CONTROL OF GLYPHOSATE-RESISTANT VOLUNTEER MAIZE USING ACCase INHIBITING HERBICIDES¹

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ABSTRAC – Controlling volunteer plants was aggravated due to the emergence of genetically modified glyphosate-resistant cultivars, requiring new technical recommendations. This work was developed with the objective of evaluating alternatives for chemical control of glyphosate-resistant maize infesting soybean crops, using ACCase inhibiting herbicides. Two experiments were carried out, one in Santa Cruz das Palmeiras (SP) and other in Não-Me-Toque (RS), Brazil. Volunteer maize plants were simulated with seeds of the DKB 390 YGRR2 hybrids, distributed between rows of soybean crops, at the density of 6 plants m⁻². Combinations of the herbicides clethodim (65 and 84 g ha⁻¹), sethoxydim (138 and 165 g ha⁻¹), and haloxyfop (36 and 45 g ha⁻¹) were applied at two phenological stages of the soybean crop [three (3T) and six (6T) trifoliate leaves], corresponding to the V4/V5 and V7/V8 stages of the volunteer maize, respectively, resulting in 14 treatments. A treatment without herbicide application and a treatment with manual weeding were included as check plots. Applying graminicides on maize plants at V4/V5 stage resulted in more consistent results and general efficacy. Soybean yield was lower when applying herbicide only on maize plants at V7/V8, in both locations. Therefore, management practices for glyphosate-resistant volunteer maize should be adopted early, on maize plants at the V4/V5 stage, mainly when the maize plants are from lost grains in the harvesting process. The use of the herbicides clethodim and haloxyfop results in consistent control efficacy of volunteer maize plants.

Keywords: Glycine max. Graminicides. Management. Tolerance. Zea mays.

CONTROLE DE MILHO VOLUNTÁRIO RESISTENTE AO GLYPHOSATE COM HERBICIDAS INIBIDORES DA ACCase

RESUMO - O controle das plantas voluntárias foi dificultado com o advento das culturas geneticamente modificadas para tolerância ao herbicida glyphosate, exigindo novas recomendações técnicas para manejo. Este trabalho foi desenvolvido com o objetivo de avaliar alternativas para controle químico de plantas voluntárias de milho resistentes ao glyphosate, infestantes na cultura da soja, por meio de herbicidas graminicidas inibidores da ACCase. Dois experimentos foram desenvolvidos em campo, sendo um em Santa Cruz das Palmeiras (SP) e outro em Não-Me-Toque (RS). Para representar as plantas voluntárias de milho, sementes do híbrido DKB 390 YGRR2 foram distribuídos nas entrelinhas da soja, na densidade de 6 plantas m⁻². Adotaram-se 14 tratamentos, resultantes da combinação dos herbicidas clethodim (65 e 84 g ha⁻¹), sethoxydim (138 e 165 g ha⁻¹) e haloxyfop (36 e 45 g ha⁻¹), com dois estádios fenológicos da cultura de soja (três (3T) e seis (6T) trifólios); estando as plantas de milho voluntário nos estádios V4/V5 e V7/V8, respectivamente. Foi adicionado um tratamento sem aplicação de herbicidas e uma testemunha capinada manualmente. A pulverização dos graminicidas sobre plantas de milho em V4/V5 promoveu melhor consistência dos resultados e eficácia geral. A produtividade da soja foi reduzida em ambas as localidades com as pulverizações realizadas somente em V7/V8. Assim sendo, medidas de controle do milho voluntário resistente ao glyphosate devem ser adotadas precocemente, sobretudo quando o milho for resultante de grãos perdidos no processo de colheita. Neste sentido, destaca-se a adoção dos herbicidas clethodim e haloxyfop por sua eficácia consistente.

Palavras-chave: Glycine max. Graminicidas. Manejo. Tolerância. Zea mays.

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¹Received for publication in 05/28/2017; accepted in 09/14/2018.

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INTRODUCTION

Brazil is a world leader in grain production, including maize (*Zea mays* L.) and soybean (*Glycine max* (L.) Merril). According to the Brazilian National Supply Company (CONAB, 2018), in the 2016/2017 crop season, 33.9 million hectares of soybean were harvested, with an estimated production of 114 million Mg; and 17.6 million hectares of maize were harvested, with a total production (first and second harvests) estimated in 97.8 million Mg.

The land use intensification in current productive environments has been characterized by the adoption of crop succession or rotation systems. The most adopted crop succession in Brazil is soybean-maize, with soybean grown as first crop and maize as a second crop in succession, covering expressive production areas (ARTUZI; CONTIERO, 2006; PETTER et al., 2015; LÓPEZ-OVEJERO et al., 2016a). In the 2016/2017 crop season, maize production as second crop reached 67.4 million Mg, representing an increase of 65.2% when compared to the previous crop season (2015/2016), which accounted for 40.8 million Mg (CONAB, 2018).

The soybean-maize succession has long been used in Brazil, with a high predominance in production systems. These crops are harvested mechanically in most areas, using harvesters of different efficiency levels, especially in those areas that use high technology. Thus, grain losses during the harvest always occur on a greater or lesser extent, and the grains left in the areas can infest subsequent crops (PUZZI, 1986; SOUZA et al., 2006). Therefore, the appearance of volunteer maize plants from grains of the previous crop in the subsequent crop is frequent, and they are responsible for losses due to weed competition (BECKETT; STOLLER, 1988; CHAHAL et al., 2014).

Loose grains or lost ears during the maize harvest can remain in the soil and present germination and emergence viability throughout the dry season (winter period). After the beginning of the rainy season and sowing of the subsequent crop, i.e., the first crop of the next crop season, these grains can establish as voluntary plants and, as well as weeds, they cause significant negative effects on crop yield due to the competition of the plants for water, light, and nutrient resources (LÓPEZ-OVEJERO et al., 2016a).

Volunteer maize plants in soybean crops compete for water, light, and nutrients, with emphasis on competition for light due to their greater size (MARQUARDT; KRUPKE; JOHNSON, 2012). In these conditions, soybean yield losses of up to 69.9% may occur, mainly when there is presence of ears, which provides successive and uneven germinations, making control measures difficult (LÓPEZ-OVEJERO et al., 2016a).

For a few years, glyphosate was an effective

alternative for the control of volunteer maize in resistant soybean crops to this herbicide (RR soybean). However, after the technology of glyphosate-resistant maize, this possibility was not feasible, requiring the application of herbicides of other mechanisms of action, with emphasis on the ACCase inhibitor graminicides (DEEN et al., 2006; SOLTANI; SHORPSHIRE; SIKKEMA, 2006; STECKEL; THOMPSON; HAYES, 2009; MARCA et al., 2015). Thus, herbicide-resistant volunteer maize infestation is currently common in crops that are resistant to the same herbicides and sown in succession or rotation (THOMAS et al., 2007; CLEWIS et al., 2008; STECKEL; THOMPSON; HAYES, 2009; PETTER et al., 2015).

Considering the various options of graminicides available commercially in Brazil, as well as the agrometeorological diversity of the country, there is still a lack of information on control programs of glyphosate-resistant volunteer maize infesting soybean crops. Therefore, this work was developed with the objective of evaluating alternatives for the chemical control of glyphosateresistant volunteer maize plants, when germinated in the soybean crop, through applications of ACCase inhibitor graminicides.

MATERIAL AND METHODS

Two independent field experiments were carried out with soybean crops in the 2010/2011 crop season; one in Não-Me-Toque, RS (NMT) and other in Santa Cruz das Palmeiras, SP (SCP), Brazil. The soybean crops were grown in an area with total weed control through glyphosate applications at presowing and post-emergence of the crops. The soybean cultivars used were BMX Titan RR, with sowing on October 08, 2010 (SCP), and NA4990RG, with sowing on December 09, 2010 (NMT).

The crops were grown with spacing of 0.50 m between the soybean rows, under adequately soil fertilization and phytosanitary conditions. The experiments were performed in a randomized block design, with 14 treatments and four replications, totaling 56 plots in each experiment (Table 1). The experimental plots consisted of six 5-meter soybean rows, totaling 15 m², with an evaluation area of 5 m². The conditions of competition of soybean with glyphosate-resistant volunteer maize were simulated by manually distribution of maize seeds of the DKB390YGRR2 hybrid between rows of soybean crops, at the density of 6 plants m⁻², during the soybean sowing.

The herbicides used as chemical treatments were clethodim (65 and 84 g ha⁻¹), sethoxydim (138 and 165 g ha⁻¹), and haloxyfop (36 and 45 g ha⁻¹). They were applied on soybean plants when they had three (3T) and six (6T) trifoliate leaves. A treatment without herbicide application and a treatment with

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manual weeding were included as check plots (Table 1). The herbicide rates were chosen considering the minimum and maximum recommendations described in the product labels. Maize plants at the V4/V5 and V7/V8 stages were found at the first and second herbicide applications, respectively.

The herbicides were applied using a CO₂pressurized backpack sprayer coupled a bar with four TT110.015 nozzles spaced 0.50 m apart, set to an application volume proportional to 120 L ha⁻¹. Mineral oil (0.5% v v⁻¹) was added to all herbicides. The effect of the competition of volunteer maize with soybean was isolated by kept all soybean plots free from weed competition, using two glyphosate applications at 720 g ha⁻¹ at 14 and 28 days after the soybean sowing, apart to the treatments.

 Table 1. Detail of the experimental treatments installed using different herbicides, in two application times, besides check plots without herbicide application and a fully weeded control.

Treatment	Herbicide	Rate (g a.i. ha ⁻¹)	Application time ¹		
1	clethodim ²	65	3Т		
2	clethodim	84	3Т		
3	sethoxydim ³	138	3Т		
4	sethoxydim	165	3Т		
5	haloxyfop ⁴	36	3Т		
6	haloxyfop	45	3Т		
7	clethodim	65	6T		
8	clethodim	84	6T		
9	sethoxydim	138	6T		
10	sethoxydim	165	6T		
11	haloxyfop	36	6T		
12	haloxyfop	45	6T		
13		Check plots without herbicide	e application		
14	Check plots with manual weeding				

 ${}^{1}3T$ = Soybean plants with 3 trifoliate leaves; 6T = Soybean plants with 6 trifoliate leaves; ${}^{2}Select$ 240 EC (240 g of clethodim L⁻¹); ${}^{3}Poast$ (184 g of sethoxydim L⁻¹); ${}^{4}Verdict^{*} R$ (120 g of haloxyfop L⁻¹).

The variables analyzed were: percentage of control of maize plants at 14, 21, and 28 days after application (DAA) of each herbicide, and soybean stand and yield (kg ha⁻¹). The percentage of control was evaluated considering 0% as absence of control, and 100% as dead plants (SBCPD, 1995). The soybean crop stand was evaluated in the two central rows when the soybean plants were at phenological maturity, when the plants of the evaluation area of the plots were harvested. The soybean grains, considering a grain moisture of 13%.

All data were subjected to the F test in the analysis of variance, with a minimum of 5% of significance, considering the sites as independent experiments. Then, when the means were significant, the data were subjected to the Scott-Knott mean clustering test ($P \le 0.05$).

RESULTS AND DISCUSSION

The phenological stage of volunteer maize plants at the time of the herbicide applications was a determinant factor for the herbicide efficacy in both experimental areas (Table 2). The herbicide applications on maize plants at the V4/V5 stage showed more consistent results and general efficacy; clethodim at both used rates and haloxyfop at 45 g ha ⁻¹ achieved maximum control in all evaluations in both experiments. The use of haloxyfop at 36 g ha⁻¹ also resulted in satisfactory control of volunteer maize plants, above 80%; however, this treatment was not similar to the best treatments at 14 days after application (DAA) of herbicides in the experiment of Santa Cruz das Palmeiras, SP (SCP) (Table 2).

In general, the herbicide efficacy was low when the herbicides were applied on maize plants at the V7/V8 stage (Table 2). No treatment reached efficacy higher than 80% in SCP when the herbicides were applied on maize plants at V7/V8, i.e., below the minimum control level for herbicides (FRANS et al., 1986). Control levels above 80% were found at 21 and 28 DAA in the experiment in Não-Me-Toque, RS (NMT), when applying clethodim (84 g ha⁻¹) and haloxyfop. However, these treatments had similar efficacy to the highest efficacy levels only at 28 DAA, with applications on volunteer maize plants at V4/V5 (Table 2).

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Treatments	Maize phenological stage	SCP ²			NMT ³		
$(g ha^{-1})$		14	21	28	14	21	28
T1. Clethodim – 65	V4/V5	98.5 a	100.0 a	100.0 a	99.3 a	100.0 a	100.0 a
T2. Clethodim – 84	V4/V5	96.5 a	100.0 a	100.0 a	99.5 a	100.0 a	100.0 a
T3. Sethoxydim – 138	V4/V5	81.5 c	82.0 b	82.0 b	82.5 b	85.0 b	81.3 b
T4. Sethoxy dim – 165	V4/V5	76.5 c	82.0 b	78.8 b	87.5 b	92.5 a	87.5 b
T5. Haloxy fop -36	V4/V5	88.0 b	92.5 a	93.8 a	98.3 a	99.5 a	100.0 a
T6. Haloxy fop -45	V4/V5	97.5 a	100.0 a	100.0 a	98.3 a	100.0 a	100.0 a
T7. Clethodim – 65	V7/V8	42.5 e	68.8 c	66.3 c	61.3 d	75.0 c	93.3 a
T8. Clethodim – 84	V7/V8	50.0 d	75.0 c	76.3 b	67.5 c	83.8 b	97.8 a
T9. Sethoxy dim – 138	V7/V8	12.5 f	21.3 d	18.8 d	41.3 e	46.3 d	66.3 c
T10. Sethoxydim – 165	V7/V8	12.5 f	21.3 d	21.3 d	36.3 e	51.3 d	82.5 b
T11. Haloxy fop – 36	V7/V8	52.5 d	73.8 c	73.8 b	60.0 d	80.0 c	95.8 a
T12. Haloxy fop -45	V7/V8	47.5 d	73.8 c	73.8 b	68.8 c	88.8 b	96.5 a
T13. Check plots without application		0.0 g	0.0 e	0.0 e	0.0 f	0.0 e	0.0 d
T14. Check plots with manual weeding		100.0 a	100.0 a	100.0 a	100.0 a	100.0 a	100.0 a
F _{treat}		220.58*	140.06*	186.84*	110.62*	116.05*	168.23*
CV (%)		7.74	7.87	6.92	7.99	6.74	4.78

Table 2. Percentage of control of volunteer maize between soybean crop rows at 14, 21, and 28 days after herbicide applications on plants at the V4/V5 and V7/V8 stages.

*F test significant at 1%; ¹Means followed by the same letter in the columns are similar by the Scott-Knott test at 5% probability; ²SCP = Santa Cruz das Palmeiras, SP, Brazil; ³NMT = Não-Me-Toque, RS, Brazil; CV = coefficient of variation.

Similarly, Costa et al. (2014) found that clethodim applications were efficient on maize hybrid at V5 stage, but unsatisfactory for plants at V8. Deen et al. (2006) evaluated the control of voluntary maize based on combined applications of glyphosate and graminicides and found more efficiency with the use of the herbicide quizalofop-pethyl, followed by clethodim and fenoxaprop-pethyl; the combined application of glyphosate, graminicides, and adjuvants did not affect the herbicide efficacy or soybean tolerance.

The herbicide application, absorption, translocation, and metabolism are factors that affect the expression of symptoms. The metabolism of xenobiotics is related to the cytochrome P450 enzyme complex and to the enzyme glutathione-Stransferase, mainly to P450 for ACCase inhibitors. Thus, more developed plants may have a more complete and active enzymatic apparatus, allowing them to decompose more active ingredient with less expression of symptoms (CARVALHO et al., 2009; DIAS; CARVALHO; CHRISTOFFOLETI, 2013). Thus, as maize plants develop, they become more capable of decomposing herbicides and consequently, more tolerant to herbicide application.

Soltani, Shropshire and Sikkema (2006) found satisfactory results with applications of clethodim, fenoxaprop-p-ethyl, fluazifop-p-butyl, and quizalofop-p-ethyl to control volunteer maize plants. Furthermore, some studies found adequate control of glyphosate-resistant volunteer maize for the herbicide haloxyfop applied alone or in combination with glyphosate, 2,4-D, or fluroxypir (COSTA et al, 2014; PETTER et al., 2015).

The soybean crop stand and yield in SCP (Table 3) were higher when the graminicides were applied early, i.e., when the volunteer maize plants

were at the V4/V5 stage and the soybean plants had three trifoliate leaves (3T). The early application of graminicides ensured the full crop stand and yield, showing similar values to those of check plots (manual weeding) throughout this experiment. The presence of volunteer maize plants throughout the soybean crop cycle resulted in losses of 84.6% in crop yield in SCP (Table 3).

The soybean plants of the check plots with manual weeding in NMT presented an adequate yield, reaching 4,445.6 kg ha⁻¹. This yield was also reached only by plants under treatments with clethodim (84 g ha⁻¹) and haloxyfop (36 g ha⁻¹) applied on maize plants at V4/V5, showing correlation between the volunteer maize control and soybean yield. The treatments had no effect on the soybean stand in NMT (Table 3).

Marquardt and Johnson (2013) also found full soybean crop yield when applied clethodim (79 g ha⁻¹), but combined with glyphosate (840 g ha⁻¹). The selectivity of ACCase inhibitors for soybean crops is known; it is due to enzymatic insensitivity, since dicotyledonous plants have herbicide-insensitive enzyme isoforms (SASAKI; NAGANO, 2004; LÓPEZ-OVEJERO et al., 2016b). Therefore, losses are due to competition with volunteer plants. According to Beckett and Stoller (1988), soybean yields were 2%, 6%, 12%, 19%, and 27% lower due to the presence of volunteer maize plants when herbicides were applied at 2, 4, 6, 8, and 10 weeks after the soybean emergence, respectively. The use of graminicides is an alternative for the control of glyphosate-resistant weed biotypes, with emphasis on Lolium multiflorum and Digitaria insularis (MACIEL et al., 2013, COSTA et al., 2014).

Table 3. Soybean crop stand and yield (kg ha⁻¹) in two areas with infestation of glyphosate-resistant volunteer maize plants subjected to chemical control when the soybean plants were at the 3T (three trifoliate leaves) or 6T (6 trifoliate leaves) stages.

Treatments (g ha ⁻¹)	Soybean stage at	SCP^2		NMT ³	
Treatments (gila)	application	Stand	Yield kg ha ⁻¹	Stand	Yield kg ha ⁻¹
T1. Clethodim – 65	3T	91 a	3529.1 a	102	4270.7 b
T2. Clethodim – 84	3T	95 a	3571.4 a	117	4661.2 a
T3. Sethoxy dim – 138	3Т	101 a	3448.2 a	116	4098.5 b
T4. Sethoxy dim – 165	3Т	100 a	3160.6 a	114	4167.9 b
T5. Haloxy fop – 36	3T	93 a	3765.2 a	115	4684.6 a
T6. Haloxy fop -45	3T	100 a	3424.3 a	109	4306.2 b
T7. Clethodim – 65	6T	87 b	1523.3 b	95	3494.5 c
T8. Clethodim – 84	6T	82 b	1974.3 b	103	3926.3 b
T9. Sethoxydim – 138	6T	84 b	600.3 d	105	3295.1 c
T10. Sethoxydim – 165	6T	70 c	1159.6 c	98	3383.7 c
T11. Haloxy fop – 36	6T	74 c	1614.8 b	111	3508.6 c
T12. Haloxy fop – 45	6T	85 b	2089.6 b	107	3443.0 c
T13. Check plots without application		86 b	500.5 d	100	1079.7 d
T14. Check plots with manual weeding		105 a	3673.9 a	106	4445.6 a
F _{Treat}		8.837**	33.799**	1.437 ^{NS}	27.728**
CV (%)		7.74	17.06	10.76	9.18

*F test significant at 1%; NS = F test not significant; 1 Means followed by the same letter in the columns are similar by the Scott-Knott test at 5% probability; 2 SCP = Santa Cruz das Palmeiras, SP, Brazil; 3 NMT = Não-Me-Toque, RS, Brazil; CV = coefficient of variation.

López-Ovejero et al. (2016a) evaluated the control of glyphosate-resistant maize from ear fragments using clethodim and found that soybean yield was maintained when using a single application of clethodim (108 g ha⁻¹) on the soybean plants at the stage of six trifoliate leaves (6T) or two applications of clethodim (108 g ha⁻¹ and 84 g ha⁻¹) at plants at 3T and 6T stages, respectively. Thus, different from infestation of maize plants from ears, delaying the control of maize plants from grains of previous harvests hinders soybean yield. This is explained by the uneven and, possibly, slower germination of grains in ears and by the competition between the plants.

Therefore, control of volunteer maize plants in soybean crops should begin during the maize crop cycle, with emphasis on the maize harvesting (LÓPEZ-OVEJERO et al., 2016a). According to Puzzi (1986), delaying mechanical harvesting after maize maturation causes a decrease in grain water content and, consequently, significant loss increases. Souza et al. (2006) found a total loss of 9% during the mechanized maize harvesting process, which may have been favored by the delay in the harvest. The harvest time in the present work was at approximately 30 days after plant maturation.

The results indicate that the control of glyphosate-resistant volunteer maize should be carried out early, when the maize plants are at the V4/V5 stage and soybean plants have three trifoliate leaves, mainly when maize plants are from grains lost during the harvesting process. Thus, the adoption of the herbicides clethodim and haloxyfop is recommended for their consistent effectiveness.

CONCLUSIONS

Early application of graminicides on glyphosate-resistant maize plants results in a higher control level. The herbicides clethodim and haloxyfop are adequate options for the control of glyphosate-resistant volunteer maize, mainly when they are applied on maize plants at the V4/V5 stage. The satisfactory effectiveness of these herbicides can ensure an adequate soybean stand and yield.

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