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# MANAGEMENT OF GLYPHOSATE-RESISTANT HAIRY FLEABANE AND CONTRIBUTION OF THE PHYSIOLOGICAL POTENTIAL OF SEEDS TO RESISTANCE[1]

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**ABSTRACT** - Hairy fleabane (*Conyza bonariensis* L.) is a major weed of the conventional crop systems. Therefore, the objectives of the present study were to assess the responses of glyphosate-susceptible (S) and -resistant (R) *C. bonariensis* at various developmental stages and evaluate the physiological potential of seeds to propose alternative herbicides for the control of this weed. Two experiments were performed in replicates. The first experiment was performed in a greenhouse, arranged in a 2 × 3 × 10 factorial design. Specifically, two hairy fleabane biotypes (S and R) at different developmental stages (I, II, and III) were subjected to various treatments (glyphosate, chlorimuron-ethyl, metsulfuron-methyl, diclosulam, ammonium glufosinate, paraquat, paraquat+diuron, diquat, 2,4-D, and control). Percentage control was evaluated at 7, 14, 21, and 28 days after the application of the treatments (DAT), and shoot dry mass (SDM) was measured at 28 DAT. The second experiment was performed in a laboratory to evaluate the physiological potential of seeds based on the weight of 1000 seeds (TSW); shoot length (SL), radicle length (RL), total length (TL), fresh seedling mass (FSM), dry seedling mass (DSM), accelerated aging (AA) and cold test (CT), and germination (G) in response to cold and accelerated aging. The alternative herbicides tested effectively controlled biotype R up to the stage-I. Seeds of biotype R showed higher physiological potential in terms of all analyzed variables and exhibited greater tolerance to adverse conditions during seedling establishment. Therefore, strategies for the management of glyphosate-resistant hairy fleabane should aim at preventing new seed production.

**keywords**: *Conyza bonariensis*. Alternative herbicides. Control. Physiological performance.

## MANEJO DE BUVA RESISTENTE AO GLYPHOSATE E CONSEQUÊNCIAS DO POTENCIAL FISIOLÓGICO DE SEMENTES À RESISTÊNCIA

**RESUMO** - A buva (*Conyza bonariensis* L.) é umas das principais plantas daninhas dos sistemas de cultivos conservacionistas, especialmente por sua evolução à resistência ao herbicida glyphosate.  Os objetivos deste trabalho foram verificar a resposta de *Conyza bonariensis* suscetível (S) e resistente (R) ao glyphosate em diferentes estádios a herbicidas alternativos e avaliar o potencial fisiológico de sementes destes biótipos. Dois estudos foram realizados e repetidos, sendo o primeiro em casa de vegetação em esquema fatorial 2x3x10, sendo A: biótipos de buva (S e R); B: estádios de desenvolvimento (I (1-2 folhas), II (5-6 folhas) e III  (30-35 folhas)) e C: herbicidas (glyphosate, chlorimuron-etílico, metsulfuron-methyl, diclosulam, amônio-glufosinato, paraquat, paraquat+diuron,  diquat, 2,4-D além de testemunha não aplicada).O segundo estudo foi desenvolvido em laboratório, realizando-se avaliações do potencial fisiológico das sementes, a partir do por peso de mil sementes (PMS), germinação (G), primeira contagem da germinação (PG),  índice de velocidade de germinação (IVG) e emergência (IVE), comprimento da parte aérea (CPA), raiz (CR) e total (CT), matéria seca da parte aérea (MSPA), raízes (MSR) e total (MST), testes de frio e envelhecimento acelerado. O biótipo CR foi eficientemente controlado pelos herbicidas alternativos ao glyphosate, utilizados até o estádio de 6 folhas. As sementes do biótipo CR apresentaram desempenho fisiológico superior em todas as variáveis analisadas, com maior tolerância a condições adversas em seu estabelecimento. O manejo de buva resistente a glyphosate demanda a utilização de estratégias que visem evitar a produção de novas sementes.

**Palavras-chave**: *Conyza bonariensis.* Herbicidas alternativos. Controle. Desempenho fisiológico.

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# INTRODUCTION

In hairy fleabane, the evolution of resistance to glyphosate, a primary herbicide used for the management of this weed, has led to a gradual increase in its infestation in agricultural areas of Brazil, with *Conyza bonariensis* L. and *C. canadensis* L. being abundant in the south of the country (LAMEGO; VIDAL, 2008; HEAP, 2020). Infestation of glyphosate-resistant hairy fleabane during the establishment and initial development of crop led to substantial losses in soybean production, compelling the search for alternatives to this herbicide. In case of severe weed infestation, the productivity may be reduced by up to 70% (VARGAS et al., 2007).

Typically, for the management of *Conyza* spp. at the soybean pre-sowing stages, several glyphosate-related herbicides, such as 2,4-D, diclosulam, metsulfuron-methyl, chlorimuron-ethyl, diuron+paraquat, and/or paraquat, are used. Diuron+paraquat and paraquat complement the use of glyphosate, but are not associated with it (VARGAS et al., 2007). In addition, ammonium-glufosinate and saflufenacil are effective in controlling hairy fleabane (CESCO et al., 2019). However, following soybean seedling emergence, only chlorimuron-ethyl and diclosulam act as selective herbicides. In addition to the use of alternative herbicides, the efficiency of management is associated with the stage of weed development and weed control may even be impossible at advanced developmental stages (VARGAS et al., 2007; MOREIRA et al., 2010).

During the management of resistant hairy fleabane, the potential or adaptive value of the resistant biotype, which is related to the production, survival, and development of viable seeds in the presence of specific resources under a certain environment, must also be taken into account (NEVE et al., 2011). Previous studies have reported that the adaptive capacity of herbicide-resistant biotypes is inferior, similar, or superior to that of the susceptible biotypes (TRAVLOS; CHACHALIS, 2013). In *C. bonariensis*, however, herbicide resistance does not affect its adaptive value (MYLONAS et al., 2019). In addition, the glyphosate-resistant biotype of *C. bonariensis* shows a shorter development cycle, higher photosynthetic pigment content, greater height, and more dry mass accumulation (KASPARY et al., 2014; KASPARY et al., 2017).

Furthermore, seed production is a crucial factor for consideration in weed management. For instance, a single glyphosate-resistant *C. bonariensis* plant produces over 800,000 seeds, reflecting the potential for the spread of resistance (KASPARY et al., 2017). In this context, the production of numerous seeds per plant is one of the most important adaptive characteristics related to the evolution and spread of herbicide resistance (DAUER et al., 2006). However, successful dissemination also depends on the dispersal capacity and physiological potential of seeds, which determine their survival in soil and germination over time to establish and generate competitive plants (PEDERSEN et al., 2007).

Understanding the population dynamics of a species requires the knowledge of its physiological characteristics, and seed viability is a crucial parameter for defining the competitive capacity of a species and, therefore, managing herbicide resistance (NEVE et al., 2011; SCHAEDLER et al., 2013). To this end, the objectives of the present study were to test the responses of glyphosate-susceptible and -resistant biotypes of *C. bonariensis* at different developmental stages to alternative herbicides as well as to evaluate the physiological potential of seeds of these biotypes to control the spread of herbicide resistance.

# MATERIAL AND METHODS

## Experiments and biotypes

The present study involved two experiments. In the first experiment, the responses of glyphosate-resistant and -susceptible *C. bonariensis* at different developmental stages to alternative herbicides were tested. In the second experiment, the physiological potential of seeds of glyphosate-resistant *C. bonariensis* was tested. The resistant (R) biotype was obtained from an agricultural area with a history of soybean cultivation and glyphosate use (Jaboticaba, Rio Grande do Sul; 27°40′30.18″S, 53°17′.12″W), and the susceptible (S) biotype was obtained from an agricultural area without a history of glyphosate use (Frederico Westphalen, RS; 27°23′46.31″S, 53°25′ 39.50″W) (40 km from the area from where biotype R was obtained). A previous dose–response experiment has confirmed glyphosate resistance in biotype R (KASPARY et al., 2017).

## Experiment 1: Management of glyphosate-resistant *C. bonariensis*

This experiment was performed in a greenhouse from November to December 2012 and repeated from March to April 2013. The experimental design was completely randomized, arranged in a 2 × 3 × 10 factorial design. Specifically, two *C. bonariensis* biotypes (S and R) at various developmental stages [I (1-2 true leaves), II (5-6 true leaves), and III (30-35 true leaves or 20-25 cm tall)] were subjected to control (without application) or herbicide treatment [glyphosate (720 $g{\cdot}ha^{-1}$), chlorimuron-ethyl (40 $g{\cdot}ha^{-1}$), metsulfuron-methyl (3.6 $g{\cdot}ha^{-1}$), diclosulam (33.6 $g{\cdot}ha^{-1}$), ammonium-glufosinate (400 $g{\cdot}ha^{-1}$), paraquat (400 $g{\cdot}ha^{-1}$), paraquat+diuron (400+200 $g{\cdot}ha^{-1}$), diquat (400 g·ha$^{-1}$), and 2,4-D (1,007.5 g·ha$^{-1}$)].

Hairy fleabane was planted in a staggered manner to guarantee plants at three different developmental stages for a single application of the treatments. The plants were grown in 500 mL pots containing an agricultural substrate (TecnoMax®) and maintained in the greenhouse until reaching the pre-established stages of application. The treatments were applied with a $CO_2$-pressurized backpack sprayer equipped with a spray boom with four flat-fan nozzles (DG 110.015) at a spray volume of 200 L·ha$^{-1}$. During treatment application in November and December 2012, temperature was 26° C and relative humidity (RH) was 73%. During treatment application in March and April 2013, temperature was 24°C and RH was 81%.

Percentage control (%) was evaluated at 7, 14, 21, and 28 days after the application of the treatment (DAT) according to the scale proposed by SBCPD (1995) [0% (no weed control) to 100% (eradication)]. Moreover, the aerial parts were harvested at 28 DAT to determine shoot dry mass (MSPA). The results of the two replicates (I and II) were subjected to analysis of variance using SISVAR (FERREIRA, 2011); data without significant differences were pooled for further analysis. Significant differences in mean results of various treatments ($p \leq 0.05$) were analyzed using Tukey test. To assess interactions between factors, DMS average test with 5% probability was adopted.

**Experiment 2: Physiological potential of seeds and spread of herbicide resistance**

This experiment was conducted in a laboratory using seeds of biotypes R and S obtained from prior multiplication in a greenhouse during two periods, namely autumn–winter with harvest in December 2012 and spring–summer with harvest in January 2013 (replicates I and II, respectively). Following harvest, the seeds were dried at room temperature for 7 days and stored in a refrigerator for at least 30 days. Subsequently, the seeds were subjected to germination and physiological quality tests (described below) from January to February 2013.

Seed tests were performed as described in the Rules for Seed Analysis (RAS) (BRASIL, 2009) for *Lacuta sativa* belonging to the same family (Asteraceae) and with seeds of approximately the same size as *C. bonariensis*. Weight of 1000 seeds was evaluated with eight subsamples of 100 seeds weighed on a precision analytical balance (mg). For germination tests, a germination paper moistened with distilled water at an amount equivalent to 2.5× the mass of the dry paper was used as the substrate. Each paper sheet contained 100 seeds, and the samples were placed in a growth chamber at 20°C under a 12/12 h photoperiod. Germination counts were performed daily for 14 days, with only seeds with root protrusion longer than 2 mm considered germinated. At 7 and 14 days, normal seedlings were counted, corresponding to the first germination count (FGC) and final germination (G), respectively. The germination speed index (GSI) was calculated according to Maguire (1962). The emergence speed index (ESI) was determined using 100 seeds of each biotype (4 sub-samples of 25 seeds), manually sown in trays (20 × 10 × 7 cm$^3$) containing an agricultural substrate and incubated in the greenhouse. The counts were performed daily for 21 days, and only seedlings taller than 2 mm were considered to have emerged. At 28 days after the initiation of the experiment, 10 normal seedlings were removed and the shoot length (SL), radicle length (RL), total length (TL) were measured (cm seedling$^{-1}$). Subsequently, each replicate was separately dried in an air circulation oven at 70°C. After 48 h, shoot dry mass (SDM), root dry matter (RDM), and total dry matter (TDM) were determined using an analytical balance (g seedling$^{-1}$).

The accelerated aging test was performed with four subsamples of 50 seeds per biotype using transparent acrylic boxes with a lid (Gerbox), serving as humid mini-chambers containing 40 mL distilled water. A screen was placed above water, and the seeds were placed on this screen. The boxes were placed in a biochemical oxygen demand (BOD) chamber at 42°C for 48 h (VIEIRA; CARVALHO, 1994). Thereafter, as described in the germination tests, the germinated seeds and normal seedlings were counted at 7 days.

The cold test was performed with four subsamples of 50 seeds per biotype. The seeds were placed on a paper within a Gerbox and incubated in a BOD chamber at 10°C for 7 days. Thereafter, the germination test was conducted as described above at a constant temperature of 20°C for 14 days.

The results of two replicates (I and II) were submitted to analysis of variance using SISVAR (FERREIRA, 2011) and analyzed separately according to the period of seed harvest. Significant differences in mean results for the two biotypes ($p \leq 0.05$) were analyzed using *t*-test.

# RESULTS AND DISCUSSION

**Management of glyphosate-resistant *C. bonariensis***

There was a significant biotype × herbicide × developmental stage interaction ($p \leq 0.05$). At 7 DAT, the tested herbicides were the most effective against plants at early developmental stages (Table 1). However, the effectiveness of glyphosate did differ significantly across development stages, although this herbicide had no effect on biotype R. These results indicate that the stage of application and degree of control are correlated, which is consistent with previous reports. Vargas et al. (2007) also obtained favorable results with the application of 2,4-D, paraquat, chlorimuron-ethyl, and metsulfuron-methyl at early developmental stages of *Conyza* sp. Meanwhile, a significant loss of *C. sumatrensis* control with chlorimuron-ethyl has been reported in plants taller than 10 cm due to the higher density of foliar trichomes, which blocked herbicide penetration (SANTOS et al., 2015).

**Table 1**. Control (%) of glyphosate-susceptible (S) and -resistant (R) *Conyza bonariensis*, at 7 and 14 days after application of treatments (DAT).

| Herbicides | Susceptible | | | Resistant | | |
|---|---|---|---|---|---|---|
| | Stages[1] | | | | | |
| | I | II | III | I | II | III |
| | 7 DAT | | | | | |
| Control | A 0.00[2]ns e | A 0.00ns f | A 0.00 ns h | A 0.00 e | A 0.00 g | A 0.00 g |
| Glyphosate | A 35.71* c | A 37.86* d | A 36.87* e | A 0.00 e | A 0.00 g | A 0.00 g |
| Chl. Ethyl[3] | B 23.12* d | A 32.14* e | C 18.57* g | A 38.75 d | B 21.87 e | C 14.29 f |
| Met. methyl[4] | A 39.57* c | A 39.17* d | B 23.33ns f | A 44.37 c | B 27.50 d | C 21.25 e |
| Diclosulam | A 42.00* c | A 43.75* c | B 24.37* f | A 36.25 d | B 19.33 f | B 20.86 e |
| Glufosinate | A 100.00* a | A 99.28* a | B 80.00* c | A 96.25 b | B 92.62 b | C 65.00 c |
| Paraquat | A 100.00ns a | A 99.00* a | B 84.37* b | A 100.00 a | B 94.75 b | C 77.50 b |
| Par.+Diuron[5] | A 100.00ns a | B 90.25* a | C 86.25* b | A 98.12 ab | B 92.85 b | C 77.14 b |
| Diquat | A 100.00ns a | B 95.71* a | C 90.62* a | A 99.75 ab | A 98.71 a | B 83.57 a |
| 2,4-D | A 53.57* b | A 52.86* b | B 40.00*d | A 43.10 c | B 41.25 c | C 34.16 d |
| Average | 32.33 | 59.00 | 48.44 | 50.73 | 48.89 | 39.38 |
| C.V. (%)[6] | 6.04 | | | | | |
| | 14 DAT | | | | | |
| Control | A 0.00ns f | A 0.00ns e | A 0.00ns g | A 0.00 e | A 0.00 d | A 0.00 d |
| Glyphosate | A 85.29* c | A 84.29* b | B 70.62* c | A 0.00 e | A 0.00 d | A 0.00 d |
| Chl. Ethyl | A 60.62* e | A 58.57* d | B 39.28ns f | A 76.87 c | B 66.87 b | C 33.57 c |
| Met. methyl | A 69.29* d | A 69.17ns c | B 47.50ns e | A 83.12 b | B 65.62 b | C 47.00 b |
| Diclosulam | A 75.00* d | A 74.37* c | B 54.37ns d | A 83.75 b | B 58.33 c | C 49.28 b |
| Glufosinate | A 100.00ns a | A 99.71ns a | B 83.12ns b | A 99.25 a | A 99.75 a | B 82.85 a |
| Paraquat | A 100.00ns a | AB 99.28ns a | B 94.62ns a | A 100.00 a | A 98.62 a | B 89.17 a |
| Par.+ Diuron | A 100.00ns a | B 94.25ns a | AB 97.25* a | A 100.00 a | A 95.71 a | B 87.14 a |
| Diquat | A 100.00ns a | A 99.71ns a | A 97.00* a | A 99.50 a | A 99.00 a | B 90.00 a |
| 2.4-D | A 89.16* b | A 86.42* b | B 80.62* b | A 71.25 d | B 60.00 c | C 51.50 b |
| Average | 77.94 | 76.58 | 63.04 | 71.37 | 64.39 | 52.95 |
| C.V. (%) | 8.07 | | | | | |

[1]Stage I – plants with 1-2 true leaves, Stage II- plants with 5-6 true leaves; and Stage III - plants with 30-35 leaves (20-25 cm in height). [2]Means preceded by different capital letters, compared in rows (stadiums) or followed by different lower-case letters, compare in columns (herbicides), differ by the t test (p≤0.05). ns and * compare means of the biotypes (S and R), within the same stages and herbicides. [3]Chlorimuron-ethyl, [4]Metsulfuron-methyl, [5]paraquat + diuron, [6]Coefficient of variation.

Paraquat and diquat were more effective when applied to smaller plants, reaching control close to 100% at stages I and II of development in both biotypes (Table 1). This rapid control can be attributed to the high capacity of these herbicides to inhabit photosynthesis by acting as electron receptors to generate free radicals ($O_2$ superoxides), which in turn damage the plasma membrane, allowing the extravasation of cellular content and ultimately causing cell death in just a few hours (MARKWELL; NAMUTH; HERNÁNDEZ-RIOS, 2006). However, contact herbicides require an effective coverage of the target, and satisfactory control may therefore not be achieved in large plants, leading to lateral regrowth, as observed in plants at stage III of development (25 cm tall). These results are consistent with the findings reported by Vargas et al. (2007), who obtained satisfactory results with the application of paraquat and paraquat+diuron at the 4-leaf stage of *Conyza* sp. Moreover, Santos et al. (2015) could achieve satisfactory control by applying glufosinate ammonium and paraquat+diuron at the 7-8 leaf stage in five *Conyza* biotypes. Conversely, Moreira et al. (2010) reported that paraquat+diuron and ammonium-glufosinate were ineffective when applied to plants with 10 leaves and led to lateral regrowth.

At 14 DAT, all tested herbicides were the most effective when applied at the early stages of development (Table 1) but relatively less effective when applied to plants at stage III of development (up to 25 cm tall). As glyphosate alternatives, systemic herbicides were inferior to contact herbicides; as such, when applied to biotype S at stage I of development, chlorimuron-ethyl achieved only 60% control, while paraquat achieved 100% control (Table 1). Overall, there were no significant differences between the biotypes and herbicides applied at each developmental stage, except for glyphosate, which achieved 0% control in biotype R.

At 21 DAT, all tested herbicides achieved 100% control when applied at the early stages of development in both biotypes (Table 2). Overall, there were no significant differences in control between the two biotypes. However, when applied at stage III of development, 2,4-D achieved a lower control in biotype R (66%) than in biotype S (88%). Of note, diclosulam, metsulfuron-methyl, and chlorimuron-ethyl achieved unsatisfactory control in both biotypes when applied at stage III of development (65%, 60%, and 40%, respectively).

At 28 DAT, all tested alternative herbicides were efficient in controlling biotype R when applied to up to stage II of development (six leaves) (Table 2). These results are consistent with the findings reported by Vargas et al. (2007), who achieved complete control of biotype R by applying chlorimuron-ethyl or metsulfuron-methyl to plants bearing four to five leaves. Our results also corroborate previous observations that spraying of *C. bonariensis* plants at early developmental stages could achieve better control by reducing regrowth capacity, as young plants are more sensitive to herbicides (MOREIRA et al., 2010 OKUMU et al., 2019). In addition, when applied at early stages of development, 2,4-D achieved better control (80%) than glyphosate in biotype R (VARGAS et al., 2007). However, hairy fleabane has a high seed production capacity, reaching a rate of over 800,000 seeds per plant (KASPARY et al., 2017). Therefore, 80% control of the population in a specific area may become ineffective to prevent the spread of the resistant biotypes of *C. bonariensis.*

When applied at stage III of development (plants with 30–35 leaves or 25 cm tall), metasulfuron-methyl and chlorimuron-ethyl did not achieve satisfactory control in both biotypes and 2,4-D did not efficiently control biotype R (Table 2). These results indicate that the developmental stage at the time of application affects the efficiency of herbicides (MYLONAS et al., 2019). This is because plants accumulate more dry matter as they develop, acquiring a greater capacity to survive under adverse conditions and recover from the phytotoxic effects of herbicides (OKUMU et al., 2019).

At 28 DAT, MSPA was higher as the plants were taller at this stage of application (Table 3). Moreover, little control could be achieved when herbicides were applied at advanced developmental stages. This is consistent with our results of visual assessment of percentage control using the scale proposed by SBCPD as well as with observations reported by Olivera Netto et al. (2010). In general, herbicides showed little or no difference in dry mass accumulation between stages I and II of development. However, at stage III of development, contact herbicides markedly reduced MSPA due to their rapid action, whereas systemic herbicides (e.g., chlorimuron-ethyl, metsulfuron-methyl, diclosulam, and 2,4-D) had little overall effects on MSPA, specifically in biotype R (Table 3). *C. canadensis* is highly susceptible to 2,4-D at doses below 70 g $ha^{-1}$ (MCCAULEY; YOUNG, 2019), but *C. sumatrensis* is resistant to 2,4-D (PINHO et al., 2019; QUEIROZ et al., 2020). However, in the present study, the recommended dose of 2,4-D achieved over 85% control in biotype R at 28 DAT when applied at stages I and II of development (Table 2). However, at stage III of development, this value decreased to approximately 75%, indicting a clear effect of developmental stage at the time of 2,4-D application stage on its efficiency and its direct impact on dry mass accumulation (Table 3).

**Table 2**. Control (%) of glyphosate-susceptible (S) and -resistant (R) *Conyza bonariensis*, at 21 and 28 days after application of treatments (DAT).

| Herbicides | Susceptible | | | Resistant | | |
|---|---|---|---|---|---|---|
| | Stages[1] | | | | | |
| | I | II | III | I | II | III |
| | | | 21 DAT | | | |
| Control | A 0.00$^{2ns}$ c | A 0.00$^{ns}$ d | A 0.00$^{ns}$ e | A 0.00 c | A 0.00 e | A 0.00 e |
| Glyphosate | A 99.71* a | A 99.71* a | B 91.87* ab | A 0.00 c | A 0.00 e | A 0.00 e |
| Chl. Ethyl[3] | A 98.87$^{ns}$ a | B 77.14$^{ns}$ c | C 45.00$^{ns}$ d | A 98.00 a | B 78.75 c | C 39.28 d |
| Met. methyl[4] | A 100.00$^{ns}$ a | B 81.67* bc | C 65.83$^{ns}$ c | A 98.12 a | A 96.62 b | B 59.37 c |
| Diclosulam | A 100.00$^{ns}$ a | B 85.62* b | C 68.75$^{ns}$ c | A 100.00 a | B 70.00 d | B 64.28 bc |
| Glufosinate | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | B 89.37$^{ns}$ b | A 99.50 a | A 100.00 a | A 92.86 a |
| Paraquat | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 97.50$^{ns}$ a | A 100.00 a | A 99.75 a | B 91.67 a |
| Par.+Diuron[5] | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 94.37$^{ns}$ ab | A 100.00 a | A 99.71 a | A 93.00 a |
| Diquat | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 97.50$^{ns}$ a | A 99.12 a | A 98.14 a | A 96.00 a |
| 2.4-D | A 100.00* a | A 98.00* a | B 88.50* b | A 83.12 b | A 82.50 b | B 66.67 b |
| Average | 89.86 | 84.21 | 73.87 | 77.79 | 72.55 | 60.31 |
| C.V. (%)[6] | | | 9.35 | | | |
| | | | 28 DAT | | | |
| Control | A 0.00$^{ns}$ b | A 0.00$^{ns}$ c | A 0.00$^{ns}$ e | A 0.00 b | A 0.00 c | A 0.00 e |
| Glyphosate | A 100.00* a | A 100.00*a | B 91.87* a | A 0.00 b | A 0.00 c | A 0.00 e |
| Chl. ethyl | A 99.00$^{ns}$ a | B 85.00* b | C 51.43* d | A 99.75 a | A 99.37 a | B 37.14 d |
| Met. methyl | A 100.00$^{ns}$ a | B 78.83* b | C 63.33$^{ns}$ c | A 100.00 a | A 96.87 a | B 67.50 c |
| Diclosulam | A 100.00$^{ns}$ a | B 78.12* b | B 81.37* b | A 100.00 a | A 97.50 a | B 73.57 bc |
| Glufosinate | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 93.87$^{ns}$ a | A 100.00 a | A 100.00 a | A 94.00 a |
| Paraquat | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 96.25$^{ns}$ a | A 100.00 a | A 100.00a | A 94.13 a |
| Par.+ Diuron | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 93.12$^{ns}$ a | A 100.00 a | A 100.00 a | A 96.42 a |
| Diquat | A 100.00$^{ns}$ a | A 100.00$^{ns}$ a | A 97.50$^{ns}$ a | A 99.75 a | A 98.57 a | A 98.57$^{a}$ |
| 2.4-D | A 100.00* a | A 95.00* a | A 92.50* a | A 91.25 a | A 85.62 b | B 76.66 b |
| Average | 89.90 | 83.70 | 76.42 | 79.08 | 77.79 | 63.80 |
| C.V. (%) | | | 9.33 | | | |

[1]Stage I – plants with 1-2 true leaves, Stage II- plants with 5-6 true leaves; and Stage III - plants with 30-35 leaves (20-25 cm in height). [2]Means preceded by different capital letters, compared in rows (stadiums) or followed by different lower-case letters, compare in columns (herbicides), differ by the t test ($p \leq 0.05$). ns and * compare means of the biotypes (S and R), within the same stages and herbicides. [3]Chlorimuron-ethyl, [4]Metsulfuron-methyl, [5]paraquat + diuron, [6]Coefficient of variation.

**Table 3**. Dry shoot weight (mg plant$^{-1}$) of glyphosate-susceptible and -resistant *Conyza bonariensis*, 28 days after application of treatments (DAT).

| Herbicides | Susceptible | | | Resistant | | |
|---|---|---|---|---|---|---|
| | Stages[1] | | | | | |
| | I | II | III | I | II | III |
| Control | C 288.7[2] ns a | B 1161.3ns a | A 4885.2ns a | C 273.7 a | B 1201.0 a | A 4783.8 a |
| Glyphosate | B 10.6* b | B 10.7* b | A 1071.0* d | C 269.0 a | B 1232.1 a | A 4936.2 a |
| Chl. Ethyl[3] | B 6.4 b | B 53.5* b | A 2033.0* c | C 12.2 b | B 73.7 c | A 2661.7 b |
| Met. methyl[4] | B 4.7ns b | B 34.2* b | A 3092.4ns b | C 12.2 b | B 49.8 b | A 2995.7 b |
| Diclosulam | B 5.1ns b | B 72.0ns b | A 1885.7* c | B 15.9 b | B 78.0 b | A 2878.4 b |
| Glufosinate | B 5.9ns b | B 6.6ns b | A 984.7* d | B 20.1 b | B 21.5 b | A 1247.2 c |
| Paraquat | B 2.6ns b | B 5.9ns b | A 1080.0* d | B 18.7 b | B 16.6 b | A 1606.0 c |
| Par.+Diuron[5] | B 3.0ns b | B 9.1ns b | A 976.8* d | B 3.0 b | B 27.2 b | A 1346.2 c |
| Diquat | B 5.4ns b | B 6.1ns b | A 1078.0* d | B 5.9 b | B 38.4 b | A 1426.3 c |
| 2.4-D | B 6.6ns b | B 19.7ns b | A 1829.7ns c | B 35.5 b | B 64.5 b | A 2435.1 b |
| Average | 33.9 | 137.9 | 1.891.7 | 66.7 | 280.3 | 2.631.7 |
| CV (%)[6] | | | 21.43 | | | |

[1]Stage I - plants with 1-2 true leaves, Stage II - plants with 5-6 true leaves; and Stage III - plants with 30-35 leaves (20-25 cm in height). [2]Means preceded by different capital letters, compared in rows (stadiums), or followed by different lower-case letters, compare in columns (herbicides), differ by the t test ($p \leq 0.05$). ns and * compare means of the biotypes (S and R), within the same stages and herbicides. [3]Chlorimuron-ethyl, [4]Metsulfuron-methyl, [5]paraquat + diuron, [6]Coefficient of variation.

Overall, the use of alternative herbicides with diverse mechanisms of action proved to be efficient in controlling glyphosate-resistant and -susceptible *C. bonariensis* (Tables 1, 2 and 3). The sensitivity of biotype R to these herbicides also rules out the possibility of multiple resistance to the evaluated products. However, herbicides acting on the acetolactate synthase (ALS) should be cautiously used, since for *C. canadensis* and *C. sumatrensis* were already reported to have developed resistance to multiple ALS-inhibiting herbicides (SANTOS et al. 2015; PINHO et al., 2019).

In addition, previous studies have reported that *C. bonariensis* exhibited multiple resistance to glyphosate and paraquat (Resistance Factor = 149 for a 5-enolpyruvylshikimate-3-phosphate synthase inhibitor) (MORETTI et al., 2013; ZOBIOLE et al., 2019). These results demonstrate the worsening of the problem of herbicide resistance in *C. bonariensis*, restricting the number of alternative herbicides for weed control considering the risk of development of multiple resistance. Recently, however, the combined use of herbicides was approved, and alternatives such as halauxifen-methyl, diclosulam, dicamba, and 2,4-D are increasingly being used for managing the populations of glyphosate-resistant weeds (ZIMMER et al., 2018; KRENCHINSKI et al., 2019).

**Physiological potential of seeds and spread of herbicide resistance**

There were significant differences ($p<0.05$) in the physiological potential of seeds between biotypes S and R (Table 4). In replicate I, weight of 1000 seeds in biotype S was lower than that in biotype R (3.90 and 5.10 mg, respectively). Seeds with a higher vigor containing greater amounts of reserves are heavier and can germinate and emerge faster, better surviving under adverse conditions (VIEIRA; CARVALHO, 1994). For weeds, greater initial vigor represents greater potential to compete with the crop once it is established first in an area; this offers an competitive advantage to weeds for resources and leads to the loss of productivity of crops.

The germination test, the first germination count, and the germination speed index were higher for seeds of biotype R than for biotype S, with 17, 10, and 15% higher values, respectively (Table 4). The ESI showed superior ability to establish for the

R biotype. This indicates its priority in the use of resources when in the field, dominance in the ecological niche early and, consequently, that it will have greater competitive potential when in contact with plants of the same species or with cultures (GUSTAFSON et al., 2004). However, in a study with biotypes originating from Greece, similar development of successful biotypes and those resistant to glyphosate by *C. bonariensis* was shown (MYLONAS et al., 2019). This fact shows the differential behavior according to the origin of the biotypes being analyzed.

**Table 4**. Thousand seed weight (TSW), first Germination Count (FGC), Germination (G), germination speed index (GSI), emergence speed index (ESI), shoot length (SL), radicle length (RL), total length (TL), fresh seedling mass (FSM), dry seedling mass (DSM), accelerated aging (AA) and cold test (CT), in seeds of of glyphosate-susceptible (S) and -resistant (R) *Conyza bonariensis.*

| Experiment I | | | | | | |
|---|---|---|---|---|---|---|
| Biotype | TSW (mg) | FGC (%) | G (%) | GSI | ESI | SL (cm) |
| S | 3.91* | 24* | 67* | 24.66* | 10.83* | 0.65* |
| R | 5.12 | 33 | 84 | 38.02 | 16.43 | 1.08 |
| Average | 4.51 | 28 | 75 | 31.34 | 13.63 | 0.87 |
| C.V. (%)[1] | 4.38 | 9.30 | 5.48 | 6.74 | 4.90 | 5.59 |
| Biotype | RL (cm) | TL (cm) | FSM (mg) | DSM (mg) | AA (%) | CT (%) |
| S | 2.53* | 3.18* | 10.70* | 1.60* | 35* | 47* |
| R | 4.16 | 5.24 | 24.80 | 3.80 | 53 | 79 |
| Average | 3.35 | 4.21 | 17.80 | 2.70 | 44 | 62 |
| C.V. (%) | 7.70 | 6.59 | 16.09 | 8.23 | 4.47 | 5.12 |
| Experiment II | | | | | | |
| Biotype | TSW (mg) | FGC (%) | G (%) | GSI | ESI | SL (cm) |
| S | 4.00* | 22* | 59* | 31.34* | 10.98* | 1.05* |
| R | 5.50 | 34 | 86 | 45.15 | 16.93 | 1.64 |
| Average | 4.80 | 28 | 72 | 38.25 | 13.96 | 1.34 |
| C.V. (%)[1] | 2.66 | 5.43 | 8.05 | 2.96 | 7.46 | 6.20 |
| Biotype | RL (cm) | TL (cm) | FSM (mg) | DSM (mg) | AA (%) | CT (%) |
| S | 1.67* | 2.72* | 10.91* | 1.30* | 43* | 57* |
| R | 3.17 | 4.81 | 21.60 | 3.82 | 54 | 95 |
| Average | 2.42 | 3.76 | 16.25 | 2.56 | 48 | 76 |
| C.V. (%) | 5.38 | 4.19 | 10.09 | 12.94 | 5.80 | 2.94 |

[1]Coefficient of variation. *Means of different biotypes (susceptible (S) and resistant (R)) in the columns, differ by the t test ($p \leq 0.05$). ns Not significant ($p \geq 0.05$).

The higher speed of establishment of the biotype R culminated in a greater length of the aerial part, root, and total length of the seedlings (Table 4). These results differ from those obtained for plants of the *Echinochloa* sp. that were resistant to quinclorac. The plants obtained inferior root performance, resulting in lower ability to explore the environment, speed of establishment, and survival under competitive conditions (SCHUCH et al., 2008). The results obtained for fresh and dry seedling mass, showed a 50% lower capacity of biotype S in biomass accumulation in comparison with biotype R; in this case, the seedling dry mass was 1.60 and 3.80 mg for S and R, respectively (Table 4). This lower accumulation results from the lower speed of establishment and development, which directly interferes with the use of soil nutrients, and the quality of incident light. This results in a lower photosynthetic rate and accumulation of photoassimilates.

Under natural conditions, seeds are often exposed to environmental stresses that limit germination and the possibility of survival. The accelerated aging test and the cold test, which reproduce stressful conditions, reduced the germination of biotype S by approximately 30, 20, 31, and 4%, for the first germination count, germination, germination speed index, and emergence speed index in comparison with the biotype R, respectively (Table 4). In this way, the depreciative effect on the seeds of both the *C. bonariensis* biotypes was contacted. However, even with a high reduction in germination by the accelerated aging test, seeds of biotype R maintained germination above 50%, while for S it was only 35%. These results corroborate the weight per thousand seeds as higher for the biotype R, because even though part of its reserves were consumed under conditions of high temperature and humidity as in the aging test, there was still enough quantity to germinate, a fact, which at the field level, would allow for its best establishment.

A study developed with different temperature regimes on *C. bonariensis* seeds showed no variation in the germination percentage and vigor between resistant and susceptible biotypes (TRAVLOS; CHACHALIS, 2013). However, *Bromus tectorum*, which is resistant to ACCase-inhibiting herbicides (acetyl-coenzyme-A-carboxylase), demonstrated superior performance for physiological quality and germination variables (PARK et al., 2004). For the biotype of *Fimbristylis miliacea*, which is susceptible to inhibitors of the ALS enzyme, a higher percentage of germination and germination speed index were observed when compared to biotopes of the same resistant species (SCHAEDLER et al., 2013). Similarly, the final germination of the seeds of the *Kochia scoparia* biotype, which is susceptible to the herbicides dicamba and fluroxypyr, was 40% higher than that of the resistant biotype at different temperatures evaluated (KUMAR; JHA, 2016). Emphasis is given to the distinct effect of resistance to different herbicides on seed quality and germination in different species and even for distinct biotypes of the same species. In this context, it is not possible to outline management strategies for one species based on knowledge of the competitive capacity of another.

The performance of the seeds from Experiment 2 (harvested in January) was similar to that observed for the seeds of Experiment 1 (harvested in December), showing a superior tendency of the biotype R for the analyzed variables (Table 4). The seeds of R presented greater weight per thousand seeds than for S, at 5.50 and 4.00 mg, respectively. The greater accumulation of reserves in the seeds may be linked to the higher level of photosynthetic pigments in plants of biotype R when compared to S and, consequently, greater efficiency in the use of light, as reported in studies with the same biotypes carried out by Kaspary et al. (2014). However, for the same biotypes, less accumulation of leaf area by S was observed, possibly influencing light uptake (KASPARY et al., 2017). Thus, the greater mass of the biotype R results from its superior characteristics, which together culminate in a greater quantity of reserves, high germination potential, and superior physiological quality. The weight per thousand seeds for *Lolium rigidum* resistant to glyphosate were also superior in comparison with seeds of susceptible plants (PEDERSEN et al., 2007).

The conyza biotypes R and S cultivated in two different periods of development and that gave rise to the seeds used in this research, showed a reduction in the cycle duration and in the number of seeds produced per plant in the second growing season (KASPARY et al., 2017). However, the weight per thousand seeds observed shows a small increase for the seeds of both biotypes grown in the spring/summer period (Table 4). Therefore, a partial compensatory effect is inferred, with a reduction in the number of seeds produced, but with an increase in the accumulation of reserves by them and, consequently, greater physiological potential of the seeds obtained in the second growing season. However, in general, the other variables analyzed did not reflect superior behavior for the seeds obtained in the spring/summer, but only for the difference between the biotypes.

Susceptible *C. bonariensis* seeds, produced under the same cultivation conditions, showed lower physiological quality when compared to R, based on the first germination count, germination, germination speed index, and emergence speed index (Table 4). In this scenario, it is inferred that the biotype R has priority in the use of the resources of the environment, accelerating its capacity of establishment and competitiveness with the biotype S or with the existing culture, without affirming that this is an isolated consequence of the resistance. In addition, the superior root length presented by the resistant biotype provides rapid growth of the roots, and can confer a competitive advantage in cases in which interactions below ground predominate (SEIBERT; PEARCE, 1993). However, in a similar study, Piasecki et al. (2019) observed a reduction in the physiological quality of glyphosate-resistant *C. bonariensis* seeds, demonstrating a possible effect of the origin of the biotypes on the physiological capacity of the seeds.

The accelerated aging test is related to the conservation potential of seed reserves and, for this reason, it is considered as one of the most sensitive tests for the assessment of physiological quality and/ or vigor (SILVA; ROSSETTO, 2012; VENSKE et al., 2015). The cold test is also a vigor test, linked to the ability of the seeds to resist adverse conditions. In general, the possibility that these seeds will germinate in a wide range of soil moisture and temperature conditions increases with the closeness of the vigor tests results with those obtained in the germination test (VIEIRA; CARVALHO, 1994). In this context, the ability of resistant biotype seeds to remain viable for a longer period in the soil seed bank is emphasized until ideal germination conditions are reached. The other variables evaluated also point to better physiological quality of glyphosate-resistant *C. bonariensis* seeds, and even in tests that simulate adverse conditions of temperature and humidity, the germination of this biotype was good and excellent (54 and 95%), respectively, for accelerated aging and cold testing. Seeds of the S biotype showed values of 43 and 57%, respectively, for the same tests (Table 4). Thus, in adverse conditions of establishment, the biotype R shows an adaptive advantage and can thus occupy the area at the expense of biotype S, ensuring the spread of resistance.

Seeds of *C. bonariensis*, from two growing seasons by biotype resistant to the herbicide glyphosate, presented superior physiological potential when compared to a susceptible biotype, although the finding cannot be based only on the question of resistance. Proper management of this species should be prioritized, either with alternative herbicides or by methods that prevent seed production, since it is linked to superior quality by propagules, increasing the potential for seedling establishment and allowing the perpetuation of resistance to the herbicide.

## CONCLUSION

The biotype of *Conyza bonariensis* resistant to the herbicide glyphosate can be efficiently controlled by the alternative herbicides chlorimuron-ethyl, metsulfuron-methyl, diclosulam, ammonium-glufosinate, paraquat, paraquat+diuron, diquat, and 2,4-D at a stage of up to 6 leaves, discarding it if there is a possibility of multiple resistance.

Seeds of the glyphosate-resistant *C. bonariensis* biotype can show superior physiological performance, as observed in this study, with greater tolerance to adverse conditions in its establishment as a seedling, requiring the management of resistance in the forest by using strategies that prevent the production of new seeds.

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## REFERENCES

BRASIL. **Regras para análise de sementes**. Ministério da Agricultura, Pecuária e Abastecimento. Secretaria de Defesa Agropecuária. Brasília, DF: Mapa/ACS, 2009.

CESCO, V. J. S. et al. Management of resistant conyza spp. during soybean pre-sowing. **Planta Daninha**, 37: e19181064 , 2019.

DAUER, J.T., MORTESEN, D.A., HUMSTON, R. Controlled experiments to predict horseweed (*Conyza canadensis*) dispersal distances. **Weed Science**, 54: 484– 489, 2006.

FERREIRA, D. F. Sisvar: a computer statistical analysissystem. **Ciencia e Agrotecnologia**, 35: 1039 -1042, 2011.

GUSTAFSON, D. J. et al. Competitive relationships of *Andropogon gerardii* (Big Bluestem) from remnant and restored native populations and select cultivated varieties. **Functional Ecology**, 18: 451-457, 2004.

HEAP, I. **The International survey of herbicide resistant weeds**. Available in: <www.weedscience.com>. Accessed in: 15 mar. 2020.

KASPARY, T. E. et al. Determinação de pigmentos fotossintéticos em biótipos de buva suscetível e resistente ao herbicida glyphosate. **Planta Daninha**, 32: 417-426, 2014.

KASPARY, T. E. et al. Growth, phenology, and seed viability between glyphosate-resistant and glyphosate-susceptible hairy fleabane. **Bragantia**, 76: 92-101, 2017.

KRENCHINSKI, F. H. et al. Halauxifen-Methyl+Diclosulam: New option to control *Conyza spp*. prior soybean sowing. **Planta Daninha**, 37: e019189000, 2019.

KUMAR, V.; JHA, P. Differences in germination, growth, and fecundity characteristics of dicamba-fluroxypyr-resistant and susceptible Kochia scoparia. **PLoS ONE**, 11: e0161533, 2016.

LAMEGO, F. P.; VIDAL, R. A. Resistência ao glyphosate embiótipos de *C. bonariensis e C. canadensis* no Estado do Rio Grande do Sul, Brasil.

**Planta Daninha**, 26: 467-471, 2008.

MAGUIRE, J.D. Speed of germination-aid in selection and evaluation for seedlig emergence and vigor. **Crop Science**, 2: 176-177,1962.

MARKWELL, J.; NAMUTH, D.; HERNÁNDEZ-RIOS, I. **Introducción a los herbicidas que actúan através de la fotosintesis**. 2006. Disponível em: <https://passel.unl.edu/pages/informationmodule.php?idinformationmodule=1024932941>. Acesso em: 02 out. 2013.

MCCAULEY, C. L; YOUNG, B. G. Differential response of horseweed (*Conyza canadensis*) to halauxifen-methyl, 2,4-D, and dicamba. **Weed Technology**, 33: 673-679, 2019.

MOREIRA, M. S. et al. Herbicidas alternativos para o controle de biótipos de *Conyza bonariensis* e *Conyza canadensis* resistentes ao glyphosate. **Planta Daninha**, 28: 167-175, 2010.

MORETTI, M. L. et al. Glyphosate resistance is more variable than paraquat resistance in a multiple-resistant hairy fleabane (*Conyza bonariensis*) population. **Weed Science**, 61: 396-402, 2013.

MYLONAS, P. N. et al. Dose-response and growth rate variation among glyphosate resistant and susceptible *Conyza albida* and *Conyza bonariensis* populations. **Journal of Plant Protection Research**, 59: 32-40, 2019.

NEVE, P. et al. Modellingevolution and management of glyphosate resistance in *Amaranthus palmeri*. **Weed Research**, 51: 99–112, 2011.

OKUMU, M. N. et al. Growth-stage and temperature influence glyphosate resistance in *Conyza bonariensis (L.) Cronquist*. **South African Journal of Botany**, 121: 248-256, 2019.

OLIVEIRA NETO, A. M. et al. Estratégias de manejo de inverno e verão visando ao controle de *Conyza bonariensis* e *Bidens pilosa*. **Planta Daninha**, 28: 1107-1116, 2010.

PARK, K. W. et al. Ecological fitness of acetolactate synthase inhibitor-resistant and -susceptible downy brome (*Bromus tectorum*) biotypes. **Weed Science**, 52: 768–773, 2004.

PEDERSEN, B. P. et al. Ecological fitness of a *glyphosate*-resistant *Lolium rigidum* population: growth and seed production along a competition gradient. **Basic and Applied Ecology**, 8: 258–268, 2007.

PIASECKI, C. et al. Glyphosate resistance affect the physiological quality of *Conyza bonariensis* seeds. **Planta Daninha**, 37: e019213902, 2019.

PINHO, C. F. et al. First evidence of multiple resistance of Sumatran Fleabane (*Conyza sumatrensis* (Retz.) E. Walker) to five- mode-of-action herbicides. **Australian Journal or Crop Science**, 13: 688-1697, 2019.

QUEIROZ, A. R. S. et al. Rapid necrosis: a novel plant resistance mechanism to 2,4-D. **Weed Science**, 68: 6-18, 2020.

SANTOS, F. M. et al. Herbicidas alternativos para o controle de *Conyza sumatrensis* (Retz.) E. H. Walker resistentes aos inibidores da ALS e EPSPs. **Revista Ceres**, 62: 531-538, 2015.

SBCPD - Sociedade Brasileira da Ciência das Plantas Daninhas. **Procedimentos para instalação, avaliação e análise de experimentos com herbicidas**. Londrina: SBCPD, 1995. 42 p.

SCHAEDLER, C. E. et al. Germination and growth of *Fimbristylis miliacea* biotypes resistant and susceptible to acetolactate synthase-inhibiting herbicides. **Planta Daninha**, 31: 687-694, 2013.

SCHUCH, L. O. B. et al. Crescimento de raízes de biótipos de capim-arroz resistente e suscetível ao quinclorac em competição. **Planta Daninha**, 26: 893 -900, 2008.

SEIBERT, A. C.; PEARCE, R. B. Growth analysis of weed and crop species with reference to seed weight. **Weed Science**, 41: 52-56, 1993.

SILVA, L. F.; ROSSETTO, C. A. V. Physiological potential of sunflower seeds as by the artificial moistening**. Ciencia Rural**, 42: 1161-1167, 2012.

TRAVLOS, I. S; CHACHALIS, D. Relative competitiveness of *glyphosate*-resistant and *glyphosate*-susceptible populations of hairy fleabane, *Conyza bonariensis*. **Journal of Pest Science**, 86: 345-351, 2013.

VARGAS, L. et al. *Conyza bonariensis* resistente ao glyphosate na Região Sul do Brasil. **Planta Daninha**, 25: 573-578, 2007.

VENSKE, E. et al. Fatores abióticos sobre o efeito de herbicidas na qualidade fisiológica de sementes de arroz. **Revista Ciência Agronômica**, 46: 818-825, 2015.

VIEIRA, R. D.; CARVALHO, N. M. **Testes de vigor em sementes**. Jaboticabal, SP: FUNEP, p. 164, 1994.

ZIMMER, M. et al. Weed Control with Halauxifen-

Methyl Applied alone and in Mixtures with 2,4-D, Dicamba and Glyphosate. **Weed Technol**, 32: 597-602, 2018.

ZOBIOLE, L. H. S. et al. Paraquat resistance of sumatran fleabane (*Conyza sumatrensis*). **Planta Daninha**, 37: e019183264, 2019.