



Evaluation of Rice Genotypes for Tolerance to Imazethapyr Herbicide in Direct-Seeded Rice Systems

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Authors' contributions

This work was carried out in collaboration among all authors. Author GSK conceptualized the study and designed the experiment. Authors DRH, AH and THS carried out data collection, performed the analysis, and contributed to interpreting the results. The manuscript was drafted by Authors GS and GSK, reviewed by Authors SM, CB, RSP and PP. All authors read and approved the final manuscript.

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ABSTRACT

Aims: The aim of this study was to evaluate the resistance of different rice genotypes to the herbicide imazethapyr.

Study Design: Pot culture experiment.

Place and Duration of Study: The experiment was conducted in pot culture at the Department of Agronomy, Tamil Nadu Agricultural University, Coimbatore, Kharif 2024.

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Methodology: Pot culture experiment was conducted with three replications by using fourteen rice genotypes (G1, G2, G3, G4, G5, G6, G7, G8, G9, G10, G11, G12, G13, and G14) to assess the phyto-toxicity tolerance. A herbicide spray was applied at the 2-3 leaf stage of weeds in the pot culture. Imazethapyr 10% SL was used at a concentration of 2.5 ml/L, applied at 23 days after sowing (DAS). The herbicide solution was prepared by adding 5 ml of Imazethapyr to 2 L of water. The spray was applied using a 500 ml capacity hand sprayer. The experiment was carried out under controlled (Field) environmental conditions to assess the response of rice genotypes in terms of phytotoxicity, including any visual damage or growth reduction caused by the herbicide treatment. The data collected were used to identify tolerant and susceptible rice genotypes to imazethapyr herbicide.

Results: The results revealed significant variation in tolerance among the rice genotypes. Genotype G14 exhibited the highest tolerance, displaying minimal phytotoxicity symptoms such as chlorosis and necrosis, and showed no adverse effects on growth. Moderate tolerance was observed in genotypes G1, G2, G3, G4, G5, G6, G7, and G8, which exhibited some injury but were able to recover and maintain growth potential. In contrast, the susceptible genotypes G9, G10, G11, G12, and G13 showed severe phytotoxicity symptoms and completely dried out.

Conclusion: The tolerant genotypes G1, G2, G3, G4, G5, G6, G7, G8 and G14, can be utilized for further rice breeding programme for the development of herbicide tolerant rice varieties.

Keywords: Direct seeded rice; genotypes; imazethapyr; phyto-toxicity; weed management.

1. INTRODUCTION

Rice is a key staple food for more than half of the world's population, particularly in Asia, where it sustains the livelihoods of 80 per cent of the rural population (Medina, 2011). It plays a vital socio-economic role in the region, being not only a main food source but also an essential part of cultural traditions. Sustainable rice production is crucial for ensuring long-term food security in Asia (Amanullah & Fahad 2017). Rice transplanting is a traditional farming practice where seedlings are grown in nurseries and then manually transplanted into flooded fields (Dixit et al., 2007). This method helps control weeds and creates optimal conditions for growth, but it is labour-intensive and faces challenges such as water shortages and a lack of available labour (Nagargade et al., 2018). Direct-seeded rice (DSR) is a promising alternative to traditional transplanting methods. It can reduce labour costs by up to 97% and decrease water consumption by 12-17% compared to puddled transplanted rice (Kumar et al., 2015).

Additionally, DSR helps lower greenhouse gas emissions and improves resource efficiency (Singh et al., 2024). This method also enables faster planting, optimizing time and resource use (Bista, 2018). On average, DSR requires four fewer irrigations than traditional methods, resulting in significant water savings (Ali et al., 2016). While DSR yields may be, on average, 12% lower than those of traditional transplanted rice (Xu et al., 2019).

Weed management presents a significant challenge in direct-seeded rice systems, as uncontrolled weeds can cause yield losses ranging from 40% to 100% (Rathika et al., 2020). The simultaneous growth of rice and weed seedlings, combined with the absence of standing water, exacerbates weed competition. Weeds are controlled through herbicides or manual labour. Manual weeding is less effective due to labour shortages and higher labour costs. Herbicides are replacing manual methods, but concerns include weed resistance, changing weed populations, high management costs and environmental impact (Hossain et al., 2016). Imazethapyr is typically applied as a post-emergence or early post-emergence treatment at 20-25 DAS in crops such as soybean, greengram, black gram, and sunflower to manage various weed species. It is particularly effective against annual and perennial grasses, as well as against certain weeds of the broadleaf type (Lal et al., 2017). Herbicides such as Imazethapyr, an imidazolinone-class compound, are widely used for their broad-spectrum control of weed species. Herbicide Imazethapyr is selective for pulse crops and non-selective for rice and other crops. Of the various herbicides, imidazolinones are the most widely targeted ones for developing herbicide-tolerant crops through a non-GM approach (Chandana et al., 2024; Kowsalya et al., 2022). Imazethapyr tolerant mutant resource was developed by EMS mutagenesis approach from a drought tolerant variety Nagina 22 which is named as HTM-N 22 (Shoba et al., 2017). Imazethapyr is a broad-

spectrum herbicide inhibiting aceto-lactate synthase, which reduces the production of branched-chain amino acids in weeds. Therefore, identifying rice varieties that are tolerant to imazethapyr is crucial for effective weed management in DSR systems, where weed infestations significantly reduce yields. The present study was conducted to evaluate the intensity of phytotoxicity caused by imazethapyr herbicide to identify the tolerant cultivars among the 14 different rice genotypes.

2. MATERIAL AND METHODS

An experiment was conducted at the Department of Agronomy, Tamil Nadu Agricultural University, Coimbatore, located at 11°N latitude and 77°E longitude, with an elevation of 426.7 m above sea level. The pot culture research was carried out during *Kharif* 2024, and aimed to evaluate the rice genotypes for their resistance to the herbicide Imazethapyr. Pot culture media consisted of clay loam soil chosen for its dry and tilled condition. During soil collection, debris was removed and 75 kg of the top 15 cm of soil was excavated to retain weed seeds. A hammer was used to break the clods and create a fine texture. A pot culture experiment was conducted with three replications by using fourteen rice genotypes (G1, G2, G3, G4, G5, G6, G7, G8, G9, G10, G11, G12, G13, and G14) to assess the phyto-toxicity tolerance. A total of 42 pots (8 inches in height) were arranged in three rows, with 14 pots per row. Each pot was filled with 2.5 kg of soil, measured using a weighing balance. Plant markers were used to label the pots. Seeds were randomly dibbled at a depth of 3 cm in each pot. After sowing, the first irrigation was applied, and a thin film of water was maintained. One week after germination, 0.5% urea solution was applied to the soil by dissolving 10g of urea in 2 litres of water and the solution was equally distributed across all the pots. To assess the phytotoxicity tolerance of different rice genotypes, imazethapyr herbicide spray was applied at the 2-3 leaf stage of weeds in the pot culture. Imazethapyr 10% SL was sprayed at a rate of 2.5 ml/litre, prepared by dissolving 5 ml of herbicide in 2 litres of water. The application was made using a 500 ml hand sprayer, 23 DAS. The rice seedlings began to show signs of drying 5 days after the herbicide application. The extent of drying was observed and recorded for 2 weeks following the spray. Phyto-toxicity was assessed using visual observations and a scorecard

method, recording symptoms such as leaf tip injury, wilting, chlorosis and necrosis in all pots. The percentage of crop injury was calculated using the formula:

$$\text{Percentage of crop injury} = \left(\frac{\text{Number of seedlings dried}}{\text{Number of seedlings germinated}} \right) \times 100$$

Weekly observations were averaged to calculate the mean values and phyto-toxicity was scored on a 1-10 scale. Tolerance was assessed by monitoring the changes in symptoms over time using these methods.

List 1. Phyto-toxicity scoring

Crop injury	Rating
0	0
1-10%	1
11-20%	2
21-30%	3
31-40%	4
41-50%	5
51-60%	6
61-70%	7
71-80%	8
81-90%	9
91-100%	10

3. RESULTS AND DISCUSSION

Among the 14 rice genotypes, significant variations in phyto-toxicity tolerance were observed over two weeks. In the first week, genotypes G2 and G14 showed tolerance, exhibiting minimal signs of toxicity, such as slight chlorosis and necrosis in a few seedlings. Genotypes G1, G3, G4, G5, G6, G7 and G8 were moderately tolerant, displaying mild symptoms of crop discoloration, in some seedlings, but remained relatively unaffected overall.

On the other hand, genotypes G9, G10, G11, G12 and G13 were highly susceptible, with most seedlings showing severe symptoms like chlorosis, necrosis, and drying, leading to significant crop damage (Table 1). By the second week, the phyto-toxicity situation evolved. Genotypes G1, G2, G3, G4, G5, G6, G7, G8 and G14 remained tolerant. Meanwhile, the genotypes G9, G10, G11, G12 and G13 remained susceptible, with severe toxicity effects continuing to damage the seedlings (Table 2).



Pic. 1. Genotype 14



Pic. 2. Genotype 2



Pic. 3. Genotype 9



Pic. 4. Genotype 12

Table 1. Evaluation of rice genotypes response to imazethapyr herbicide application in the first week on crop injury and phyto-toxicity analysis

Genotypes	1 DAH		2 DAH		3 DAH		4 DAH		5 DAH		6 DAH		7 DAH	
	%CI	PRS	%CI	PRS	% CI	PRS	% CI	PRS	%CI	PRS	%CI	PRS	%CI	PRS
Genotype 1	34.7	4	47.2	5	47.2	5	47.2	5	47.2	5	47.2	5	47.2	5
Genotype 2	8.3	1	8.3	1	8.3	1	8.3	1	8.3	1	8.3	1	8.3	1
Genotype 3	68.6	7	68.6	7	68.6	7	68.6	7	68.6	7	68.6	7	68.6	7
Genotype 4	42.5	5	42.5	5	42.5	5	42.5	5	50	5	50	5	50	5
Genotype 5	38.6	4	38.6	4	38.6	4	38.6	4	38.6	4	38.6	4	47.6	5
Genotype 6	46.7	5	46.7	5	46.7	5	55	6	55	6	55	6	55	6
Genotype 7	16.7	2	16.7	2	16.7	2	33.3	4	33.3	4	33.3	4	33.3	4
Genotype 8	16.7	2	16.7	2	35	4	53.3	6	63.3	7	63.3	7	63.3	7
Genotype 9	78.6	8	78.6	8	85.7	9	92.9	10	100	10	100	10	100	10
Genotype 10	53.3	6	53.3	6	61.7	7	61.7	7	71.6	8	71.6	8	76.7	8
Genotype 11	83.3	9	83.3	9	83.3	9	100	10	100	10	100	10	100	10
Genotype 12	30.5	3	30.5	3	30.5	3	44.4	5	55.5	6	55.5	6	80.5	8
Genotype 13	33.3	4	41.7	5	41.7	5	64.3	7	64.3	7	78.6	8	78.6	8
Genotype 14	0	0	0	0	0	0	0	0	0	0	0	0	0	0

DAH- Day after herbicide spray; CI – Crop injury; PRS – Phyto-toxicity rating scale

Table 2. Evaluation of rice genotypes response to imazethapyr herbicide application in the second week on crop injury and phytotoxicity analysis

Genotypes	8 DAH		9 DAH		10 DAH		11 DAH		12 DAH		13 DAH		14 DAH	
	% CI	PRS	% CI	PRS	% CI	PRS	% CI	PRS	% CI	PRS	% CI	PRS	% CI	PRS
Genotype 1	59.7	6	59.7	6	59.7	6	59.7	6	59.7	6	59.7	6	59.7	6
Genotype 2	8.3	1	37.5	4	54.2	6	54.2	6	54.2	6	57.5	6	57.5	6
Genotype 3	68.6	7	68.6	7	68.6	7	68.6	7	75.7	8	75.7	8	75.7	8
Genotype 4	50	5	62.5	7	62.5	7	62.5	7	62.5	7	75	8	75	8
Genotype 5	47.6	5	47.6	5	47.6	5	47.6	5	47.6	5	47.6	5	47.6	5
Genotype 6	60	6	60	6	73.3	8	73.3	8	73.3	8	73.3	8	73.3	8
Genotype 7	33.3	4	46.7	5	46.7	5	46.7	5	57.2	6	57.2	6	57.2	6
Genotype 8	63.3	7	63.3	7	73.3	8	73.3	8	73.3	8	73.3	8	73.3	8
Genotype 9	100	10	100	10	100	10	100	10	100	10	100	10	100	10
Genotype 10	81.7	9	86.7	9	100	10	100	10	100	10	100	10	100	10
Genotype 11	100	10	100	10	100	10	100	10	100	10	100	10	100	10
Genotype 12	80.5	8	91.7	10	100	10	100	10	100	10	100	10	100	10
Genotype 13	78.6	8	92.9	10	92.9	10	100	10	100	10	100	10	100	10
Genotype 14	0	0	0	0	0	0	7.1	1	7.1	1	7.1	1	28.6	3

DAH- Day after herbicide spray; CI – Crop injury; PRS – Phyto-toxicity rating scale

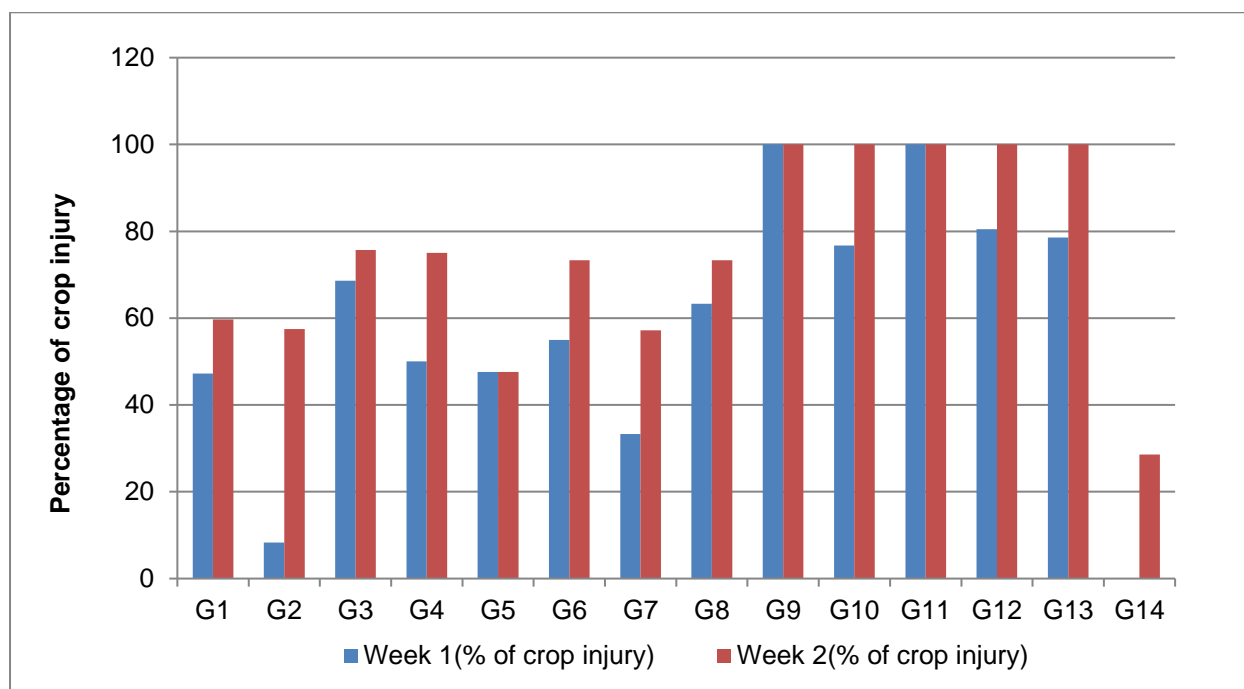


Fig. 1. Effect of Imazethapyr herbicide application on rice genotypes over two weeks after spraying about crop injury

Direct-seeded rice cultivation faces the challenge of effective weed management, and broad-spectrum non-selective herbicides are often used to control weeds. To address this issue, researchers developed an EMS-induced rice mutant named 'HTMN22' (HTM), which exhibits tolerance to the herbicide Imazethapyr (Shoba et al., 2017). Imidazolinone-tolerant rice cultivars show varying levels of tolerance to imazethapyr herbicide. Application of imazethapyr + imazapic, applied both pre- and post-emergence, effectively controlled red rice while reducing phyto-toxicity in tolerant varieties (Villa et al., 2006). Even herbicide-tolerant cultivars can be prone to herbicide drift as observed with IRGA 417, which showed increased sensitivity to simulated drift of imazethapyr and imazapic (Dal Magro et al., 2006). Additionally, tolerant cultivars may still display phyto-toxicity symptoms, such as leaf discoloration and stunted growth, especially when exposed to higher application rates, with these effects persisting for one year (Marchesan et al., 2011). Among the tested varieties, HTM-N 22 and N 22 were the only ones to survive the early post-emergence imazethapyr application at 75 g ai/ha 30 DAS, with minimal plant drying due to their herbicide tolerance (Senthil Kumar et al., 2022). Imazethapyr has been shown to effectively control weedy rice and barnyardgrass in

imidazolinone-tolerant rice, without affecting grain yield (Wright et al., 2020).

4. CONCLUSION

This experiment conducted a phytotoxicity study with 14 rice genotypes in pot culture, using imazethapyr herbicide at a rate of 2.5 ml per liter during the 2-3 leaf stage of weeds, the results indicated that while some genotypes initially show resistance to toxicity, prolonged exposure typically leads to increased susceptibility and greater crop damage. Genotypes G1, G2, G3, G4, G5, G6, G7, G8 and G14 showed the highest resilience, maintaining their tolerance with minimal damage, while genotypes G9, G10, G11, G12 and G13 exhibited susceptibility to the herbicide and it showed phyto-toxicity. Hence, the tolerant genotypes G1, G2, G3, G4, G5, G6, G7, G8 and G14 can be used for further rice breeding programs aimed at developing herbicide-tolerant rice varieties.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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