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POST-EMERGENCE CONTROL OF *Eriochloa villosa* (Thunb.) Kunth IN SILAGE CORN

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Abstract

Weed interference in agricultural fields poses an important challenge globally due to direct competition with vulnerable crops such as corn. Our study is focused on the chemical control of Eriochloa villosa, commonly known as the woolly cupgrass, an emerging invasive Poaceae species in Romania that germinates in waves and was reported to significantly reduce corn yield. We conducted a post-emergence experimental trial in which six herbicidal formulations were tested against E. villosa in a field located near Buzas, Timis County, Romania. In this practical screening, three visual assessments were carried out and data statistically analysed. Our findings revealed that treatments, assessments dates and their interactions were significant, and treatments based on Tembotrione 44 g/l and Mesotrione 75 g/l + Nicosulfuron 30 g/l provided the highest efficacy, with over 90% control 24 days after the application, while other treatments provided only moderate control. The use of these post-emergence active ingredients is immediately applicable in agricultural field management strategies by farmers and other specialists, and the results establish an important foundation for future research of E. villosa.

Key words*: weed suppression, chemical control, phytotoxicity, post-emergence, Poaceae.*

INTRODUCTION

Segetal weed species have long negatively impacted agricultural crops, though numerous control techniques were used to cope with their pressure. While chemical control existed in rudimentary form since antiquity in the form of salts, plant extracts, mineral and organic mixes, the creation of synthetic herbicides revolutionized agriculture (Horvath et al., 2023). Despite their popularity and efficacy, extensive and irresponsible herbicide usage led to severe ecological impact and health issues in humans as well as animals, and often to the development of herbicide resistance in numerous weed species (Heap, 2014; Hongoeb et al., 2025; Pogurschi et al., 2024; Pratt et al., 1997). In response to legislative measures taken against harmful substances to human health and the environment, numerous herbicides were banned, which led to the need to optimize management strategies using newer alternatives (Mallory-Smith & Roma-Burgos, 2020).

Corn, *Zea mays* L., is one of the most relevant crop species due to its high adaptability, genetic variability and high yield potential, with a yearly production of over 1 billion metric tons globally (García-Lara & Serna-Saldivar, 2019). Despite these traits, corn is still very susceptible to the presence of weed species in early developmental stages due to high weed density, allelopathy and resource competition (Casimero et al., 2022; Horvath et al., 2023; Meissle et al., 2010; Soltani et al., 2016; Vila-Aiub et al., 2008). Due to these factors, losses, especially during early stages of crop development, are significant and consistent, leading to serious economic strain (Cerrudo et al., 2012).

Weed biodiversity within crop fields may vary greatly depending on location, season and management strategies, although recently, a loss in species richness was noted in Europe, but not in abundance (Petit et al., 2016). *Eriochloa villosa* (Thunb.) Kunth (woolly cupgrass) is an invasive species whose populations germinate in waves throughout the season and are often abundantly found in infested corn fields, which can lead to significant competitive pressure and yield loss of over 80% (Darbyshire et al., 2003; Lele et al., 2025). In our experimental field trial, this species had the highest and most homogenous presence in the plots, occurring in

all of them. *E. villosa* is the focus of our study as its response to newer herbicides is important to evaluate to better manage its fast spread.
Although *E. villosa* had the largest population in this particular trial, it was not the only species encountered and generally, herbicides with a broader control spectrum may still be preferred for fields that present both grassy and broadleaf weed species in order to properly manage the wider variety, so our herbicide choice was based on this aspect (Kushwah, 2020; Susha et al., 2018).
Our objective was to assess the chemical efficacy of 7 herbicide treatment variants against the most abundant species encountered in the trial field, *E. villosa*. We hypothesize that herbicide efficacy against *E. villosa* would significantly differ among treatments, but that they will achieve higher efficacy than the untreated control. We also hypothesize that efficacy will increase after each evaluation, with the highest control by the last assessment, although efficacy increase rate will depend on the herbicide, with some showing faster improvement than others. As the herbicide used in T5 clearly labels high efficacy against *E. villosa*, we expect it to achieve great control. No phytotoxicity is expected in corn, as all chosen herbicides are within recommended rates.

MATERIALS AND METHODS

The experimental trial was located on a field near Buziaș town, in Timiș County, Romania, in open field conditions. The location was chosen for its homogenous *E. villosa* populations, ensuring representative results. Environmental and weather data on temperature, precipitation and humidity were acquired from DTN LLC, collected from May 16th 2024 to June 8th 2024 for this area (Figure 1). Soil data was collected using a standard soil sampler tube at 0.5 m below the surface and texture analyses performed at OSPA Timiș laboratories.
During May, temperatures averaged 18.5°C, with the minimum temperatures averaging at 12.7°C and maximum temperatures averaging at 24.5°C. Rainfall averaged 2.00 mm per day with a total of 31.2 mm during the period of the trial.

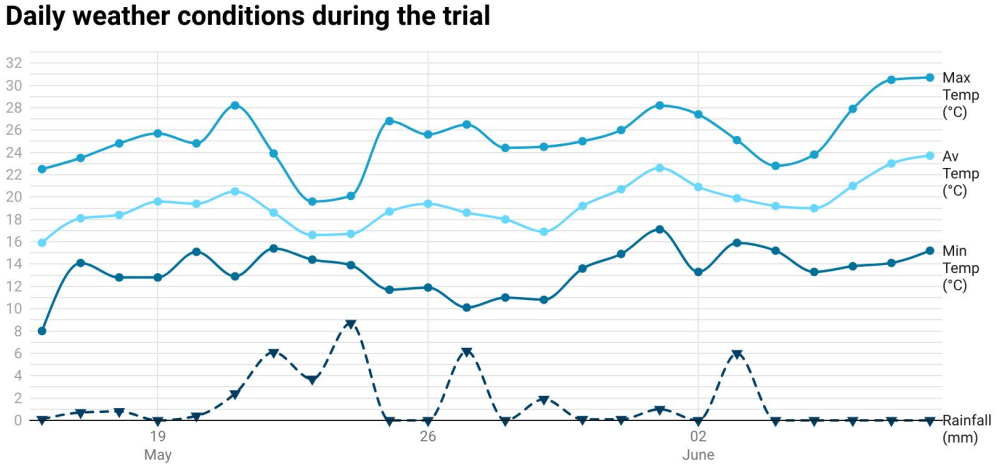

Figure 1. Visual representation of minimum daily temperatures, average daily temperatures and maximum daily temperatures throughout the trial, represented by continuous lines with one dot along for each day. Daily rainfall is represented by a dashed line, with triangles along the line representing each day. Graph created with datawrapper.de

During June, temperatures averaged at 21.2°C, with minimum temperatures averaging at 14.7°C and maximum temperatures averaging at 27.1°C. Rainfall averaged at 0.87 mm per day, with a total of 7.00 mm during the trial period. The field was not irrigated.

Pedological laboratory analyses of particle sizes presented the following soil texture properties: 13.5% coarse sand, 28% fine sand, 30.3% silt, 28.2% colloidal clay and 47.6 physical clay. According to these analyses, the soil is classified as clay. The pH was 6.51 with a percentage of 9.5% organic matter.

Corn variety Stine 9734-G was sown on May 2nd and presented, on average, 3 true leaves at the date of application. Crop yield analyses were not carried out as only the efficacy of the treatments *E. villosa* was included in the final analysis, and the results would not reflect the reality of the field due to the presence, albeit heterogeneous, of other weed species.

The weed developmental stage at the application timing did not exceed 4 true leaves for most individuals, and only a fraction presented grown tillers. Although multiple weed species were present in this field, we chose to focus on *E. villosa* only due to the heterogeneous nature of the other weed populations, which disallowed representative or reliable statistical analyses.

The experimental trial design is a randomized complete block design (RCBD), with 7 treatments including the control (T1) and 6 replicates. One-meter paths between each row of replicates were kept for easy access to each plot. Plots were sized at 5 x 3 m (15 m^2) each. Treatments consisted of 6 commercially formulated herbicides and one adjuvant for T6 only as per product indications. Each treatment was applied once, as post-emergence.

The complete list of treatments, along with formulations and dosages, is represented in Table 1.

Table 1. Complete list of treatments used during this trial

Trt. no.	Active Ingredient(s)	Rate of application
1	-	-
2	S metolachlor 960 g/l	1.5 l/ha
3	Nicosulfuron 40 g/l	0.8 l/ha
4	Dicamba 312,5 g/kg + Mesotrione 150 g/kg + Nicosulfuron 100 g/kg	0.6 kg/ha
5	Tembotrione 44 g/l	2.25 l/ha
6	Mesotrione 75 g/l + Nicosulfuron 30 g/l	2 l/ha
7	Tembotrione 345 g/l + Thiencarbazone 68 g/l	0.29 l/ha
	Adjuvant (esterified rapeseed oil)	2 l/ha

They are represented by the control (T1), one pre-emergence type treatment (T2) and 5 post-emergence treatments (T3-T7). All were applied as post-emergence, in one timing. All except T3 were applied at the maximum allowed rate.

Nicosulfuron was chosen as it was often used against *E. villosa* in experimental trials in the past (Arnold et al., 2005; Lele et al., 2025). The lower than allowed dosage was used as the maximum dose is intended for applications of fields where *Sorghum halepense* L. Pers. is present, which was not the case. Newer active ingredients such as Tembotrione, Mesotrione and Thiencarbazone were chosen for this trial because they contain modern active ingredients, all registered after the year 2000, as well as because no research papers reported the response of *E. villosa* populations in a controlled trial against any of them. All treatments contained post-emergence formulations, with the exception of treatment 2 (containing S-metolachlor) which, although a pre-emergent herbicide, it is often used in combination mixes by farmers and was included in this post-emergence application to observe any possible effect on the crop and on already emerged weed species under field conditions. Several months after the final assessment, this active ingredient was banned from use in the European Union based on regulation (EU) 2024/20, though we will still present the results as other countries may still use formulations using this active ingredient (European Commission, 2023).

The application took place on May 16th 2024. It started at 7:12 PM and ended at 7:46 PM. Air temperature during the application averaged 20.8°C while soil temperatures averaged 23.8°C. Humidity averaged 40.3%. Wind speeds averaged 3.53 meters per second (minimum 1.4 and maximum 6.6). The application was completed using an experimental grade backpack sprayer with compressed air at 2.6 bars, a spraying band 3 m wide with 6 flat fan type nozzles spaced 50 cm between each other and the application performed at 50 cm above the ground to reduce drift.

The chemical control against *E. villosa* was visually assessed taking into consideration symptoms of chlorosis, bleaching, necrosis and stunted growth or deformation. We used percentages from 0 (no effect) to 100 (all dead) in increments of 10 for each plot during every assessment date. Crop phytotoxicity was visually assessed according to these guidelines as well. The assessments were carried out during three different dates, at 9 (May 25th), 18 (June 2nd) and 24 (June 8th) days after the first application.

RStudio version 2026.01.0 (Build 392) was used for data analysis and graph creation within this experimental trial. Several packages were used to facilitate handling of data, statistical analyses

as well as table and figure creation. The packages “dplyr”, “tidyr” and “tibble” were used to handle data and organize the code; “car”, “emmeans” and “multcomp” and “multcompview” were used for extended statistical analyses that were not available in base RStudio; while “ggplot2” and “gt” were used in the creation of publication grade visual representations (Fox & Weisberg, 2019; Graves et al., 2026; Hothorn et al., 2008; Iannone et al., 2026; R. Lenth & Piaskowski, 2025; Müller & Wickham, 2026; Wickham, 2016; Wickham, François, et al., 2026; Wickham, Vaughan, et al., 2026).

Treatments, assessment dates and replicates were taken into consideration as factors. Descriptive statistics analyses were performed to gain a better understanding of the data and factor interaction. Following the descriptive statistics analyses, normality was evaluated using the Shapiro-Wilk test on the response values and model residuals, as well as a visual evaluation using Q-Q plots (Shapiro & Wilk, 1965). Homogeneity of variances among treatments was determined using Levene’s test. These analyses were chosen due to their suitability for small to medium sample sizes (Yang & Berdine, 2021).

A Type II blocked analysis of variance (ANOVA) based on an ordinary least squares (OLS) linear model was applied, with assessment date (DAA) and treatment as categorical factors, replicate as a fixed block, and their two-way interactions included (Efficacy ~ Replicate + DAA (Days after application) + Treatment + DAA:Treatment)); this model was chosen as it is recommended for unbalanced factorial designs (Langsrud, 2003; Shaw & Mitchell-Olds, 1993).

Based on the ANOVA results, post-hoc comparisons were performed using estimated marginal means (EMMs) with Šidák adjusted pairwise comparisons. Compact letter groupings were created at $\alpha = 0.05$ and confidence intervals reported at 95% adjusted confidence level (R. V. Lenth, 2016; Sidak, 1967).

Effect sizes were analysed to determine which factors or interactions were the strongest drivers of variation in efficacy. Additionally, comparisons of treatments to control were carried out for a clear statistical view of the most efficient treatments.

RESULTS AND DISCUSSIONS

Results

In total, 10 species were found in the experimental field trial: *Hibiscus trionum* L. (31 plots), *Xanthium strumarium* L. (14 plots), *A. artemisiifolia* L. (9 plots), *Convolvulus arvensis* L. (6 plots), *Echinochloa crus-galli* (L.) P. Beauv. (5 plots), *Rubus caesius* L. (4 plots), *Cirsium arvense* (L.) Scop. (1 plot), *Polygonum aviculare* L and *Polygonum maculosa* Gray (both in 1 plot each). Due to their low representation and low frequency across and within plots, these species were excluded from the statistical analyses.The only species considered in this trial was *E. villosa*, and raw mean efficacy values were calculated for all treatments and visually represented in Table 2 and Figure 2.

Table 2. Visual representation of the raw mean efficacy of the treatments against *E. villosa*, with DAA = Days after application. The values are expressed as % efficacy

Mean treatment efficacy (%) against *Eriochloa villosa*							
Assessment day	Treatment						
	1	2	3	4	5	6	7
9 DAA	0	20	40	56.7	81.7	61.7	36.7
18 DAA	0	38.3	60	66.7	91.7	90	46.7
24 DAA	0	40.0	55	61.7	93.3	98.3	48.3

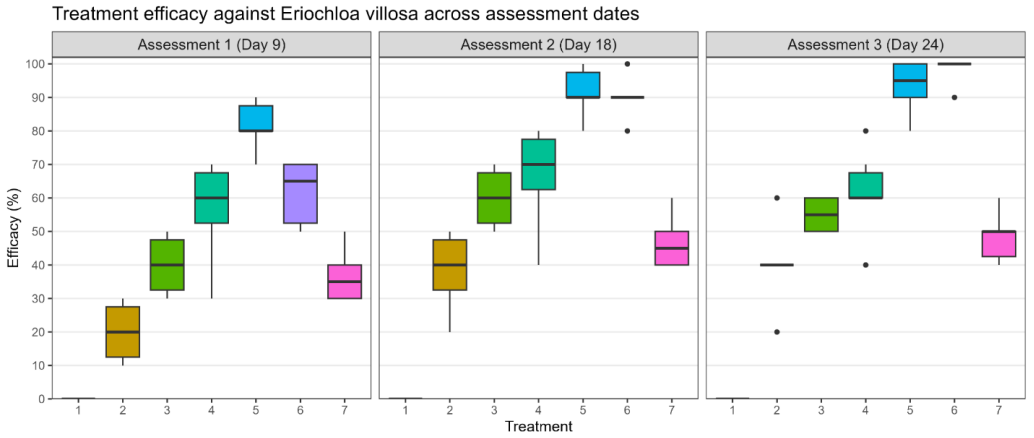

Figure 2. Boxplot representation of raw mean efficacies of all treatments against *E. villosa* across three assessment dates

Treatment 2, although a pre-emergence herbicide, was applied post-emergence in order to assess any potential effect upon already germinated and emerged weed seedlings. Its efficacy was low overall, reaching 38.3% by the second assessment and up to 40% by the third. Field observations indicated that established individuals were not visibly affected, and the control was associated with newly emerged seedlings, which presented clear signs of chlorosis. Treatment 3, a grass selective herbicide, reached up to 60% control against *E. villosa* at the second assessment, although efficacy declined by the third, settling on 55%. Treatment 4 performed similarly to T3, reaching a control of 66.7% at the second assessment and dipping in efficacy by the third, settling on 61.7%. Treatment 5 provided high control throughout the trial against *E. villosa*, with 81.7% by the first assessment, reaching 91.7% by the second assessment and 93.3% by the last. Treatment 6 provided excellent control by the final assessment, reaching 98.3%, although control was lower compared to T5 during the first two assessments. Treatment 7 provided moderate control, reaching 46.7% by the second assessment, though stagnating at 48.3% by the third and final assessment date. No signs of crop phytotoxicity symptoms were visible on corn throughout the trial, therefore the data, which was consistently 0 for every plot, is not presented.

Shapiro-Wilk normality tests were performed on both the raw mean efficacy values and the residuals of the final model. Raw mean efficacy values deviated significantly from normality ($W = 0.9388$, $p = 2.295 \times 10^{-5}$), while the residuals of the final model did not significantly differ from a normal distribution ($W = 0.9799$, $p = 0.05771$). Levene's test resulted in $F = 1.477$, $p = 0.105$, which was not significant at $\alpha = 0.05$, indicating that the homogeneity of variances was acceptable across groups.

Diagnostic plots supported the normality test results presented above.

The residuals were distributed around 0 without a clear pattern in the residuals vs fitted plot, while in the normal Q-Q plot, most residuals followed the reference line closely, only presenting small deviations at the tails, indicating that residual normality was broadly acceptable for the chosen model (Figure 3).

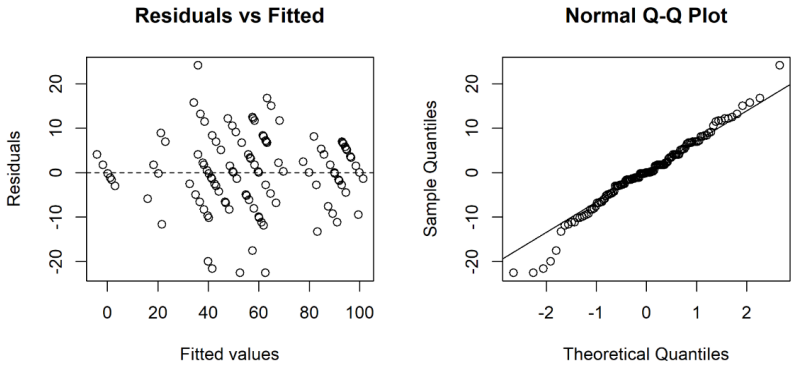

Figure 3. Residuals vs fitted and Normal Q-Q plot visual assessments of the model assumptions

A type II blocked OLS ANOVA model that included main effects as well as their two-way interaction was used to evaluate the efficacy of treatments (α=0.05). Assessment dates (DAA) and treatments had a significant effect on efficacy, while replicates did not. The DAA x Treatment was also statistically significant (Table 3).

Table 3. Blocked OLS ANOVA

Blocked OLS ANOVA (Type II) (*Efficacy (%) ~ Replicate + DAA + Treatment + DAA x Treatment*)					
Effect	Sum Sq.	Df	F value	Pr (>F)	Sig
Replicate	692.0635	5	1.7503	0.1301	*ns*
DAA	5530.1487	2	34.9659	0	***
Treatment	100349.2063	6	211.4947	0	***
DAA x Treatment	2936.5097	12	3.0945	0	***
Residuals	7907.9365	100	NA	NA	*ns*

Note: Significance levels (Sig): *** p < 0.001; ** p < 0.01; * p < 0.05; ns p ≥ 0.05.

We performed post-hoc comparisons based on the estimated marginal means (EMMs) with Šidák correction in order to further examine differences within each assessment date for *E. villosa*. The resulting compact letter display (CLD) showed considerable differences among treatments across all assessments. Treatments sharing at least one letter were not considered significantly different, while treatments assigned to completely different letter groups differed significantly (α=0.05). These results are visually represented in Figure 4.

At the first assessment date, 9 days after the application, the highest efficacy against *E. villosa* was recorded by Group E (T5), which was significantly different from all other treatments, followed by Group D (T4 and T6) and Group C (T3 and T7).

At the second assessment date, 18 days after the application, several treatments shifted statistical groups as efficacy increased over time. The highest efficacies were associated with T5 and T6 (Group E), which were significantly different from all other treatments, followed by T3 and T4 (Group D).

By the third and final assessment date, 24 days after the application, the treatment statistical groups shifted again, becoming more compact. Group D (T5 and T6) contained the most efficient treatments and differed significantly compared to all others. Group C (T3, T4 and T7) contained the following most efficient treatments, partially overlapping with Group B (T7, T3, T2).

Overall, the most effective treatments in this trial were T5 and T6, with a difference of over 30% between these and the following treatments, T3, T4 and T7, which showed moderate efficacy against this species. Images of the experimental plots can be found in figure 6, where each treatment is represented in two pictures, one overall, and one closer to the ground. The significant treatment effect as well as the significant Treatment:DAA interaction confirmed that herbicide efficacy greatly differed among treatments and changed over time.

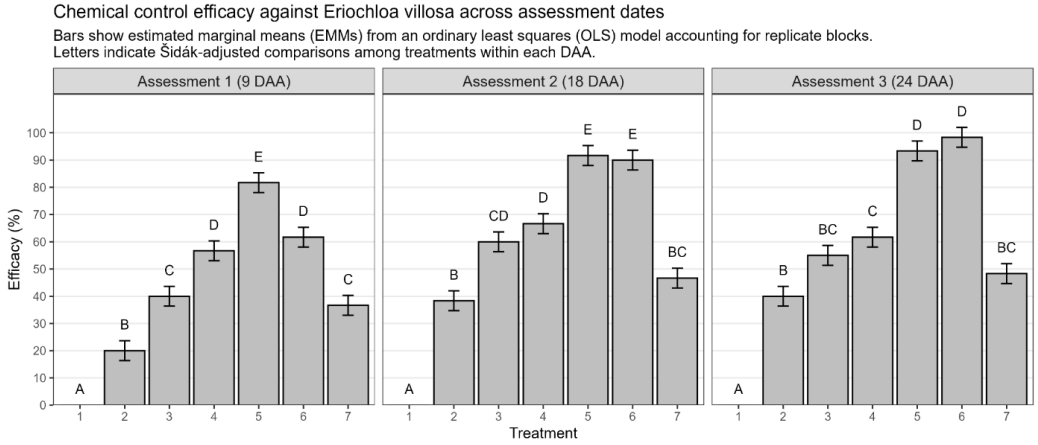

Figure 4. Estimated marginal mean efficacy of the treatments across assessments. Bars represent the predicted mean efficacy based on a type II blocked OLS ANOVA model and it accounts for the treatment, assessment date as well as their interaction ($\alpha = 0.05$). Treatments with the same letter within the same assessment are not significantly different

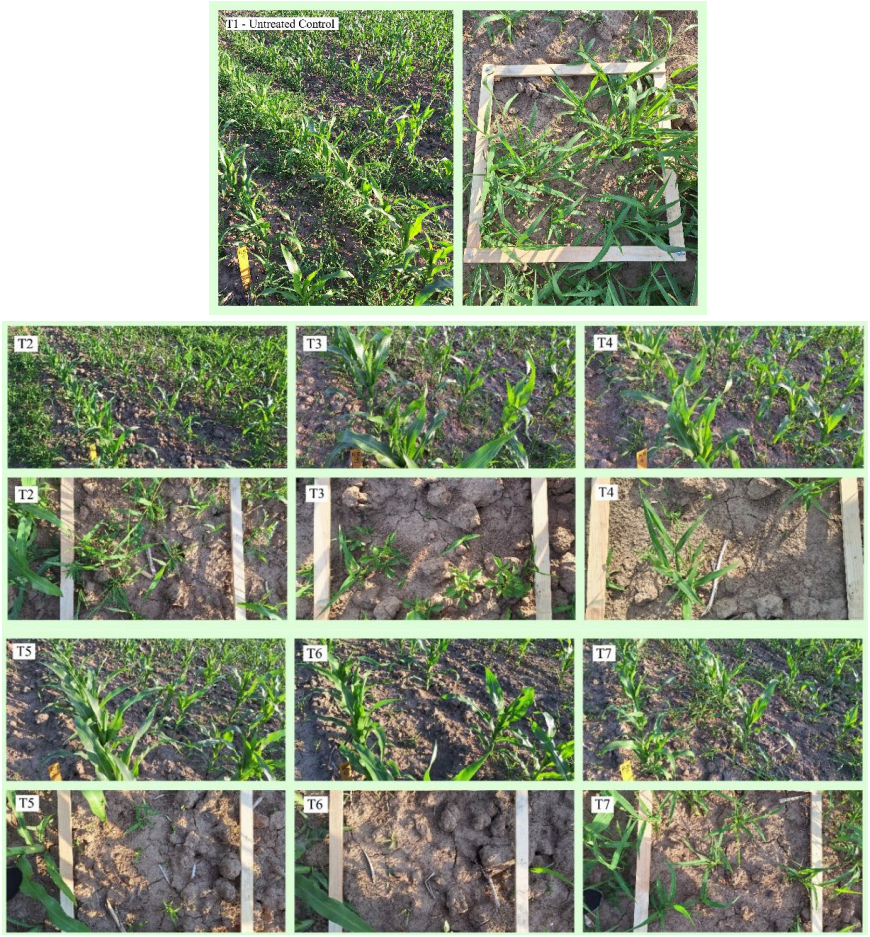

Figure 5. Representative plot images of all treatments at 24 DAA

Discussions

The response of *E. villosa* to 6 different herbicide formulations was studied in this experimental field trial, several of which were not officially tested against this species and reported in research papers before. Only reports of S-metolachlor and Nicosulfuron being used against this species have been published before, with varied results (Lele et al., 2025). Treatments 5 and 6 provided excellent control against *E. villosa*, while treatments 3, 4 and 7 only provided moderate control against it. As expected in our hypothesis, it showed a low response towards treatment 2 most likely because it was a pre-emergence herbicide applied as post-emergence. Our trial suggests only a limited number of these herbicides provided strong, reliable control of this species, leading to a direct relevant choice of treatments in infested corn fields.

Our hypotheses proved to be mostly correct, as all treatments were more effective than untreated control and their efficacy increased across assessment dates, although they slightly dropped for T3 and T4 after the second assessment. The efficacy of all treatments evolved differently throughout the trial, despite some of them reaching similar final results. This difference in efficacy is important from the perspective of prospective management choices because even though the final result is similar, early suppression of weeds reduces early competitive pressure for corn, when it is the most vulnerable (Horvath et al., 2023). Corn did not show any phytotoxic symptoms following the application, likely because all treatment doses were within approved safety thresholds.

As this was a field trial under natural, typical environmental conditions, several variables may have influenced the results. The weed population was abundant, as the field was selected specifically for its high and relatively homogeneous *E. villosa* infestation. Even so, the individual plants' growth stage did not exceed 4 true leaves and only a small proportion developed tillers by the time of application. The trial geographical location was that of a typical agricultural field subjected in a temperate continental climate, making it easy to replicate in other areas with similar climatic and pedological conditions, even in other countries or continents. Average temperatures during the trial were approximately 2°C higher than the historical mean from the past 30 years in this area, while rainfall in June was half of the historical amount for the corresponding study time period, which could have affected the response of *E. villosa* to the selected herbicides due to increased drought and heat stress. Regardless, these field variables reflect common types of variability under natural agricultural conditions.

E. villosa response to Nicosulfuron is known to vary depending on factors such as application timing, weather conditions, growth stage and possible genetic resistance to this active ingredient, as well as the number of applications, where it was often recommended as a double application. Considering the conditions of the experimental field, its design, the application timing and the population abundance, our results of treatment 3 are in line with previous studies (Lele et al., 2025). While T5 and T6 showed a very high efficacy, T7 only provided moderate control, even with the addition of the adjuvant. This contrast suggests that formulation differences and active ingredient combinations, species substance uptake and translocation can significantly influence their performance, even for herbicides that appear promising based on their composition.

Overall, our findings of previously tested herbicide formulations or active ingredients fit within existing knowledge on *E. villosa* control and add new, relevant information, especially for farmers in regions with similar environmental conditions, as they may contribute to the immediate improvement of management strategies against this problematic species. These results also present a starting point in choosing experimental designs for further chemical control studies.

Future studies of this difficult to control species may include multiple trial years, multiple dosages of the same active ingredient or formulation and multiple applications throughout the experimental period. In this type of research, other weed species are often included and studied if their populations are abundant and homogeneous enough for reliable statistical analyses. Advanced herbicide trials also include the study of crop yield quantity and content analyses for a better understanding of

the interaction between the crop and competing weeds. The biochemical response of *E. villosa* against the most efficient herbicides is a relevant aspect of understanding the mechanisms of herbicide efficacy and the pathways of active ingredient action within the metabolism of this species. Local genetic analysis may be required in the future, as weed tolerance becomes an increasingly important management aspect, with widely used active ingredients like Nicosulfuron becoming less effective as populations adapt to repeated use.

CONCLUSIONS

Weed control became an increasingly relevant challenge in modern agriculture, both because numerous widely used herbicides were banned and many weed species, like *E. villosa*, started exhibiting genetic tolerance to widely used alternatives in certain populations. Despite this, research focused on its management declined in the past two decades, which resulted in relatively few studies on modern herbicides with more sustainable formulations.

In this trial, we studied the efficacy of several modern herbicides against *E. villosa* and obtained very satisfactory results for two of them, treatments 5 and 6, which provided over 90% control in field conditions. Even though the other treatments only provided moderate control, the results nonetheless offer a useful insight into the most promising formulations for post-emergence applications. These results also help identify future research directions which may include testing lower doses, alternative mixtures of the same active ingredients, different or multiple application timings and most importantly, the interaction between these factors with the weed species and the infested crop.

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