

Control and translocation of saflufenacil in fleabane (*Conyza* spp.) according to plant integrity

Control y translocación de saflufenacil en yerba canicera (*Conyza* spp.) en función de la integridad de la planta

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ABSTRACT

Keywords:

Conyza spp
Herbicide mobility
Regrowth

Efficient herbicide absorption and translocation, and satisfactory weed control can be affected by the site of herbicide application. However, during harvesting of crops of previous soybean sowing, the cutting process made by harvesters on the fleabane may generate a difficult management in pre-sowing of the crop by limiting the leaf area of the absorption of the herbicide. Experiments were conducted to evaluate the control efficiency and translocation potential of saflufenacil in fleabane plants with different leaf and stem conditions. Experiment I was arranged in a 2x10 factorial scheme, with factor A corresponding to leaf integrity, and factor B corresponding to different levels of injury and saflufenacil application. Weed control was evaluated at 7, 14, and 21 days after herbicide treatment (DAT), and dry matter was evaluated at 21 DAT. Experiment II consisted of applying saflufenacil to different fleabane structures, where the percentage of necrotic area was evaluated at 1, 3, 5, and 7 DAT. Fleabane control was higher than 75% in all treatments with saflufenacil application, with greater control in plants previously defoliated. Saflufenacil application on 10 and 20 cm hairy fleabane plants was also efficient in all treatments. Saflufenacil application in old stem showed a larger necrotic area, while application in the site of the cutting resulted in a lower necrotic area. The main pathway for translocation of saflufenacil is via xylem and the stem proved to be the absorption element of the herbicide when leaf area is limited.

RESUMEN

Palabras clave:

Conyza spp
Movilidad herbicida
Rebrote

La eficiencia de absorción, translocación y control satisfactorio de malezas pueden ser afectados por el lugar de aplicación del herbicida. Sin embargo, durante la recolección de cultivos previos a la soja, el corte realizado por la cosechadora en la yerba carnífera dificulta el manejo de cultivos en pre-siembra al limitar el área foliar de absorción. Se realizaron experimentos para evaluar la eficiencia del control y el potencial de translocación de saflufenacil en plantas de yerba carnífera en diferentes condiciones. El primer experimento se organizó en esquema factorial 2x10. El factor A corresponde a la integridad de la hoja y el factor B corresponde a los diferentes niveles de daño y aplicación de saflufenacil. El control de *Conyza* se evaluó a los 7, 14 y 21 días después del tratamiento herbicida (DAT), y el peso seco a los 21 DAT. El experimento II consistió en aplicar saflufenacil a diferentes estructuras de *Conyza*, donde se evaluó el porcentaje de área necrótica a 1, 3, 5 y 7 DAT. El control de *Conyza* obtenido fue superior al 75% en todos tratamientos usando saflufenacil, con mayor control en plantas previamente defoliadas. La aplicación de saflufenacil en plantas de 10 y 20 cm fue eficaz en todos los tratamientos. Además, saflufenacil aplicado en el tallo viejo mostró un área necrótica mayor, mientras la aplicación en el sitio del corte generó como resultado un área menos necrótica. La translocación de saflufenacil se da a través del xilema y el tallo demostrando ser el elemento a través del cual se da la absorción del herbicida.

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Species of *Conyza* spp. (hairy fleabane) are one of the main weeds found in soybean (*Glycine max* (L.)) and corn (*Zea mays*) in Brazil (Kaspary *et al.*, 2016). The hairy fleabane has high rusticity, abundance in the seeds production and high potential regrowth; besides, the existence of biotypes resistant to the most used herbicides in the broad-leaves management (Osipe *et al.*, 2013). These features contribute to the increased occurrence of these weeds in agricultural areas, which hinder their control.

Methods for integrated management of resistant weeds must be continuously carried out in production areas. In this context, the winter management is highlighted, since allows the control of plants before they produce seeds and feed the soil seed bank (Constantin *et al.*, 2013). Autumnal management is a promising alternative in the context of limited postemergence herbicide options for *Conyza* spp. control, allowing its management in early stages or during regrowth (Dan *et al.*, 2013). However, during harvesting of certain crops, the cutting made by harvesters on the fleabane may generate a difficult management in pre-sowing of the crop by limiting the leaf area of the herbicide absorption.

The use of protoporphyrinogen oxidase-inhibiting herbicides (PROTOX) such as saflufenacil has stood out as an alternative for managing glyphosate-resistant fleabane biotypes (Mellendorf *et al.*, 2013). Saflufenacil belongs to the chemical group pyrimidinedione, and is recommended in preharvest desiccation, presowing burndown, and pre-emergence of dicotyledonous weeds (Grossmann *et al.*, 2011). Due to rapid action on plant tissues, these herbicides usually have low translocation potential, being predominantly translocated via xylem, except for saflufenacil, which presents some mobility via phloem (Ashigh and Hall, 2010; Oliveira Júnior, 2011).

Efficient herbicide absorption, translocation and satisfactory weed control can be affected by the site of product application in plant structures, requiring applications at the correct stages and good leaf coverage, mainly when referring to contact herbicides (Trezzi *et al.*, 2009). PROTOX-inhibiting herbicides can be absorbed by different plant structures, including roots, stems, and leaves. The contact place where the herbicide is applied can interfere the translocation and

efficiency of the herbicide. Moreover, plant age and turgor and leaf surface characteristics, can influence herbicide absorption and translocation (Trezzi *et al.*, 2009; Grossmann *et al.*, 2011).

Impaired height and leaf area in *Conyza* spp. plants that have been cut in the harvest of winter crops, can be a limiting factor for adequate coverage of saflufenacil spray, making the control a difficult target. Nevertheless, the ability of this herbicide to penetrate and damage plants when in contact with the remaining stems in the area has scarce studies. Thus, the present study aimed to evaluate the control efficiency and translocation potential of the herbicide saflufenacil when applied in fleabane plants with different leaf and stem conditions.

MATERIALS AND METHODS

The study was conducted from January to April 2018 in a greenhouse, which consisted of two experiments. Plants from an area with a record of glyphosate resistance in the city of Santa Maria, Rio Grande do Sul, Brazil, were used in both experiments. Experimental units consisted of 5 L plastic pots filled with Gray-Brown Argisol. Each experimental unit contained a single *Conyza* spp. plant collected in the field at a standard height of 20 cm and immediately was transplanted. Plants were irrigated daily until the average height of 25 cm, when treatments were established in the different experiments.

Experiment I aimed to evaluate the efficacy of the saflufenacil herbicide in *Conyza* spp. plants with different injuries, and a experimental design was carried out in completely randomized design, with four replicates. Treatments were arranged in a 2x10 factorial scheme, in which factor A represented leaf integrity and factor B consisted of different levels of injury and saflufenacil application, according to Table 1. For factor B, cutting process was made using a scissors, and the different periods were used to obtain plants without regrowth (cutting at 1 Day before application (DBA)) and with regrowth (cutting at 8 DBA), simulating situations of loss of *Conyza* spp. shoots during harvest.

The herbicide saflufenacil was applied at a rate of 35 g (active ingredient) a.i ha⁻¹, with addition of 0.5% mineral oil, at the time indicated in the Table 1. Treatments were applied with a CO₂ pressurized backpack sprayer

equipped with 110.015 nozzles and calibrated to provide an application volume of 150 L ha⁻¹. The variables analyzed were controlled at 7, 14, and 21 days after herbicide treatment (DAT), in a percentage scale where zero (0) represents the absence of

injuries, and one hundred (100) represents complete plant death. At 21 DAT, shoots were collected and placed in a greenhouse with forced air circulation at 65 °C for three days to determine the dry matter (DM) of fleabane plants.

Table 1. Treatments related to Experiment I arranged in a factorial scheme.

Factor A Leaf Integrity	With leaves Without leaves
Factor B With/Without cutting and Saflufenacil application	Whole plant
	Cutting 10 cm 8 DBA ¹
	Cutting 20 cm 8 DBA
	Cutting 10 cm 1 DBA
	Cutting 20 cm 1 DBA
	Whole plant + SAFL ²
	Cutting 10 cm 8 DBA + SAFL
	Cutting 20 cm 8 DBA + SAFL
	Cutting 10 cm 1 DBA + SAFL
	Cutting 20 cm 1 DBA + SAFL

¹DBA=Days before application of saflufenacil. ²SAFL=Saflufenacil.

Experiment II aimed to evaluate the translocation capacity of the herbicide saflufenacil when applied in different places in fleabane plants. It was conducted in a completely randomized design, with three replicates. Treatments consisted of the localized application of 35 g a.i. ha⁻¹ of the herbicide saflufenacil, with an addition of 0.5% mineral oil, in the following plant structures: leaf located close to the apex (new leaf); leaf located close to the base (old leaf); stem located close to the apex (new stem); stem located close to the base (old stem); and the region of the cut in 10 cm plants. The herbicide was deposited on plant structures using a flexible brush in a region of approximately 1.5 cm at the end of the day (7:00 PM). The variable analyzed was the percentage of necrotic area in fleabane plants at 1, 3, 5 and 7 DAT.

To measure the percentages of necrotic area, fleabane plants were photographed in all experimental units at each evaluation period. The software ImageJ® was used for image processing, where green plant parts were selected. Subsequently, non green parts were suppressed from the image, which was considered to be necrotic areas due

to the action of the herbicide (Figure 1). Afterward, the percentage of the necrotic area was calculated by the difference between the total plant area and the green area, that means, without necrosis symptoms from herbicide activity, according to Ramos *et al.* (2015).

The collected data were subjected to analysis of variance by the F test ($P < 0.05$). When statistically significant, the means were compared by the Tukey test ($P < 0.05$) using R software (R Core Team, 2020).

RESULTS AND DISCUSSION

Analysis of variance showed statistical significance for all variables analyzed in both experiments. In experiment I, fleabane control at 7 DAT was higher than 75% in all treatments with saflufenacil application, differing from the other treatments without applying of the herbicide (Table 2). Saflufenacil proved to be an efficient alternative in the control of glyphosate-resistant fleabane biotypes and acetolactate synthase (ALS)-inhibiting herbicides, and can be mixed with herbicides with other mechanisms of action (Dalazen *et al.*, 2015; Davis *et al.*, 2010).

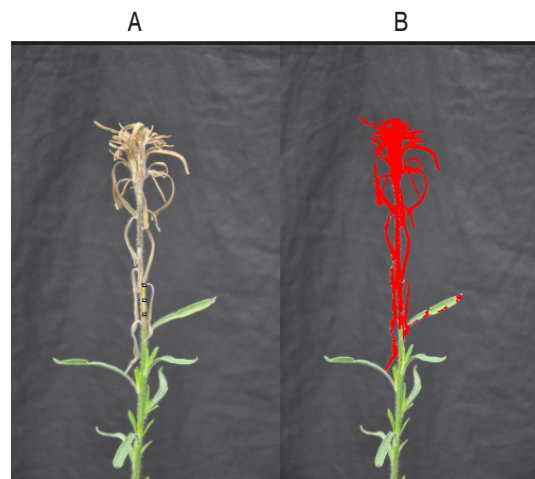


Figure 1. Identification and quantification of the percentage of the necrotic area of fleabane plants (*Conyza* spp.) after localized application of saflufenacil, indicating the original image without manipulation (A) and the selection of damage areas underlined in red color (B).

Comparing the leaf integrity of plants, the treatments in which plants were previously defoliated showed a higher control percentage at 7 DAT, except for the treatment with cutting+herbicide of 10 cm plants at 1 DBA (Table 2). Treatments with and without leaves, the most significant weed controls (higher than 90%) were observed when saflufenacil was added to treatments on defoliated plants. Moreover, approximately 20% higher control was achieved by applying the herbicide to whole plants (uncut) without leaves compared to treatment with leafy plants. This result is related to the deposition of saflufenacil on the stem of fleabane plants due to the absence of leaves. Thus, once absorbed in the stem, saflufenacil is translocated to the other parts of the plant, acting on the chlorophyll synthesis pathway (Oliveira Júnior, 2011). The green stem of fleabane plants stands out regarding the presence of this photosynthetic pigment, which can favor the mode of action of the herbicide.

The results showed that saflufenacil application on cut-leaf plants, simulating the harvest of winter cereals, was higher to the treatment of uncut-leafy plants at 7 DBA (Table 2). However, defoliated cut plants showed lower controls when compared with treatments mixed with herbicides. In this sense, fleabane plants have a high regrowth capacity after management practices with potential for recovery (Oliveira Neto *et al.*

et al., 2013). This regrowth capacity highlights the need to adopt alternatives that contribute to chemical and mechanical control through other practices such as crop management (Oliveira Neto *et al.*, 2010).

At 7 DAT, the treatment with the cutting of 10 cm plants at 1 DBA plus herbicide application did not differ between plants with and without leaves, showing satisfactory control (93%). This shows greater sensitivity of 10 cm fleabane plants for saflufenacil application (Table 2). Also, 20 cm plants that were cut and received saflufenacil did not differ from whole plants that received the herbicide, regardless of leaf integrity (Table 2). Growth and development characteristics must be observed for proper control of *Conyza* spp. through saflufenacil. Higher plants in an advanced growth stage have a high capacity for lateral shoot emission to recover from herbicide exposure (Moreira *et al.*, 2010). However, this study showed reasonable control of *Conyza* spp. plants up to 25 cm height.

After 14 and 21 DAT, the application saflufenacil in *Conyza* spp. plants of 10 and 20 cm was efficient in all treatments, with control equal to or greater than 98% (Table 2). Treatments without saflufenacil application generally showed a decrease in fleabane mechanical injuries from 14 DAT, differing from herbicide treatments and indicating plant recovery (Table 2).

Table 2. *Conyza* spp. control (%) in different tissue conditions at 7, 14, and 21 days after application (DAT) of saflufenacil.

Treatment	7 DAT		14 DAT		21 DAT	
	With leaves	Without leaves	With leaves	Without leaves	With leaves	Without leaves
Whole plant	0 eB	53 bA	0 dB	48 bA	0 cB	33 cA
Cutting 10 cm 8 DBA ¹	24 dB	64 bA	16 cB	56 bA	10 cB	41 bcA
Cutting 20 cm 8 DBA	16 dB	61 bA	8 cdB	49 bA	8 cB	35 cA
Cutting 10 cm 1 DBA	55 cB	65 bA	46 bA	48 bA	38 bA	48 bA
Cutting 20 cm 1 DBA	14 deB	62 bA	12 cB	50 bA	9 cB	47 bA
Whole plant + SAFL ²	76 bB	95 aA	95 aA	100 aA	98 aA	100 aA
Cutting 10 cm 8 DBA + SAFL	88 aB	97 aA	97 aA	100 aA	99 aA	100 aA
Cutting 20 cm 8 DBA + SAFL	83 abB	96 aA	93 aB	100 aA	98 aA	100 aA
Cutting 10 cm 1 DBA + SAFL	93 aA	93 aA	99 aA	100 aA	100 aA	100 aA
Cutting 20 cm 1 DBA + SAFL	85 abB	96 aA	99 aA	100 aA	100 aA	100 aA
C.V.(%) ³	8.11		7.50		9.59	

Means followed by the same uppercase letter in the row, comparing leaf integrity for each evaluation period, and by the same lowercase letter in the column, comparing injury levels for each treatment, indicate no significant difference by the Tukey test ($P < 0.05$). ¹DBA=Days before application of saflufenacil. ²SAFL=Saflufenacil. ³Coefficient of variation.

When associating saflufenacil and glyphosate, a similar study showed satisfactory control (97%) at 14 DAT of *Conyza bonariensis* plants with regrowth and glyphosate resistance in an apple orchard (Pereira *et al.*, 2016). Furthermore, mechanical weed control of *Eryngium horridum* with 10 to 15 cm mowing in natural pasture removed the shoots of weeds; however, it did not reach lateral buds, allowing regrowth at 20 DAT (Pellegrini *et al.*, 2007), a similar situation was observed in the present study.

Treatments with saflufenacil application decreased DM in comparison to those that have not applied herbicide in whole plants with or without leaves (Table 3). The DM of whole leafy plants decreased by 64% after saflufenacil application compared to an entire leafy plant without herbicide treatment. Cesco *et al.*, (2019) evaluated *Conyza* spp. control at different stages of plant development, the saflufenacil rate of 60 g a.i. ha⁻¹ decreased DM by approximately 77 and 54% in fleabane plants up to 5 and 20 cm, respectively, and led to a low regrowth percentage in plants up to 20 cm height.

In treatments with saflufenacil application, DM differed only between whole plants with and without leaves, with lower DM in defoliated plants (Table 3). Besides, defoliation of *Conyza* spp. plants decreased DM approximately by 50 to 70%, in the control treatment (whole leafy plant) and in treatments in which the plants had regrowth (cutting at 8 DBA, without herbicide application), showing statistical difference (Table 3). The results indicated the efficiency of saflufenacil in controlling *Conyza* spp. plants with injured leaves, for example, after harvesting damage. Moreover, the control and reduction of DM in defoliated *Conyza* spp. plants may be related to herbicide absorption in the plant stem.

Treatments with saflufenacil application on leaves (new and old ones) showed a necrotic area less than 20% in all evaluations (Table 4). Foliar applications of saflufenacil tend to concentrate in the meristematic regions of the plant and not translocate towards the roots (Budd *et al.*, 2017). Furthermore, it is known that this herbicide has a weak acid character, with greater translocation via xylem and limited translocation via phloem (Grossmann *et al.*, 2011), which may justify the low mobility from leaves. A

Table 3. Dry matter (DM) of *Conyza* spp. in different tissue conditions at 21 days after application (DAT) of saflufenacil.

Treatment	DM (g plant ⁻¹)	
	With leaves	Without leaves
Whole plant	7.72 aA	2.70 aB
Cutting 10 cm 8 DBA ¹	4.48 abA	1.70 abB
Cutting 20 cm 8 DBA	3.97 bcA	1.83 abB
Cutting 10 cm 1 DBA	1.28 cdA	0.75 bA
Cutting 20 cm 1 DBA	3.61 bcA	1.44 abA
Whole plant + SAFL ²	2.77 bcA	0.55 bB
Cutting 10 cm 8 DBA + SAFL	1.06 cdA	0.30 bA
Cutting 20 cm 8 DBA + SAFL	1.51 cdA	0.45 bA
Cutting 10 cm 1 DBA + SAFL	0.40 dA	0.30 bA
Cutting 20 cm 1 DBA + SAFL	0.52 cdA	0.37 bA
C.V.(%) ³	81.15	

Means followed by the same uppercase letter in the row, comparing leaf integrity for each evaluation period, and by the same lowercase letter in the column, comparing injury levels for each treatment, indicate no significant difference by the Tukey test ($P < 0.05$). ¹ DBA=Days before application of saflufenacil. ² SAFL=Saflufenacil. ³ Coefficient of variation.

study on saflufenacil absorption and translocation in wheat (*Triticum aestivum* L.) found that the absorbed herbicide showed low translocation from treated leaves, corroborating the data observed in the present study (Frihauf *et al.*, 2010).

The lowest average necrotic areas were observed in the treatment where the herbicide was applied only in the region of the cutting, being lower to the other treatments at 3, 5, and 7 DAT, with a necrotic area less than 2% (Table 4).

Table 4. Necrotic area (%) of *Conyza* spp. evaluated at different times after localized application of saflufenacil.

Treatment	Necrotic area (%)			
	1 DAT	3 DAT	5 DAT	7 DAT
New leaf	0.42 b	11.44 c	15.15 b	18.45 b
Old leaf	5.13 ab	9.28 c	15.10 b	15.70 b
New stem	8.33 a	23.04 b	22.12 b	24.45 b
Old stem	6.28 a	58.00 a	68.34 a	76.00 a
Cutting site	0.00 b	1.63 d	1.00 c	0.92 c
C.V.(%) ¹	51.64	19.60	14.87	3.56

DAT: days after application of treatments. Means followed by the same lowercase letter in the column, comparing the percentage of necrotic area for each treatment, indicate no significant difference by the Tukey test ($P < 0.05$). ¹ Coefficient of variation.

On the contrary, picloram application in the cut stem of a woody species (*Tectona grandis*) showed translocation via phloem, being efficient in controlling future shoots (Caldeira and Castro, 2012), which shows a difference in systemic herbicides about those with less mobility and support the

importance of knowing the behavior of the applied herbicide and the target weed. In the present study, the necrotic region resulting from stem application must be related to chlorophyll in the fleabane stem, which has a photosynthetically active area. The interaction of protoporphyrin IX with oxygen in

presence of light generates singlet oxygen, resulting in lipid peroxidation of membranes and interruption of chlorophyll and heme synthesis (Owen *et al.*, 2011). Moreover, products tend to produce hydrogen peroxide and lead to rapid necrosis and wilting, disrupting cell membranes and reducing herbicide translocation (Eubank *et al.*, 2013).

Saflufenacil application on the stem located close to the base (old stem) showed a larger necrotic area at 3, 5, and 7 DAT than the other treatments, demonstrating that this region has greater herbicide translocation capacity (Table 4). The biochemical characteristics of saflufenacil, such as pKa 4.4, octanol-water partition coefficient (Kow 2.6), and ion trapping mechanism, provide it with metabolic stability and assist in its translocation in the plant (Kleier *et al.*, 1996). When this herbicide is applied to the basal region of *Conyza* spp. plants, these characteristics allow transport via xylem for a longer period before total destruction of plant structures occurs, since damage to vascular tissues occurs more slowly after absorption. Moreover, it is known that photoassimilate distribution occurs from the producing region (source) to the metabolic and/or storage regions (sink) (Taiz and Zeiger, 2017). These factors may explain the larger necrotic area when saflufenacil was applied in the region close to the base, considered a source region.

CONCLUSION

Saflufenacil is efficient in the control of *Conyza* spp. having a higher action in leafless and cutting plants. The saflufenacil absorption is carried out by the stem in *Conyza* spp. plants. The leaves showed low absorption and translocation from the located application of saflufenacil and this herbicide translocated from the stem to the leaves.

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