PLANTA DANINHA

SBCPD
SOCIEDADE BRASILEIRA DA CIÊNCIA DAS PLANTAS DANINHAS

<http://www.sbcpd.org>

ISSN 0100-8358 (print)
1806-9681 (online)

Article

RAPID DETECTION OF HORSEWEED AND BLACK PICKER SENSITIVITY LEVELS TO SAFLUFENACIL

Detecção Rápida de Níveis de Sensibilidade de Buva e Picão-Preto ao Saflufenacil

VARGAS, L.[1]*
HENCKES, J.R.[2]
GAVA, F.[3]
AGOSTINETTO, D.[2]
CARVALHO, I,R.[2]
RAMOS, R.[3]
RODRIGUES, R.[3]

ABSTRACT - Resistance to herbicides is one of the main factors responsible for weed control failures. Once uncontrolled and stressed, resistant weeds demand a new application of herbicides from different modes of action, which results in higher monetary costs and environmental impact. Knowledge of weed sensitivity levels to herbicides before making decisions for their management in the field is paramount to increasing the efficiency of integrated weed management. Thus, methods that allow the rapid determination of sensitivity levels can help to the correct recommendation before the application, and thus, favor the greater technical, economic and environmental efficiency of weed management. The objective of this work was to validate a rapid method to detect the levels of sensitivity of black picker (*Bidens pilosa*) and horseweed (*Conyza sumatrensis*) to saflufenacil. Dose-response experiments were performed in growth chamber, greenhouse and field. The control provided by different doses of the saflufenacil herbicide was evaluated in each environment, and the results correlated within each species. The results showed a significant positive correlation for the results obtained in growth chamber and greenhouse for black picker and horseweed. These results indicate that the method developed in the growth chamber can replace greenhouse tests for determination of the sensitivity of black picker and horseweed to saflufenacil.

Keywords: integrated weed management, resistance, control, efficiency.

*RESUMO - A resistência aos herbicidas é um dos principais fatores responsáveis por falhas no controle de plantas daninhas. Uma vez não controladas e estressadas, plantas daninhas resistentes demandam nova aplicação de herbicidas de diferentes modos de ação, o que resulta em maior custo monetário e impacto ambiental. O conhecimento dos níveis de sensibilidade das plantas daninhas aos herbicidas antes de serem tomadas decisões para seu manejo no campo é primordial para o aumento na eficiência do manejo integrado das plantas daninhas. Dessa forma, métodos que possibilitem a rápida determinação dos níveis de sensibilidade podem auxiliar na correta recomendação antes da aplicação e, desse modo, favorecer uma maior eficiência técnica, econômica e ambiental do manejo das plantas daninhas. O objetivo deste trabalho foi validar um método rápido para detecção dos níveis de sensibilidade de picão-preto (**Bidens pilosa**) e buva (**Conyza sumatrensis**) ao saflufenacil. Foram realizados experimentos dose-resposta em câmara de crescimento, casa de vegetação e em campo. O controle proporcionado por diferentes doses do herbicida saflufenacil foi avaliado em cada ambiente, sendo os resultados correlacionados dentro de cada espécie. Os resultados demonstraram correlação positiva significativa para os resultados obtidos em câmara de crescimento e casa de vegetação para picão-preto e buva. Esses*

*** Corresponding author:**
<leandro.vargas@embrapa.br>

Received: November 8, 2017
Approved: January 4, 2018

Planta Daninha 2019; v37:e019187603

[1] Embrapa Trigo, Passo Fundo-RS, Brazil. [2] Universidade Federal de Pelotas, Pelotas-RS, Brasil; [3] Basf S/A, São Paulo-SP, Brazil.

Doi: 10.1590/S0100-83582019370100022

resultados indicam que o método desenvolvido em câmara de crescimento pode substituir ensaios em casa de vegetação para determinações dos níveis de sensibilidade de picão-preto e buva ao saflufenacil.

Palavras-chave: manejo integrado de plantas daninhas, resistência, controle, eficiência.

INTRODUCTION

From the emergence of genetically modified crops that tolerate post-emergence herbicide applications, the weed chemical control method prevails. This is due to its high efficiency, low cost and easy management, in comparison to other methods (Agostinetto and Vargas, 2014). However, the indiscriminate application of herbicides has favored the selection of resistant biotypes (Yuan et al., 2007; Délye, 2013). The resistance of biotypes refers to the inherited ability to survive and reproduce after an exposure to a dose of herbicide that is usually lethal for the other individuals of the same population (Agostinetto and Vargas, 2014).

Weed resistance to herbicides is a serious and evolving problem in the world and a reason for concern among researchers (Heap, 2017). Resistant and uncontrolled weeds interfere in crops and reduce their productivity, and, in order to control them, the application of herbicides with different modes of action (isolated or mixed in tanks, or in sequential applications) is necessary, elevating the costs of the agricultural activity (Agostinetto and Vargas, 2014).

Failures in weed control may be attributed to the application of doses below those recommended by the manufacturers and mainly to resistance, and are mostly observed only after sowing and the establishment of the crops. On these situations, in the case of control failures and the need of a new herbicide application, the possibility of application of several herbicides with a different mode of action than the previously applied herbicide is restricted, since it may lead to phytotoxicity of the crop. In these situations, rapid methods to determine the sensitivity levels of weeds may be the threshold between success or failure in managing weeds and for the productivity of the crop.

The early identification of the sensitivity levels of weeds to herbicides through rapid methods allows the adoption of more assertive weed management methods and allows a correct and timely decision-making for weed management (Vidal et al., 2006; Dias et al., 2015). In addition, rapid methods may assist in the monitoring of weed populations in agricultural areas in response to the application of herbicides, preventing a dissemination of the resistance and contributing to a reduction in costs and in the impact in the management of resistant biotypes (Dias et al., 2015).

Rapid methods are promising and easy to plan and execute, since they are developed to allow tests to be conducted without the need for specialized equipment of materials. These tests are relatively simple and may even be conducted in the farm by an engineer- agronomist, accelerating the detection of the problem. The use of the filter paper and the commercial formulation of the product make it easier to conduct the test in the farm environment (Dias et al., 2015).

On the other hand, the quick test is not adequate to make a diagnosis and report new resistance cases; for such purposes, there are specific norms described by Gazziero et al. (2009). The tests conducted in the field and in greenhouses to confirm the sensitivity or resistance demand a lot of time and a large physical room, a large amount of material and labor. In general, the confirmation as to the resistance of biotypes to herbicides occurs through dose-response curve studies, in which it is possible to determine the necessary dose to reach 50% of control (C_{50}) and the necessary dose to reduce 50% of the dry mass (GR_{50}) by using doses above and below that recommended for a certain herbicide (Agostinetto and Vargas, 2014). This process may take 40-60 days in order to confirm the resistance to a certain biotype; in case there is the need for an immediate response, it ends up damaging the crop due to the competition by uncontrolled plants (Boutsalis, 2001).

The objective of this study was to validate a rapid method to detect the sensitivity levels of black picker (*Bidens pilosa*) and horseweed (*Conyza sumatrensis*) to saflufenacil, in experiments conducted in a Biological Oxigen Deman (BOD) germination chamber, in a greenhouse and in the field.

MATERIAL AND METHODS

Six dose-response curve experiments were conducted, one in BOD, another in a greenhouse and the third one in the field, for each reacting weed species that were biotypes for black picker (*Bidens pilosa*) and horseweed (*Conyza sumatrensis*). The experiments were conducted with a completely randomized design with four replicates.

The BOD experiments were conducted with Petri dishes over filter paper, where one dish, with 100 seeds of each species, represented one replica. In each Petri dish, 5 mL of a solution containing saflufenacil were applied, equivalent to the Heat® doses of: 0.0; 0.071; 0.143; 0.286; 0.571; 1.143; 1.429; 2.143 and 2.857 g ha^{-1}. The volume of five milliliters was defined in previous studies and was shown to be sufficient for the germination of the seeds without the need to refill the water up to 14 days after the beginning of the study. The control treatment received only deionized water. The dishes with the respective treatments were maintained at a temperature of 25 °C and within a photoperiod of 12 hours.

In the greenhouse, the experiments were installed in 0.5 L vases, filled with the turf and vermiculite mix (1:1). The black picker and horseweed seeds were sown in each respective vase. After emergence, the plants were thinned, leaving two seedlings per vase. The Heat herbicide was applied when the plants had 4-6 leaves, at the following doses: 0.0; 0.25; 0.5; 1.0; 2.0; 4.0; 6.0; 8.0; and 12 g ha^{-1}. At the time of the application of the treatments in the field, the relative humidity was 78%, and the temperature, 28 °C.

In the field, the experiments were conducted in 3 x 5 m plots (15 m^2), infested with 32 and 16 plants m^2 of black picker and horseweed, respectively, at a stage of 3-6 leaves. The applied doses of Heat® were: 0.0, 10; 20; 30; 40; 50; 60; 70; and 80 g ha^{-1}. At the time of the application of the treatments in the field, the relative humidity was 82%, the temperature, 26 °C, and the wind, 5 km h^{-1}.

In the greenhouse and in the field, the treatments were applied with a precision back sprayer, pressurized with CO_2, equipped fan-type spray nozzles DG 110.02 and spray solution volume of 150 L ha^{-1}. In all experiments, the percentage control was evaluated up to 14 days after treatment (DAT), assigning percentage scores from zero to 100%, in which zero represented lack of control and 100 the complete death of the plants. The results were subjected to the Shapiro-Wilk normality test and to the analysis of variance (ANOVA) ($p \leq 0.05$). When they were significant, the logistic sigmoidal regression model suggested by Seefeldt et al. (1995) was adjusted to the data:

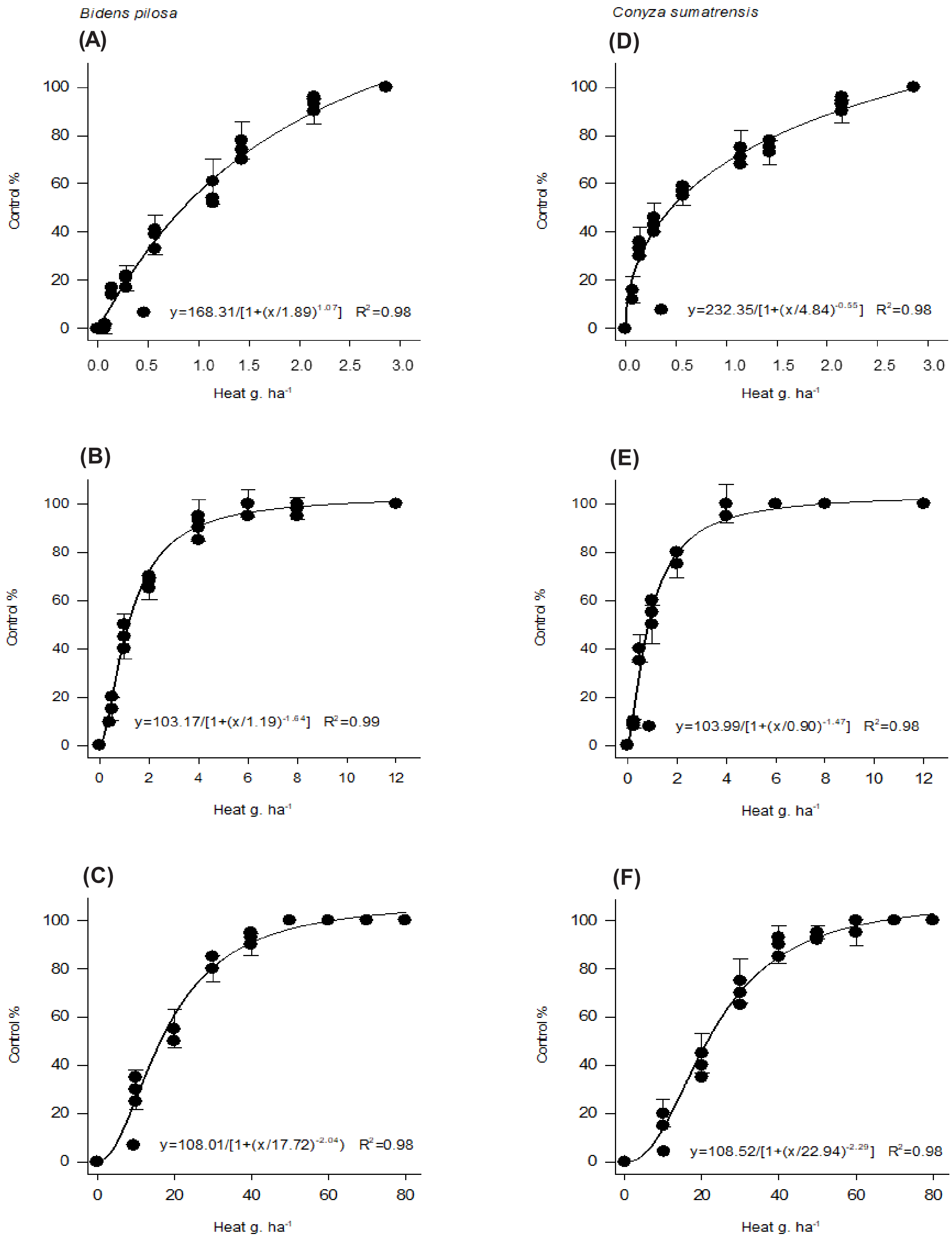
$$y = a / [1 + (x / x_0)^b]$$

where: y: control percentage; x: herbicide dose; *a*: difference between the maximal and minimal point of the curve; x_0: dose that offers 50% of the response variable; and *b*: curve declivity.

Then, Pearson's linear correlation was conducted (r) ($p \leq 0.05$ to the t test) across the variables. Pearson's correlation coefficient is a measurement of the linear relation degree between two quantitative variables. This coefficient ranges from values -1 and 1. Value 0 (zero) means that there is no linear relation; value 1 indicates a perfect linear relation; and value -1 also indicates a perfect, but inverse, linear relation, that is, when one of the variables increases, the other is reduced. The closer to 1 or -1, the stronger the linear association between the two variables; and for the analysis of the results, the magnitudes of the correlations defined by Dancey and Reidy (2006) were used, where: r = 0.10 up to 0.30 (weak); r = 0.40 up to 0.6 (moderate); and r = 0.70 up to 1 (strong). On Pearson's correlation analysis, the treatments with control over 90% were considered.

RESULTS AND DISCUSSION

The analysis of variance was significant to control black picker and horseweed subjected to doses of Heat®, observing a sigmoidal behavior throughout the doses and with adjustments of the model, shown by the determination coefficient (R^2) (Figure 1A-F). From the regression equations, the dose necessary to promote a 50% (C_{50}) and 90% (C_{90}) of the black picker and horseweed plants was calculated (Figure 1A-F; Table 1).



The points represent the mean values for the replicates, and the bars, the respective confidence intervals presented for each treatment with 95% of significance.

Figure 1 **-** Control (%) of *Bidens pilosa* and *Conyza sumatrensis* biotypes – graphs A and D, in BOD; B and E, in a greenhouse; and C and F, in the field – subjected to doses of the Heat® herbicide (saflufenacil) 14 days after the application of the treatments (DAT).

Table 1 **-** Heat® dose and control of 50% (C_{50}) and 90% (C_{90}) of the *Bidens pilosa* and *Conyza sumatrensis* population in response to the application of different doses of the saflufenacil herbicide, evaluated at 14 DAT

Control	*Bidens pilosa*			*Conyza sumatrensis*		
	BOD	Greenhouse	Field	BOD	Greenhouse	Field
C_{50}	0.85	1.16	16.5	0.47	0.86	21.5
C_{90}	2.1	3.9	39.5	2.1	3.3	46

Across the evaluated environment, a major variation of the dose was observed to reach C_{50} or C_{90}. When comparing the results in each environment, it was observed that the necessary dose to obtain 90% of control in the field for black picker was 10 and 19 times greater than the experiment in greenhouse and BOD, and for horseweed, it was 8 and 22 times greater, respectively. The differences across the control levels observed in the field may be a result of the environmental factors, such as soil humidity, radiation, temperature and relative air humidity, which may affect the characteristics of the plant, such as inducing the development of a thicker cuticle, trichomes and deeper root system, and the efficiency of the herbicide, since in BOD and in the greenhouse there is a greater control over these factors (Werlang et al., 2005). These results show the importance of the environmental conditions to the quality of the application and the control results obtained with saflufenacil in the studied species. In addition to the environment, the development stage of the plants also influences the results, that is, the more developed the plants, the lower the control (Mellendorf et al., 2013). In this study, the differences observed in the control between the greenhouse tests and those in the field are attributed to environmental effects, considering that the plants were at similar development stages in both environments.

The results of different tests were compared using Pearson's correlation, considering the control over 90% (Table 2). A significant correlation was not observed among the results obtained in the field and those in BOD and in the greenhouse for black picker and horseweed (Tables 3 and 4). On the other hand, for the studied species, there was a strong positive correlation among the results obtained by saflufenacil doses in BOD and in the greenhouse (Tables 3 and 4). The greatest correlation shown for black picker was 0.74 for Heat® doses over 2.143 g ha^{-1} in BOD, and 6 g ha^{-1} in the greenhouse, with control of 95 and 97.5%, respectively (Table 3). In relation to horseweed, the greatest correlation was 0.92 from a Heat® dose of 2.143 g ha^{-1} in BOD and 4 g ha^{-1} in the greenhouse, with control of 94 and 95%, respectively (Table 4). According to these results, it is possible to infer that greenhouse tests need a 2.8 times higher dose of Heat® to control black picker, and a 1.8 time higher dose of Heat® for horseweed, in comparison to the doses used in BOD.

The significant correlation indicates that the tests in BOD may replace those made in the greenhouse to identify the different sensitivity levels of black picker and horseweed to Heat®; for other species, the model will need to be evaluated. The practical application of the results obtained in this study, in addition to the possibility of conducting a rapid test to identify sensitivity or resistance, involves the identification of the maximal necessary dose to control weed populations (regression equation). The maximal dose value necessary to control 100% of a population may also be used as dose indication for resistance case reports.

Table 2 **-** Heat® doses and control above 90% of *Bidens pilosa* and *Conyza sumatrensis*, for correlation analyses in tests conducted in BOD and in a greenhouse at 14 DAT

Especie	BOD		Greenhouse	
	Dose (g ha^{-1})	Control (%)	Dose (g ha^{-1})	Control (%)
Bidens pilosa	2.143	95.25	6	97.5
	2.857	100	12	100
Conyza sumatrensis	2.143	94.33	4	95
	2.867	100	6	100

Table 3 **-** Pearson's linear correlation for the control variable of *Bidens pilosa* among the dose-response curve studies with herbicide saflufenacil conducted in BOD, greenhouse and in the field

	BOD	Greenhouse	Field
BOD	-	0.7403*	0.4277^{ns}
Greenhouse		-	0.3666^{ns}
Field			-

^{ns} non-significant and, * significant at p≤0.05 according to the t test.

Table 4 **-** Pearson's linear correlation for the control variable of *Conyza sumatrensis* among the dose-response curve studies with herbicide saflufenacil conducted in BOD, greenhouse and in the field

	BOD	Greenhouse	Field
BOD	-	0.9196*	0.4277^{ns}
Greenhouse		-	0.1132^{ns}
Field			-

^{ns} non-significant and, * significant at p≤0.05 according to the t test.

In general, when the companies that own the herbicide molecules are recording the product dose necessary to control a certain weed, they consider control levels between 80 and 90%. However, at the time in which a product dose is registered to offer 90% of control, it is admitting that 10% of the population survives to that dose and, consequently, that there is naturally 10% of the population that requires a dose higher than the registered one. In that sense, it is worthy to point out that the weed that survives the herbicide dose above the one registered for the species is considered a resistant weed. Companies consider that the dosage definition studies involve a large number of experiments and high cost. Considering the results of this study, it is possible to use the rapid method in Petri dishes to define the maximal control dose, which may be established as the threshold in dose-response tests to identify potential resistant biotypes to herbicides, eliminating the identification of a resistant "false-positive". For such, it is necessary to validate the method for the different species, find the equivalent doses between BOD and greenhouse for each one of them and make the necessary extrapolations.

Thus, using this method allows the identification of the sensitivity level of weed biotypes that survived to the application of herbicides and to define the weed control concomitantly to the crop cycle, as long as there are seed of the species or as long as it is possible to collect them from the soil in the relevant area, assisting in the reduction of the dissemination and of the control costs, in addition to contributing to a reduction on the evolving resistance of weeds to herbicides.

Therefore, according to the objectives, based on the correlation value obtained, it is possible to quickly evaluate the sensitivity level of black picker and horseweed in tests made in Petri dishes in BOD with a significant correlation with the results from tests conducted in the greenhouse, obtaining the results within six days for BOD, which would be obtained in two or more months for the greenhouse. It is possible to infer that doses from 2.143 g ha^{1} of Heat® are strongly and positively correlated to doses of 6 and 4 g ha^{-1} of Heat® used in the greenhouse to control black picker and horseweed, respectively. Thus, there is an equivalent between the tests conducted in BOD and in the greenhouse, and the rapid method using Petri dishes in BOD is feasible.

REFERENCES

Agostinetto D, Vargas L. Resistência de plantas daninhas a herbicidas no Brasil. Pelotas: UFPel; 2014.

Boutsalis P. Syngenta quick-test: a rapid whole-plant test for herbicide resistance. Weed Technol. 2001;15:257-63.

Dancey C, Reidy J. Estatística sem matemática para psicologia: usando SPSS para Windows. Porto Alegre: Artmed; 2006.

Délye C. Unravelling the genetic bases of non-target-site-based resistance (NTSR) to herbicides: a major challenge for weed science in the forthcoming decade. Pest Manag Sci. 2013;69:176-87.

Dias MAN, Obara FEB, Arruda N, Cursi http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0006-87052015000300307 - aff01PR, Gonçalves NR, Christoffoleti PJ. Germination test as a fast method to detect glyphosate-resistant sourgrass. Bragantia. 2015;79(3):307-10.

Gazziero DLP, Christoffoleti PJ, Vargas L, Kruse ND, Galli AJB, Trezzi MM. Critérios para relatos oficiais estatísticos de biótipos de plantas daninhas resistentes a herbicidas. In: Agostinetto D, Vargas L, editores. Resistência de plantas daninhas a herbicidas no Brasil. Passo Fundo: Berthier; 2009. p.91-101.

Heap, I. The international survey of herbicide resistant weeds. [accessed at: 25 Sept. 2017]. Available on: http://www.weedscience.org.

Mellendorf TG, Young JM, Matthews J, Young BG. Influence of plant height and glyphosate on saflufenacil efficacy on glyphosate resistant horseweed (*Conyza canadensis*). Weed Technol. 2013;27(3):463-7.

Seefeldt SS, Jensen JE, Fuerst EP. Log-logistic analysis of herbicide dose-response relationships. Weed Technol. 1995;9:218-27.

Vidal RA, Lamego FPE, Trezzi MM. Diagnóstico da resistência aos herbicidas em plantas daninhas. Planta Daninha. 2006;24(3):597-604.

Werlang RC, Silva AA, Ferreira LR, Miranda GV. Efeito da chuva sobre a eficácia de diferentes formulações de glyphosate no controle de *Bidens pilosa.* Rev Bras Herb. 2005;4:25-38

Yuan JS, Tranel PJ, Stewart CN Jr. Non-target-site herbicide resistance: A family business. Trends Plant Sci. 2007;12(1):6-13.