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RESEARCH ARTICLE

Performance of herbicide mixtures and sequential application for benghal dayflower management

Performance da mistura de herbicidas e aplicação sequencial no manejo de trapoeraba

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Abstract: **Background**: Benghal dayflower is a weed widely distributed in Brazil, known for its tolerance to glyphosate, requiring the use of alternative herbicides and more complex control strategies.
Objective: Evaluate different auxinic herbicides in combination with glyphosate for the control of Benghal dayflower and assess the efficacy of glufosinate alone or in combination with PPO inhibitors in sequential applications.
Methods: Two experiments were conducted in Ponta Grossa (PR-Brazil), using a randomized block design. The first experiment evaluated different auxinic herbicides combined with glyphosate, including 2,4-D, triclopyr, halauxifen, fluroxypyr, and dicamba. Additionally, treatments with glyphosate alone and a non-treated control were assessed. The second experiment evaluated Benghal dayflower control with glyphosate + 2,4-D followed by sequential glufosinate applications alone or with PPO inhibitors, including saflufenacil, tiafenacil, carfentrazone, and flumioxazin. In both experiments, visual weed control evaluation were performed by means of control percentage relative in comparison of untreated control.
Results: Glyphosate plus auxinic herbicides showed better control level compared to glyphosate alone, especially for 2,4-D + glyphosate mixture. The addition of PPO inhibitors did not improve Benghal dayflower control compared to glufosinate alone. However, sequential applications enhanced control, achieving an average of approximately 93% compared to only 75% in the non-sequential treatment.
Conclusions: The use of auxinic herbicides in mixture with glyphosate, mainly 2,4-D, in the burndown application and sequential treatment of glufosinate alone or combined with PPO inhibitors are strategies that enhance Benghal dayflower control.

Keywords: *Commelina benghalensis*, chemical control, glyphosate, glufosinate ammonium, PPO inhibitors, auxin mimics.

Resumo: **Introdução**: A trapoeraba é uma planta daninha amplamente difundida no Brasil, tolerante ao glifosato, exigindo o uso de herbicidas alternativos e estratégias de controle mais complexas.
Objetivo: Avaliar diferentes herbicidas auxínicos em mistura com glifosato para o controle de trapoeraba, assim como a eficácia de glufosinato isolado ou em combinação com inibidores da PROTOX em aplicações sequenciais.
Métodos: Dois experimentos foram conduzidos em Ponta Grossa (PR-Brasil), em delineamento de blocos casualizados. O primeiro avaliou diferentes herbicidas auxínicos em associação com glifosato, incluindo 2,4-D, triclopir, halauxifen, fluroxipir e dicamba. Além disso, foram avaliados tratamentos com glifosato isolado e testemunha. No segundo experimento, avaliou-se o controle de trapoeraba com glifosato + 2,4-D seguido de aplicações sequenciais de glufosinato isolado ou em mistura com os inibidores da PROTOX saflufenacil, tiafenacil, carfentrazona e flumioxazina. Em ambos, as avaliações visuais de porcentagem média de controle foram realizadas em comparação com a testemunha sem aplicação.
Resultados: Todos os tratamentos que combinaram glifosato com herbicidas auxínicos apresentaram performance superior em comparação ao glifosato isolado, com destaque para 2,4-D + glifosato. A adição de inibidores da PROTOX não melhorou o controle de trapoeraba em relação ao glufosinato isolado. No entanto, as aplicações sequenciais resultaram em aumento no controle, atingindo em média aproximadamente 93% contra apenas 75% no tratamento sem sequencial.
Conclusões: A utilização de herbicidas auxínicos na dessecação em mistura com glifosato, principalmente 2,4-D, e a aplicação sequencial de glufosinato isolado ou em associação com inibidores da PROTOX são estratégias que melhoram o controle de trapoeraba.

Palavras-chave: *Commelina benghalensis*, controle químico, glifosato, glufosinato de amônio, inibidores da PROTOX, mimetizadores de auxina.

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1. Introduction

The agricultural area designated for crop production in the 2024/2025 growing season in Brazil is projected to reach 81.6 million hectares, with an estimated grain production of 328.3 million tons (CONAB, 2025). In this context, one of the greatest challenges to agricultural production is competition with weed species, with a reduction of the yield grain greater than 80% when no control measures are implemented (Hussain et al., 2021). Among the five most problematic weed species in the Brazilian agricultural fields are *Conyza* spp., sourgrass (*Digitaria insularis*), morningglory (*Ipomoea* spp.), goosegrass (*Eleusine indica*), and Benghal dayflower (*Commelina benghalensis* L.) (Oliveira et al., 2021).

Benghal dayflower, also known as tropical spiderwort, is the most significant weed species in the Commelinaceae family, negatively affecting crops such as soybean and corn around the world due to yield losses and interference with harvesting processes (Wilson, 1981; Sabila et al., 2012; Bottcher et al., 2022; Moura et al., 2024). For favorable conditions, such as humid, warm, and shaded environments with predominantly clayey soils, this weed has become increasingly prevalent in crop fields with continuous spread and, exhibiting characteristics of perennialization (Steptoe et al., 2006).

For this weed species, mechanical control is not recommended due to its vegetative propagation. Consequently, operations such as soil disturbance or hand weeding may contribute to the dispersal of structures that give rise to new plants (Wilson, 1981). On the other hand, chemical control of Benghal dayflower is challenging due to its tolerance to the herbicide glyphosate, which is widely used in non-selective weed management across Brazilian croplands (Bottcher et al., 2022). For these reasons, the control of Benghal dayflower is complex, and there is a continuous need to explore new strategies for chemical control (Ronchi et al., 2002).

Most herbicides, when applied alone without sequential applications, are ineffective in controlling Benghal dayflower during its adult stage. Historically, glyphosate herbicide applications have been exhibiting several failure control, especially in plants with greater development and has been increasing its occurrence in cropping systems after glyphosate-resistant crops introduction. Furthermore, continuous application of the same herbicide (or herbicides with the same mode of action) poses a significant challenge due to the high selection pressure exerted on the species, increasing the risk of resistant biotype development (Vargas et al., 1999; Ronchi et al., 2002).

The use of herbicide mixtures and sequential applications has shown potential as an alternative for controlling Benghal dayflower. For instance, mixtures containing auxinic herbicides, such as 2,4-D, triclopyr, fluroxypyr, halauxifen, and dicamba, have been investigated. For sequential applications, glufosinate-ammonium, a glutamine synthetase (GS) inhibitor, can be used alone or in combination with protoporphyrinogen oxidase (PPO) inhibitors, such as carfentrazone, saflufenacil, flumioxazin, and tiafenacil (Oliveira Jr et al., 2021).

Based on this approach, the present study hypothesized that use of auxin mimics in combination with glyphosate significantly improves Benghal dayflower control during burndown applications, with differences in the level efficacy among herbicides within this group. Additionally, sequential application with glufosinate-ammonium is essential to achieving satisfactory control levels, especially when applied together with PPO inhibitors to enhance its effectiveness. Therefore, this research aimed to evaluate the efficacy of different *C. benghalensis* control strategies, including the use of auxinic herbicides mixed with glyphosate and, compared to glyphosate alone, followed to the sequential application of glufosinate-ammonium alone or in combination with PPO inhibitors.

2. Material and Methods

Experimental Site and Environmental Conditions

The experiment was carried out in the field at the Capão da Onça School Farm (FESCON, Ponta Grossa, Paraná, Brazil; 25°05' 33' S, 50°02' 53" W). According to the Köppen (1936), the region's climate is classified as Cfb (subtropical), characterized by mild temperatures and well-distributed rainfall throughout the year. The average temperature of the site is 18.3°C, with an annual average precipitation of 1505 mm (Climate Data, 2024).

Experimental Design and Treatments

Two field experiments were conducted to assess the post-emergence control of Benghal dayflower. Experiment 1 evaluated the efficacy of different auxinic herbicides in combination with glyphosate (Table 1). Experiment 2 examined the control effectiveness of the best-performing treatment from Experiment 1 when applied for burndown, followed by a sequential application of glufosinate-ammonium, either alone or in combination with PPO-inhibiting herbicides.

Table 1. List of treatments in Experiment 1, including their respective active ingredients and application rates. All treatments received the addition of the non-ionic adjuvant Assisti® at a concentration of 0.5%. Ponta Grossa, PR, 2024.

N°	Treatments	Application rate (g ai ha^{-1})
1	Control	-
2	Glyphosate[1]	1240
3	Glyphosate + 2,4-D[2]	1240 + 917.2
4	Glyphosate + Triclopyr[3]	1240 + 816
5	Glyphosate + (Halauxifen-methyl + Diclosulam)[4]	1240 + 6.3 + 31.9
6	Glyphosate + (Fluroxypyr + Clethodim)[5]	1240 + 345.6 + 168
7	Glyphosate + Dicamba[6]	1240 + 576

[1]Zapp QI 620®; [2] DMA 806 BR®;[3] Triclon®; [4] Paxeo®; [5] Araddo®; [6] XtenDicam®.

The herbicide applications for the first experiment were carried out on December 15, 2023. In Experiment 2, applications were performed on March 20, 2024, in which glyphosate + 2,4-D (Treatment T2) was used as the standard management and applied for all treatments in the first application, except to untreated control. Ten days later, on March 30, 2024, a sequential application of glufosinate-based treatments and PPO inhibitors was performed. At the time of application in both experiments, Commelina benghalensis plants were at an advanced stage of development, already in the reproductive phase. All applications were performed as part of a pre-plant burndown strategy.

The PPO inhibitors tested in Experiment 2 included saflufenacil, carfentrazone, flumioxazin, and tiafenacil (as shown in Table 2). Both experiments followed a randomized block design (RBD) with four replications. Each experimental plot measured 3 m in width and 5 m in length, covering a total area of 15 m².

Table 2. List of treatments in Experiment 2, including their respective active ingredients and application rates. All treatments received the addition of the non-ionic adjuvant Assisti® at a concentration of 0.5%. Ponta Grossa, PR, 2024.

N°	Treatments	Application rate (g ai ha^{-1})
1	Control	-
2	Glyphosate + 2,4-D (T2)	1240 + 917.2
3	T2 → (Glufosinate[1])*	400
4	T2 → (Glufosinate + Saflufenacil[2])*	400 + 35
5	T2 → (Glufosinate + Carfentrazone[3])*	400 + 30
6	T2 → (Glufosinate + Flumioxazin[4])*	400 + 25
7	T2 → (Glufosinate + Tiafenacil[5])*	400 + 118.65

[1]Finale®; [2] Heat®; [3] Aurora 400 EC®; [4] Flumyzin 500 SC®; [5] Terrad´Or 339 SC®; * Sequential applications with a 10-day interval.

The treatments were applied using a CO_2-pressurized backpack sprayer, equipped with AXI 110.02 (yellow) spray nozzles, operating at 30 psi, application speed of 1 m s^{-1}, resulting in a spray volume of 150 L ha^{-1}. The applications were carried out under favorable environmental conditions. In Experiment 1, the application day recorded an average temperature of 25.67°C, relative humidity of 60.44%, and an average wind speed of 6.21 km h^{-1}. In Experiment 2, during the first application, the recorded conditions were 23.32°C, 84.61% relative humidity, and a wind speed of 4.6 km h^{-1}, while for the second application, the temperature was 21.18°C, the relative humidity was 75.57%, and the wind speed was 7.06 km h^{-1}. The sequential treatment in Experiment 2 was conducted during a period of high light intensity, as required for optimal performance of the glufosinate-ammonium herbicide and PPO inhibitors (Takano and Dayan, 2020).

Data Collection and Statistical Analysis

In Experiment 1, evaluations were performed at 13, 20, 27, and 33 days after application (DAA). The assessments were based on visual ratings assigned to the plots, as shown in Table 3, where control efficiency was evaluated by comparing the treated plots with the untreated controls in each block.

Table 3. Visual rating scale for *Commelina benghalensis* control (adapted from Frans and Crowley, 1986). Ponta Grossa, PR, 2024.

Percentage	Main Category Description	Detailed Control Description
0	No effect	No control
10		Very poor control
20	Mild effect	Poor control
30		Poor to inadequate control
40		Inadequate control
50	Moderate effect	Inadequate to moderate control
60		Moderate control
70		Slightly below satisfactory control
80	Severe effect	Satisfactory to good control
90		Very good to excellent control
100	Total effect	Complete destruction

In Experiment 2, evaluations were conducted at 7, 10, 15, 21, 28, and 36 DAA, using the same methodology as Experiment 1. The evaluations at 7 and 10 DAA were performed before the sequential applications, Leading to uniform ratings for all treatments, except for the untreated control. The evaluation at 15 DAA was carried out 5 days after the sequential application.

Data were analyzed for normality using the Shapiro-Wilk test and for variance homogeneity using the Bartlett test, followed by analysis of variance (ANOVA) ($p<0.05$). Means were compared using the Student-Newman-Keuls (SNK) test at a 5% significance level. Data analyses and visualizations were conducted using RStudio (2024.09.0 version).

3. Results and Discussion

The results of Experiment 1 and Experiment 2 are showed below, focusing on different burndown strategies for the control of *C. benghalensis*. Data were analyzed based on visual control ratings, and statistical comparisons were made to evaluate the performance of the different treatments.

Experiment 1 - efficacy of auxinic herbicides in the control of *Commelina benghalensis*

In the first experiment, differences were observed at all evaluation periods when compared with the efficacy of glyphosate alone versus glyphosate mixed with auxin mimics for *C. benghalensis* control (Figure 1). At 13 DAA, the initial assessment indicated that the mixtures of glyphosate with 2,4-D, halauxifen, fluroxypyr, dicamba, and triclopyr resulted in the highest control levels of Benghal dayflower, with a control level higher than 67.5% for mixtures and lower than 41.2% for glyphosate applied alone.

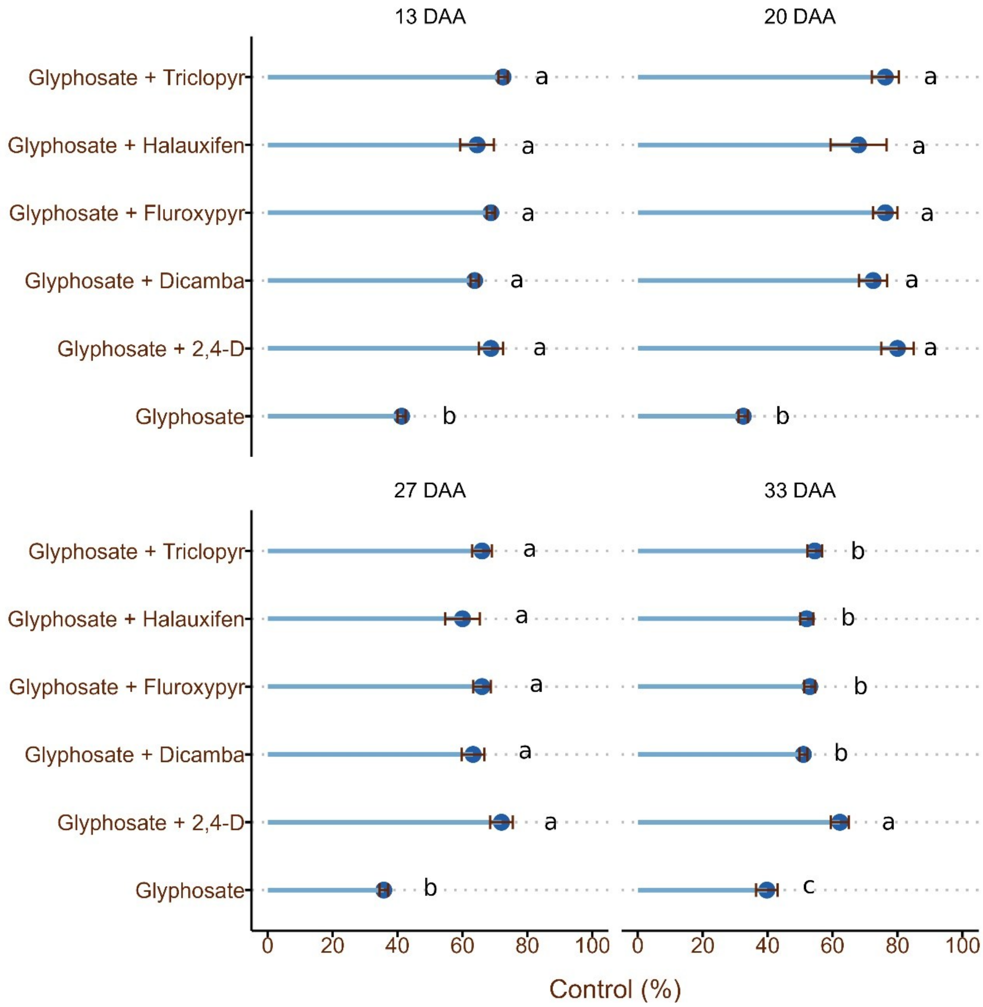

Figure 1. Control percentage data were assessed at 13, 20, 27, and 33 days after application (DAA) and are reported as the mean ± standard error of the mean. Means followed by the same letter do not differ statistically at a 5% probability level according to the SNK test.

At 20 DAA, all treatments containing glyphosate combined with auxinic herbicides exhibited high control efficacy, reaching an average of 74.6%. However, a slight decline was observed at 27 DAA, with control dropping to 65%. Despite this reduction, these combinations still outperformed glyphosate applied alone, whose effectiveness declined after 15 DAA, reaching only approximately 32%, thus highlighting the high recovery potential of mature *C. benghalensis* plants. This behavior aligns with findings from Dias et al. (2012), who also reported regrowth following glyphosate application in advanced developmental stages. Moreover, while Ronchi et al. (2002) observed a progressive increase in control up to 90% at 28 DAA with the glyphosate + 2,4-D combination, the current results suggest that although such combinations enhance initial control, their efficacy may vary depending on environmental conditions or weed phenology.

At 33 DAA, glyphosate + 2,4-D provided the highest control level, reaching approximately 62%, which was greater than other mixtures with auxinic herbicides. Additionally, glyphosate applied alone resulted controlled only 40% of the plants, performing lower than all other treatments.

Takano et al. (2013) reported similar findings, demonstrating that the combination of glyphosate and 2,4-D outperformed glyphosate alone. For plants at the 10-leaf stage, control with glyphosate alone was 6.20%, whereas the combination with 2,4-D resulted in 62.50% control. Still, in their study, only the combined application achieved acceptable control efficiency (>85%) for Benghal dayflower at the 4–6 leaf stage, emphasizing the effectiveness of this mixture for species that glyphosate alone struggles to control.

Osipe et al. (2017) evaluated 2,4-D and dicamba applied individually and in combination with glyphosate for the control of *C. benghalensis*. Their results indicated that 2,4-D alone was more effective than dicamba, and in adult plants (10–12 leaves), dicamba failed to achieve satisfactory control levels. When mixed with glyphosate, both treatments provided satisfactory control, although 2,4-D proved to be more suitable in the mixture. Similarly, Perissato et al. (2023) evaluated the control of *C. benghalensis* using glyphosate + dicamba and glyphosate + 2,4-D. Their results align with the findings of the present study, showing that the use of 2,4-D in adult plants resulted in superior performance compared to dicamba in this combination, achieving 90% control at 28 days after application (DAA).

Despite these findings, in the present study, even the most effective treatment (glyphosate + 2,4-D) failed to achieve satisfactory control levels (<80%), evidencing the necessity of sequential applications. Consequently, Experiment 2 assessed sequential applications following the glyphosate + 2,4-D treatment.

Experiment 2 - efficacy of glufosinate-ammonium alone or in combination with PPO inhibitors in the sequential application for Benghal dayflower control

In the second experiment, evaluations conducted at 7 and 10 DAA (before the sequential applications) showed average control levels of 45% and 55%, respectively, resulting solely from the initial application of the glyphosate + 2,4-D mixture (data not shown). Subsequently, at 15 DAA, all sequential applications enhanced control efficacy, with no differences among treatments, achieving approximately 80% control of *C. benghalensis* (Figure 2). In contrast, the non-sequential treatment exhibited significantly lower control, reaching only 57.5%.

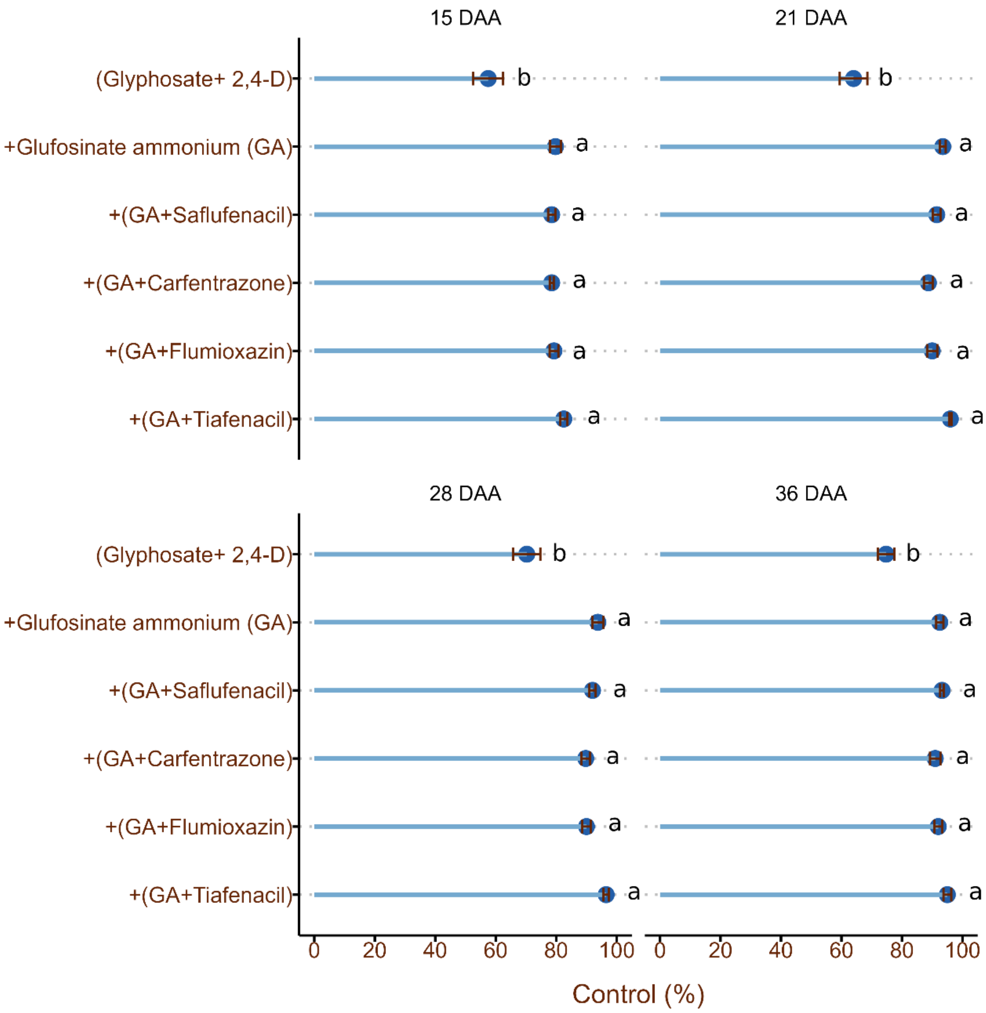

Figure 2. Control percentage data were assessed at 15, 21, 28, and 36 days after application (DAA) and are reported as the mean ± standard error of the mean. Means followed by the same letter do not differ statistically at a 5% probability level according to the SNK test.

At 21, 28, and 33 DAA (Figure 2), a progressive improvement in *C. benghalensis* control was observed across all treatments, reaching satisfactory levels, except for the glyphosate + 2,4-D treatment without a sequential application, which achieved a final control rate of 75.5%. All treatments involving sequential applications exhibited excellent control efficacy, with results exceeding 92%. However, no differences were detected between glufosinate-ammonium applied alone and its combinations with PPO inhibitors.

Correia et al. (2008) evaluated the control of *C. benghalensis* in RR soybean, concluding that sequential glyphosate applications at doses of 0.96 kg a.e. ha^{-1} followed by 0.72 kg a.e. ha^{-1} were necessary for effective control of this weed. However, the authors also observed that *C. benghalensis* exhibited tolerance to glyphosate, indicating the need for additional management strategies in transgenic soybean fields. They highlighted that the developmental stage of the plants at the time of herbicide application is a critical factor for effective control. When working with plants from seeds at stages 4 to 6 fully expanded leaves, satisfactory control was not achieved. This partially explains the low control observed in the present study, as the plants were already mature and perennial, making the glyphosate treatment insufficient to reach the desired control level.

In addition, glyphosate tolerance and plant size can reduce the control level, indicating the essential role of sequential application with contact herbicides, such as glufosinate-ammonium, for controlling *C. benghalensis* due to the need to target regrowth points in desiccated plants, preventing their survival. This finding aligns with the observations of Daramola et al. (2024) and corroborates those reported by Bottcher et al. (2022) and Albrecht et al. (2023).

Although studies, such as that of Takano et al. (2020) with *Amaranthus palmeri*, have shown improved efficacy of glufosinate-ammonium when combined with PPO inhibitors, the present study found no increase in control efficacy when PPO inhibitors were added to glufosinate for *C. benghalensis* management. Nevertheless, herbicide mixtures remain a recommended technique in integrated weed management, as they play a crucial role in preventing the development of herbicide-resistant weeds (Markus et al., 2021).

Thus, sequential applications of glufosinate-ammonium, whether used alone or in combination with PPO inhibitors, proved superior in controlling *C. benghalensis* compared to the non-sequential glyphosate + 2,4-D treatment in all evaluations. However, it is important to emphasize that the adoption of other control methods, including crop rotation, cover crops, and the use of pre-emergence herbicides, is essential for reducing infestation and promoting effective management of Benghal dayflower in agricultural systems.

4. Conclusions

The mixture of auxin mimicking herbicides with glyphosate improves the control of *C. benghalensis* compared to isolated glyphosate applications. There are differences in the efficacy of control between the various auxin mimics mixed with glyphosate, with the 2,4-D + glyphosate mixture showing the highest efficiency in controlling Benghal dayflower. The use of sequential application with glufosinate, either alone or in combination with PPO inhibitors, is necessary and effective for post-emergence control of this species.

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