 and 30 DAT of herbicides in the plots treated with T_1 to T_7 by 5 persons separately watching each plot and then averaged to obtain the mean values. Phytotoxicity symptoms viz., leaf injury on tips/surface, wilting, necrosis and chlorosis were recorded in all treatments. The phytotoxicty score was recorded using 1–10 scale (where 1=1–10 % damage and 10=91–100 % damage).

2.1.2. Chlorophyll content

Fresh leaveswere taken from plants of each plot at 10, 20 and 30 DAT of herbicide. 25 mg of shredded leaf was taken from each sample and kept in 10 ml of 80% acetone solution in 15 ml centrifuge tubes for 48 hours in darkness. Then the tubes were centrifuged in 3000 rpm for 15 minutes at 4°C. After completion of centrifugation the supernatants were separated and analysed for chlorophyll-a and chlorophyll-b in spectrophotometer in 645 and 663 nm wave lengths (Porra et al., 1989). Determination of chlorophyll-a and chlorophyll-b were done by using the following formulae 1 and 2. Then total chlorophyll (Chlorophyll-a+Chlorophyll-b) contents were calculated.

Chlorophyll - a (µg/ml)=[{(12.7 X OD_{663})–(2.69 X OD_{645})} **/** leaf weight (mg)] **×** 0.4….. (1)

Chlorophyll - b (µg/ml)=[{(22.9 X OD_{645})–(4.68 X OD_{663})} **/** leaf weight (mg)] **×** 0.4….. (2)

Where,

OD_{663}=Spectrophotometer reading at 663 nm wave length

OD_{645}=Spectrophotometer reading at 645 nm wave length

2.1.3. SPAD chlorophyll-meter readings

Five randomly selected rice plants were selected from each plot and SPAD chlorophyll-meter observations were taken from five leaves from each selected plantusing SPAD 502 Plus Chlorophyll-meter at 10, 20 and 30 DAT of herbicides.

2.2. Yield

After physiological maturity, the crop from each net plot was harvested separately. The grains were separated from straw by threshing. The weight of grains was recorded and expressed in t ha^{-1}.

3. Results and Discussion

3.1. Visual observations (phytotoxic score)

Phytotoxic symptoms on rice leaves in terms of injury in leaf tip/surface, wilting, necrosis and chlorosis as observed in the herbicide treated plots at 10, 20 and 30 DAT of herbicides are presented in the Table 1. The highest phytotoxic symptoms were found in the herbicide mixture XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 360(60+300)g ha^{-1} (T4) in which the scores were 1, 2, 3 and 5 followed by herbicide mixture XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v) @ 180(30+150) g ha^{-1} (T_3) in which the scores were 1, 1, 1 and 2 for injury in leaf tip/surface, wilting, necrosis and chlorosis respectively at 10 DAT of herbicides. In the other herbicide treatments normally 1 to 10% phytotoxicity was seen at 10 DAT of herbicides. At 20 DAT of herbicides, no injury in leaf tip/surface and wilting was observed in the field except the plots treated with T_4 in which 1–10% symptoms were observed. 1–10% symptoms as necrosis and chlorosis were observed in all herbicide treated plots except the T_4 treated plot where the symptoms were 10-20%. However, at 30 DAT of herbicides the crop was found to recover totally and no phytotoxic symptoms were observed.

3.2. Chlorophyllcontent analysis

The data regarding the chlorophyll content analysis are presented in Table 2. The data revealed that at 10 DAT of herbicides, the total chlorophyll content in rice leaves was found to be highest in the weed free plot (T_8) (9.19 µg ml^{-1}) which was at par with the weedy check (T_9) (8.50 µg ml^{-1}) and T_5 (8.09 µg ml^{-1}) followed by T_6 (7.35 µg ml^{-1}), T_1 (7.07 µg ml^{-1}), T_7 (6.74 µg ml^{-1}) and T_2 (5.96 µg ml^{-1}). The lowest chlorophyll content was found under T_4 (4.39 µg ml^{-1}) followed by T_3 (5.49 µg ml^{-1}). At 20 DAT of herbicides, the treatments i.e. T_8 (12.22 µg ml^{-1}), T_5 (10.799 µg ml^{-1}) and T_6 (9.940 µg ml^{-1}) were found to be at par with T_9 (11.67 µg ml^{-1}), T_1 (10.38 µg ml^{-1}) and T_7 (9.58 µg ml^{-1}) respectively. The lowest chlorophyll content was found under T_4 (7.42 µg ml^{-1}) followed by T_3 (8.66 µg

Table 1: Phytotoxicity in terms of visual observations (Phytotoxic score)

Treat-ments	Dose (g ha^{-1})	Phytotoxic score											
		10 DAT				20 DAT				30 DAT			
		I	W	N	C	I	W	N	C	I	W	N	C
T_1	120 (20+100)	1	1	1	1	0	0	1	1	0	0	0	0
T_2	150 (25+125)	1	1	1	1	0	0	1	1	0	0	0	0
T_3	180 (30+150)	1	1	1	2	0	0	1	1	0	0	0	0
T_4	360 (60+300)	1	2	3	5	1	1	2	2	0	0	0	0
T_5	25	1	1	1	1	0	0	1	1	0	0	0	0
T_6	30	1	1	1	1	0	0	1	1	0	0	0	0
T_7	30	1	1	1	1	0	0	1	1	0	0	0	0
T_8	-	-	-	-	-	-	-	-	-	-	-	-	-
T_9	-	-	-	-	-	-	-	-	-	-	-	-	-

I: leaf injury on tips/surface; W: Wilting; N: Necrosis; C: Chlorosis; T_1: XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v); T_2: XR-848 Benzyl Ester + Cyhalofop Butyl 12% EC (w/v); T_3: XR-848 Benzyl Ester + Cyhalofop Butyl 12% EC (w/v); T_4: XR-848 Benzyl Ester+Cyhalofop Butyl 12% EC (w/v); T_5: XR-848 Benzyl Ester 2.5% EC (w/v); T_6: XR-848 Benzyl Ester 2.5% EC (w/v); T_7: Bispyribac-Na 10% SC; T_8: Weed free; T_9: Weedy check

Table 2: Phytotoxicity in terms of total chlorophyll content (μg ml^{-1}) and SPAD chlorophyll-meter readings; and Grain yield (t ha^{-1})

Treatments	Dose (g ha^{-1})	Total Chlorophyll content (μg ml^{-1})			SPAD chlorophyll-meter readings			Grain Yield (t ha^{-1})
		10 DAT	20 DAT	30 DAT	10 DAT	20 DAT	30 DAT	
T_1	120 (20+100)	7.07	10.38	9.45	31.07	32.32	38.50	4.68
T_2	150 (25+125)	5.96	9.02	8.85	29.24	31.82	37.60	4.91
T_3	180 (30+150)	5.49	8.66	8.35	28.20	31.22	36.53	4.17
T_4	360 (60+300)	4.39	7.422	7.40	26.73	30.90	33.73	3.72
T_5	25	8.09	10.79	9.84	32.94	34.03	41.40	4.46
T_6	30	7.35	9.94	9.52	32.05	32.81	39.50	4.74
T_7	30	6.74	9.58	9.11	30.34	32.14	38.00	4.36
T_8	-	9.19	12.22	12.18	33.52	35.71	43.03	5.27
T_9	-	8.50	11.67	10.13	33.25	34.24	40.23	3.14
SEm±		0.40	0.27	0.39	1.02	0.97	1.02	0.17
CD (p=0.05)		1.20	0.83	1.17	3.06	2.91	3.07	0.52

*DAT=Days after treatment of herbicides

ml^{-1}) and T_2 (9.02 μg ml^{-1}). Similarly at 30 DAT of herbicides, the total chlorophyll content was highest under T_8 (12.18 μg ml^{-1}) followed by T_9 (10.13 μg ml^{-1}) which was at par with T_5 (9.84 μg ml^{-1}), T_6 (9.52 μg ml^{-1}), T_1 (9.45 μg ml^{-1}) and T_7 (9.11 μg ml^{-1}). The lowest total chlorophyll content was observed under T_4 (7.40 μg ml^{-1}) followed by T_3 (8.35 μg ml^{-1}) and T_2 (8.85 μg ml^{-1}).Analyzing the results,it is concluded that among the herbicide mixtures, T_4 and T_3 showed a delayed recovery compared to the other two i.e. T_2 and T_1. Whereas, among the alone applied herbicides, $T_7$was found to be more phytotoxic than the other two i.e. T_5 and T_6. This finding was in agreement with XuePing et al. (2000) who had found that 1-3 leaf stage of rice seedlings were highly vulnerable to Bispyribac-Na showing higher phytotoxicity.

3.3. *SPAD chlorophyll-meter readings*

Data regarding SPAD meter readings are presented in Table 2. The data revealed that at 10 DAT of herbicides, the highest SPAD values were obtained under T_8 (33.52) which was found at par with T_9 (33.25), T_5 (32.94), T_6 (32.05) and T_1 (31.07) followed by T_7 (30.348) which was at par with T_2 (29.24) and T_3 (28.200). The lowest SPAD value was obtained from T_4 treated plot i.e.4.39. At 20 DAT of herbicides, the highest SPAD value was obtained under T_8 (35.71) which was at par withT_9 (34.240), T_5 (34.03) and T_6 (32.81) followed by T_7 (32.14) which was at par with T_1 (32.32), T_2 (31.82), T_3 (31.22) and the lowest one T_4 (30.90). Similarly at 30 DAT of herbicides, the highest SPAD value was observed under T_8 (43.03) which was at par with T_5 (41.40) and T_9 (40.23) followed by T_6 (39.50) which was at par with T_1 (38.50), T_7 (38.00), T_2 (37.60) and T_3 (36.53). The lowest SPAD value was observed in T_4 treated plot i.e. (33.73). In the experiment, minimal phytotoxic effects of T_5, T_6 and T_1 were observed which were found at par with the untreated plots and the T_2, T_3 and T_7 were though at par with each other, found a little more phytotoxic than the former set (T_5, T_6 and T_1) of observations. Whereas, under T_4, the highest phytotoxicity was observed.

3.4. Grain yield (t ha^{-1})

Data with respect to grain yield is presented in Table 2. It is clear from the data that the different weed management treatments significantly influenced the grain yield. Among different treatments, the weed free (T_8) (5.27 t ha^{-1}) proved significantly superior producing higher grain yield, but it was found at par with T_2 (4.91 t ha^{-1}) which was followed by the yields of plots treated with T_6 (4.74 t ha^{-1}), T_1 (4.68 t ha^{-1}), T_5 (4.46 t ha^{-1}) and T_7 (4.36 t ha^{-1}). T_3 (4.17 t ha^{-1}) and T_4 (3.72 t ha^{-1}) were next in order, which performed significantly better than the untreated weedy check (T_9) (3.14 t ha^{-1}) which was the lowest producing treatment. Higher phytotoxicity in T_4 and T_3may be the reason for lower yield of grain and effective control of weeds during the critical crop-weed competition period, may be the reason for which the T_2 yielded at par with the weed free treatment i.e. T_8.

4. Conclusion

The new molecule (XR-848 Benzyl Ester) alone i.e. T_5 and T_6 shows less phytotoxicity and more grain yield than Bipyribac-Na i.e. T_7. Whereas, among the herbicide mixtures, the new molecule in combination with Cyhalofop-butyl (T_2), though initially phytotoxic, shows a timely recovery, greater weed control and higher grain yield than the other herbicide combinations i.e. T_1, T_3 and T_4.

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