

INTERFERENCE OF VOLUNTEER CORN IN GLYPHOSATE RESISTANT SOYBEAN AND CHEMICAL CONTROL IN DIFFERENT PHENOLOGICAL STAGES

INTERFERÊNCIA DO MILHO VOLUNTÁRIO NA SOJA RESISTENTE AO GLIFOSATO E CONTROLE QUÍMICO EM DIFERENTES ESTÁDIOS FENOLÓGICOS

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ABSTRACT: The successive use of Roundup Ready crops may difficult the management of volunteer plants originated from seed losses during harvest. In soybean, volunteer corn plants can exhibit higher interference and cause reduce yield depending on their density. The aim of this study was to quantify the economic threshold level (ETL) in soybean as a function of the competition of volunteer corn and to evaluate the chemical control in different phenological stages of development. The ETL and chemical control experiments were conducted in the field, under completely randomized and randomized block designs with one and three replicates, respectively. The variables analyzed were yield and ETL as functions of the competition of different volunteer corn populations (control, 1, 2, 4, 6, 8, 10, 12, 16, 20, 24 and 32 plants m⁻²) and the chemical control with acetyl coenzyme-A carboxylase (ACCCase) inhibitor herbicides alone or mixed with glyphosate in different phenological stages of development (V₂-V₃, V₄-V₅ and V₆-V₈) that were evaluated at seven, 14 and 21 days after application (DAA). The results showed higher competitive potential of volunteer corn in which the presence of one plant m⁻² reduces the soybean yield in 17%. The ETL ranged from 0.14 to 0.78 plants m⁻² and the control of volunteer corn must be carried out in low populations. The use of ACCCase inhibitors herbicides alone or mixed with glyphosate demonstrated greater than 85% control in the V₂-V₃ phenological stage independent of the period evaluated. The effectiveness of all herbicides decreased with application delay with a control level above 87%, in the V₆-V₈ phenological stage, obtained only for fluazifop and haloxyfop herbicides alone or in mixed with glyphosate at 14 and 21 days after application.

KEYWORDS: Alternative herbicides. Competition. Economic threshold level. *Glycine max*. *Zea mays*.

INTRODUCTION

Corn (*Zea mays*) and soybean (*Glycine max*) are the main commercial crops in Brazilian agriculture (CONAB, 2017). The technological advances which occurred for these crops such as the use of glyphosate-resistant crops and the use of integrated agricultural practices constitute factors that provided flexibility, improved weed control, reduced costs and contributed to increased yield (PETTER et al., 2007; BENBROOK, 2016).

Roundup Ready[®] technology has enabled the selective use of glyphosate in resistant crops for post-emergence spray, simplifying weed management (PETTER et al., 2015). The development of resistance to this herbicide occurred by the insertion of CP4 strain from *Agrobacterium* sp. responsible for coding one variant of 5-enolpyruvylshikimate-3-phosphate synthase enzyme (EPSPs), causing insensitivity to the glyphosate herbicide (PADGETTE et al., 1996). For susceptible plants, the inhibition of EPSPs enzyme blocks the

biosynthesis of aromatic amino acids tryptophan, phenylalanine and tyrosine in plastids due structural change at the active site and competition with phosphoenolpyruvate (TAN et al., 2006).

In Brazil, the Roundup Ready corn and soybean are widely used as succession crops in most agricultural areas, resulting in the appearance of glyphosate-resistant volunteer plants from grains lost during harvest (PETTER et al., 2015). The occurrence of volunteer corn plants into soybean fields causes competition for nutrients, light and water, increases control costs owing to the use of alternative herbicides and reduces the yield (DEEN et al., 2006; MARQUARDT et al., 2012). Moreover, volunteer corn plants have early development and C₄ cycle allowing for a higher competitive ability compared to soybean, which depends on the relative time of emergence, the origin (individual plant or clump), and the population present in the field (CHADAL; JHALA, 2016), reducing the time to control and the period prior to interference (PPI).

The presence of volunteer corn plants within another glyphosate-resistant crop impairs the adoption of management strategies reducing the alternatives available for weed control (ALMS et al. 2016). However, the spray of acetyl coenzyme-A carboxylase (ACCCase) inhibitor herbicides alone or mixed with glyphosate can be utilized to control volunteer corn (OWEN, 2000) with a reduction of efficacy in function of the differential phenological stage of development (CHADAL et al., 2014). Thus, studies regarding the interference of volunteer corn plants in soybean crops and the effectiveness of alternative controls are important factors that can aid in decision-making processes, reducing costs and protect yield potential.

The aim of this study was to quantify the economic threshold level (ETL) in soybean as a function of the competition of volunteer corn and to evaluate the chemical control in different phenological stages of development.

MATERIAL AND METHODS

Economic threshold level

The field experiment was conducted during 2015 in a completely randomized design with one replicate in yellow red Argisol belonging to the Pelotas mapping unit (EMBRAPA, 2013). The soil amendment was performed based on soil analysis following the technical recommendations for soybean (EMBRAPA, 2013). The population of volunteer corn provided the necessary variance to perform the statistical analyzes by the hyperbolic model proposed by Cousens (1985) and ranged from control, 1, 2, 4, 6, 8, 10, 12, 16, 20, 24 and 32 plants m^{-2} . The volunteer corn plants were randomly sown in the plot and conducted without infestation of other weeds during the soybean cycle. The soybean cultivar used was NA 5909 Roundup Ready, using a row spacing of 40cm and population of 36 plants m^{-2} . The emergence of volunteer corn and soybean occurred four to five days after sowing and, the environmental conditions are shown in Figure 1.

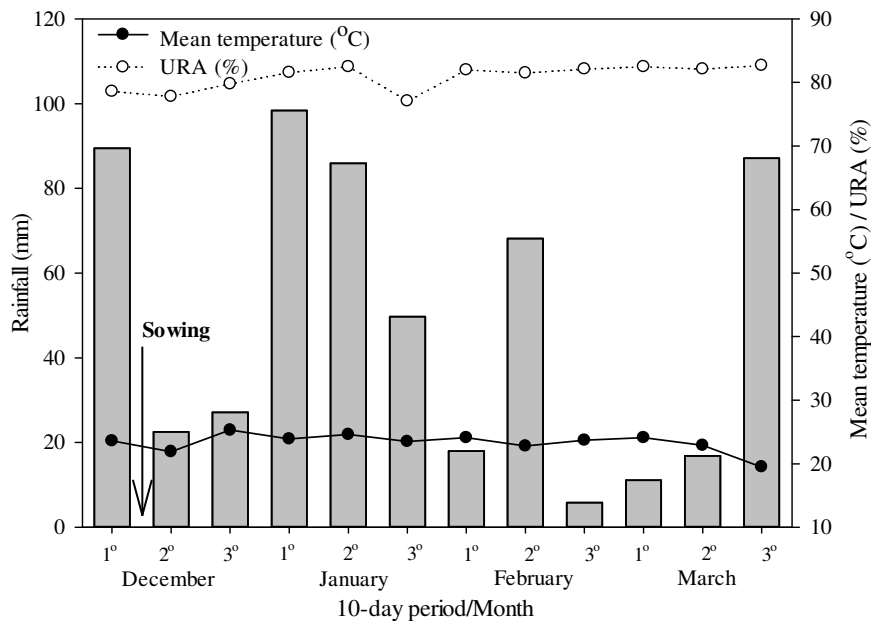


Figure 1. Environmental conditions during December 2014 to March 2015 for the experiment of soybean submitted to competition with volunteer corn.

Source: Estação Meteorológica da Embrapa Terras Baixas, Capão do Leão-RS.

The variables analyzed were soybean yield in competition with the volunteer corn populations and the ETL in function of yield expected, control cost, soybean price, and control efficiency. For the soybean yield, the total plot area ($2m^2$) was harvested and subjected to grain cleaning and drying at 13% moisture, and the yield ($kg ha^{-1}$) was obtained after weighing the grains on an analytical balance.

The percentage of soybean yield losses was calculated for each population level and compared with the control (without the presence of volunteer corn plants) according to the equation:

$$\text{Yield loss (\%)} = [(Ra - Rb) / Ra] \times 100$$

equation (1)

where: Ra and Rb : correspond to crop yield without and with the presence of volunteer corn plant, respectively.

The relationship between the percentage of soybean yield losses in each population of volunteer corn was calculated using the nonlinear regression model derived from the rectangular hyperbola (equation 2) proposed by Cousens (1985).

$$YL = [i * X / (1 + (i / a) * X)]$$

equation (2)

where: *YL*= yield loss (%); *X* = population of volunteer corn; *i* and *a* = yield loss (%) for each unit of the competing plant when the value of the variable approaches zero and when it tends to infinity, respectively.

The ETL used the parameter *i* obtained from equation 2 (COUSENS, 1985) and the equation adapted from Lindquist and Kropff (1996):

$$ETL = [Cc / (P * R * (i/100) * (H/100))]$$

equation (3)

where: *ETL*= economic threshold level (plants m⁻²); *Cc*= control cost (herbicide + application, in dollars ha⁻¹); *P*= soybean price (dollars kg⁻¹of the grains); *R*= soybean yield (kg ha⁻¹); *i*= soybean yield losses (%) per unit of volunteer corn plants when the population level approaches zero and; *H*= efficiency of the herbicide (%).

The ETL considered four reference values based on data from the last 10 years. The yield of soybean in Rio Grande do Sul State varied from 2000 to 3500 kg ha⁻¹ (CONAB, 2017), the quotations ranged from 10 to 40 dollars for 60 kg bags (BANCO CENTRAL O BRASIL, 2017). The control costs considered values between 20 to 80 dollars ha⁻¹ based on the glyphosate spray at a dosage rate of 1080 g ea. ha⁻¹ combined with an ACCase inhibitor herbicide added to the application

cost, and the efficiency of the herbicide was established at values of 70, 80, 90 and 100% of control.

Data adjustment to the model was performed with Proc Nlin from the statistical analysis system program (SAS, 1989) with 5% significance (*p*≤0.05) using the Gauss-Newton method, which enables to estimate the values of the parameters by the sum of the squared deviations from observations in relation to the adjusted values considering the lower residual mean square (RMS) (RATKOWSKY, 1983).

Chemical control of volunteer corn in different phenological stages

The field experiment was conducted during 2014/15 agricultural year in the experimental area of the Brazilian Agricultural Research Corporation localized in Passo Fundo-RS municipality on classified soil typical dystrophic Red Latosol belonging to the Passo Fundo Mapping Unit (EMBRAPA, 2013). The soil chemical analysis exhibited water pH=5.1; CTC_{pH7}= 6.7 cmolc dm⁻³; Al³⁺= 3.1 cmolc dm⁻³; Ca⁺²= 6.6 mmolc dm⁻³; Mg⁺²= 2.7 cmolc dm⁻³; K⁺= 0.23 mg dm⁻³; P= 9.9 g dm⁻³; clay= 61% and 1.5% of organic matter. The plots were arranged in a randomized block design with three replicates where each experimental unit was sown in a total area of 12 m² using a population of six plants m⁻¹ and rows spaced at 0.45 m from each other. The emergence of volunteer corn occurred four days after sowing and, the environment conditions during the development the volunteer crop are show in Figure 2.

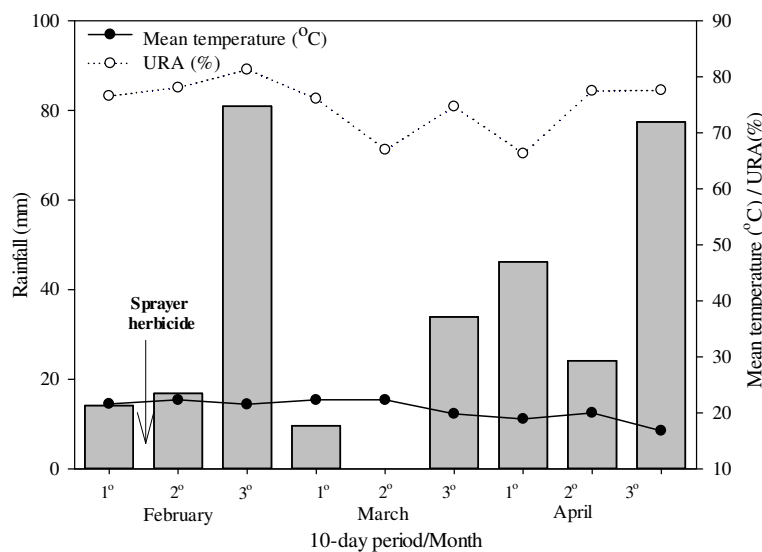


Figure 2. Environmental conditions during February to March 2015 for the experiment of volunteer corn control in different phenological stage.

Source: Estação Meteorológica da Embrapa Trigo, Passo Fundo-RS.

The treatments were arranged in factorial design (3x9+1) where factor A was the differential phenological stages of volunteer corn (V₂-V₃; V₄-V₅ and V₆-V₈) and, factor B was the ACCase inhibiting herbicides alone or mixed with glyphosate compared to the treatment without herbicide. The rates of each herbicide were glyphosate (1080 g ea. ha⁻¹), clethodim (84 g ai. ha⁻¹), sethoxydim (230 g ai. ha⁻¹), haloxyfop-p-methyl (49.8 g ai. ha⁻¹) and fluazifop-p-buthyl (150 g ai. ha⁻¹). The spray of the herbicides was performed with a CO₂ backpack sprayer equipped with four TeeJet TTI 110.15 spray nozzles, spaced at 0.5 m, and calibrated to deliver 120 L ha⁻¹ of spray volume. The meteorological conditions during the herbicide application were mean temperature of 25°C, 64% relative air humidity, and wind speeds of 1.2 m s⁻¹.

The control of volunteer corn plants was evaluated at seven, 14 and 21 days after application (DAA) using the percentage scale from 0% to 100%, where 0% means is no control and, 100% complete control of all weeds at the time of observation compared to the untreated control. The data obtained were analyzed for normality by Shapiro-Wilk test and then subjected to analysis of

variance and, the means were compared by Tukey's test ($p \leq 0.05$).

RESULTS AND DISCUSSION

Economic threshold level

The analysis of variance demonstrated significant effect of volunteer corn populations on soybean yield when in competition, with suitable adjustment of the data to the hyperbolic model proposed (Figure 3). Based on the *i* parameter (% yield loss of soybean per volunteer corn plant) was evidenced highly competitive potential in the NA5909 Roundup Ready soybean with yield reduction of 17% for each plant (Figure 3). Similar results have been observed for volunteer corn in soybean yield with losses from 15 to 20% per corn plant m⁻² in United States of America fields (MARQUARDT et al., 2012). Higher competitive ability occurs due to rapid initial growth and higher height of the corn plant compared to the soybean causing shading, physiological and morphological alterations, and yield reduction (CHADAL; JHALA, 2016).

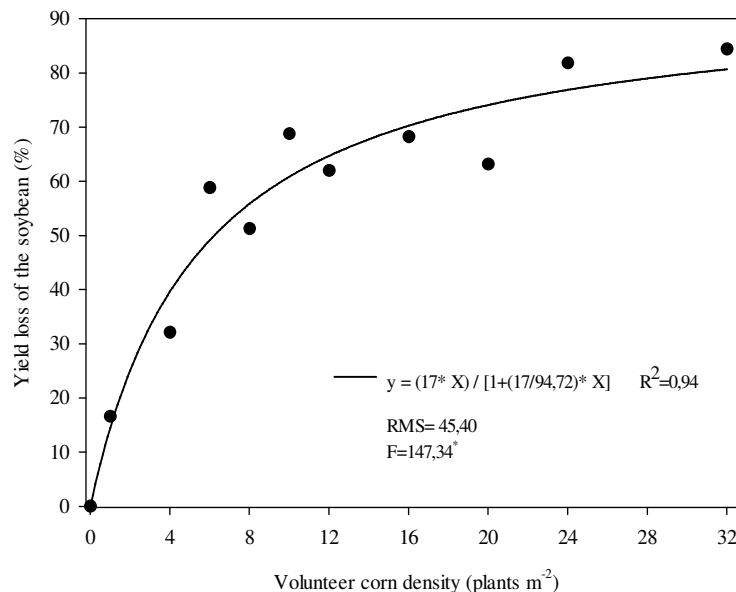


Figure 3. Yield loss (%) in NA5909 Roundup Ready soybean in function of the differential population competition of volunteer corn. UFPel/FAEM, Pelotas-RS, 2017. RMS: Residual mean square; *Significant at 5% probability.

For the *a* parameter of the model, the 94.72% value obtained indicates that the increase in the volunteer corn population may cause yield losses of approximately 100%. Similar results demonstrated that the increase in corn density can be considered a highly competitive weed which reduces the yield by more than 60%, depending on the population and the soybean cultivar used

(MARQUART et al., 2012; ALMS et al., 2016). In another study, volunteer corn plants in competition with soybean originating from seeds or clumps showed losses greater than 75% to 16 corn plants m⁻² and, greater than 90% when in competition with four clumps m⁻² (PIASECKI et al., 2018).

The suitable adjustment of the data to the hyperbolic model allows to evaluate the weed

competition in different crops and enables the calculation of the ETL (AGOSTINETTO et al., 2010). The ETL ranged from 0.14 to 0.78 plants m^{-2} depending on the parameter used, equivalent to 5% of the soybean yield based on the mean value of each parameter (Figure 4). Considering the control cost obtained, the ETL ranged from 0.14 to 0.56 plants m^{-2} in the lower and higher costs, respectively (Figure 4A). Studies evaluating the control cost for ETL determination of *Urochloa plantaginea* and *Ipomoea nil* in the common bean found that low

populations are enough to justify the control (VIDAL et al., 2010), which is similar to that found in the present study. Therefore, the greater the control cost, the higher the ETL of volunteer corn in the soybean crop. Although the ETL can generate information for control decision making, a lack of stability of the economic variables including the control costs and soybean price, might lead to changes in the ETL over time (KALSING; VIDAL, 2010).

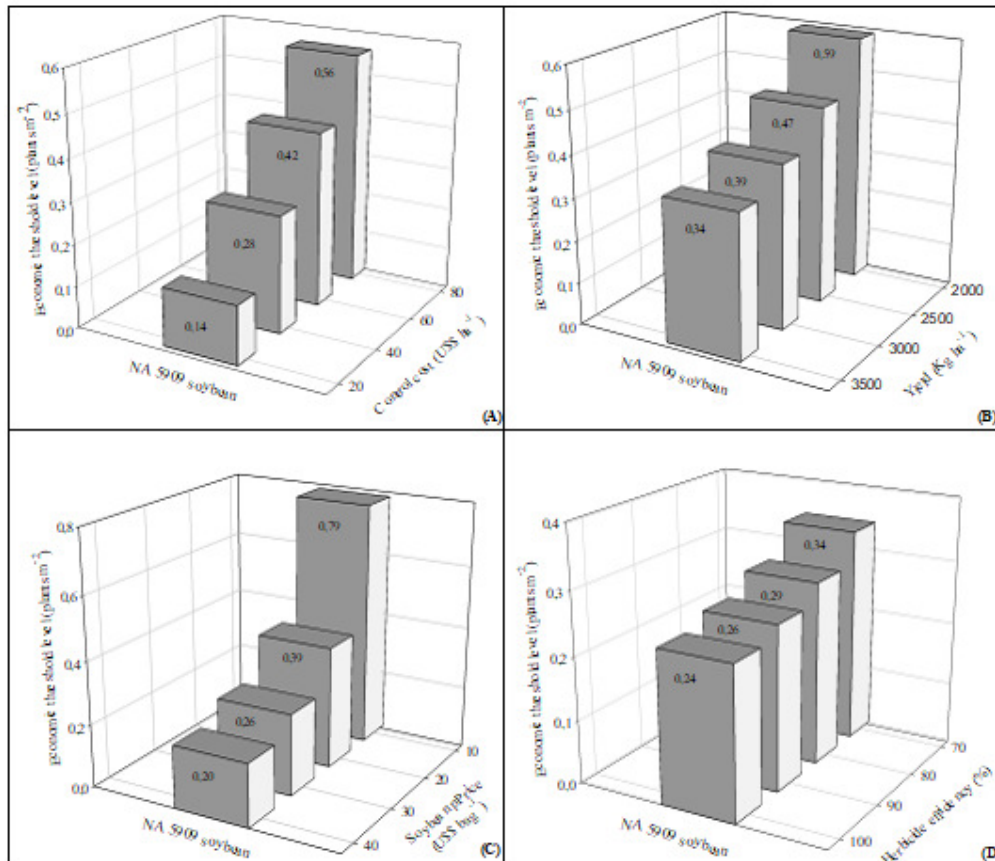


Figure 4. Economic threshold level of volunteer corn (plants m^{-2}) on NA 5909 Roundup Ready soybean in function of control cost (A), yield (B), soybean price (C) and herbicide efficiency (D) for corn management. UFPel/FAEM, Pelotas-RS, 2017.

The ETL values in function of the expected soybean yield ranged from 0.34 to 0.59 plants m^{-2} of volunteer corn (Figure 4B). In a previous study that analyzed the competition of soybean with the weed *Bidens pilosa*, the ETL ranged from 0.4 to 33 plants m^{-2} where the time of emergence of the weed compared to the crop and the yield also influencing the ETL (RIZZARDI et al., 2003). For the ETL as a function of soybean price, the control should be performed for a population level ranging from 0.2 to 0.78 plants m^{-2} (Figure 4C).

Although there is variation in the ETL, independent of the situation, the ETL was lower

than one plant m^{-2} in the present study, demonstrating higher competitive capacity of volunteer corn in reducing the soybean yield. In this way, the lower the price paid per soybean bag, the greater will be the number of volunteer corn plants needed to reach the ETL and make up for the control. Similar results were obtained in rice where the interference and ETL of *Echinochloa sp.* as a function of plant arrangement varied with the price paid, with the ETL being inversely proportional (AGOSTINETTO et al., 2010). Thus, higher crop yields and prices lead to lower ETL, supporting the adoption of control by farmers.

The ETL as a function of herbicide effectiveness demonstrated that control levels above 70% are required for populations of volunteer corn with less than 0.34 plants m^{-2} , demonstrating that the ETL is inversely proportional to herbicide effectiveness (Figure 4D). These results are in agreement with the ETL found in *Echinochloa sp.* where the values ranged from 0.20 to 2.3 plants m^{-2} depending on herbicide efficiency (GALON et al., 2007). Therefore, the adoption of control is economically viable when populations of volunteer

corn are lower than one plant m^{-2} in Roundup Ready soybean crops.

Chemical control of volunteer corn in different phenological stages

The results of chemical control demonstrated significant interaction between the herbicides treatments and different phenological stages of volunteer corn for all evaluated times, with higher effectiveness for the application at V₂-V₃ stage (Table 1).

Table 1. Control of volunteer corn in differential phenological stages at seven, 14 and 21 days after application with different ACCase inhibitor herbicides alone or mixed with glyphosate. Pelotas-RS, 2017.

Herbicide treatments	Rate g ai/ae ha ⁻¹	7 DAA					
		V ₂ -V ₃		V ₄ -V ₅		V ₆ -V ₈	
Control	-	0	Ca	0	Da	0	Ea
Glyphosate	1080	0	Ca	0	Da	0	Ea
Clethodim	84	98	Aa	83	Bb	22	Dc
Sethoxydim	230	82	Ba	65	Cb	27	Dc
Haloxifop	49.8	100	Aa	93	Aa	47	Cb
Fluazifop	150	95	Aa	77	Cb	55	Cc
Glyphosate+clethodim	1080+84	98	Aa	87	Ab	82	Ab
Glyphosate+sethoxymim	1080+230	97	Aa	73	Cb	58	Cc
Glyphosate+haloxifop	1080+49.8	100	Aa	90	Ab	72	Bc
Glyphosate+fluazifop	1080+150	100	Aa	80	Bb	80	Ab
V.C. (%)	-	9,4					
Herbicide treatments	Rate g ai/ae ha ⁻¹	14 DAA					
		V ₂ -V ₃		V ₄ -V ₅		V ₆ -V ₈	
Control	-	0	Ba	0	Ca	0	Ea
Glyphosate	1080	0	Ba	0	Ca	0	Ea
Clethodim	84	100	Aa	100	Aa	22	Db
Sethoxydim	230	98	Aa	60	Bb	47	Cc
Haloxifop	49.8	100	Aa	100	Aa	95	Aa
Fluazifop	150	100	Aa	100	Aa	87	Bb
Glyphosate+clethodim	1080+84	100	Aa	100	Aa	80	Bb
Glyphosate+sethoxymim	1080+230	100	Aa	100	Aa	68	Cb
Glyphosate+haloxifop	1080+49.8	100	Aa	100	Aa	92	Aa
Glyphosate+fluazifop	1080+150	100	Aa	100	Aa	91	Aa
V.C. (%)	-	5,9					
Herbicide treatments	Rate g ai/ae ha ⁻¹	21 DAA					
		V ₂ -V ₃		V ₄ -V ₅		V ₆ -V ₈	
Control	-	0	Ba	0	Ca	0	Da
Glyphosate	1080	0	Ba	0	Ca	0	Da
Clethodim	84	100	Aa	100	Aa	23	Cb
Sethoxydim	230	98	Aa	88	Bb	70	Bc
Haloxifop	49.8	100	Aa	100	Aa	98	Aa
Fluazifop	150	100	Aa	100	Aa	92	Ab
Glyphosate+clethodim	1080+84	100	Aa	100	Aa	100	Aa
Glyphosate+sethoxymim	1080+230	100	Aa	100	Aa	72	Bb
Glyphosate+haloxifop	1080+49.8	100	Aa	100	Aa	98	Aa
Glyphosate+fluazifop	1080+150	100	Aa	100	Aa	93	Aa
V.C. (%)	-	4,8					

*means followed by the same lowercase letter (line) and the same uppercase letter (column) do not differ by Tukey test ($p \leq 0.05$).

At seven DAA, the control was greater than 95% in V₂-V₃ stage for all herbicides, except for glyphosate and sethoxydim where the control was 0% and 82%, respectively (Table 1). Similar results were obtained for the application of haloxyfop and clethodim herbicides at the rate of 62 and 84 g ai. ha⁻¹, respectively, with control levels of volunteer corn being greater than 97% at seven DAA (MACIEL et al., 2013). For spraying during the V₄-V₅ phenological stage, only haloxyfop alone or mixed with glyphosate provided a control level above 90% and, for the V₆-V₈ stage, the best herbicides were the mixture of glyphosate with clethodim and haloxyfop where the control levels were 82% and 80%, respectively. The reduced control observed in advanced phenological stages, can be attributed to a greater amount of wax and cuticle thickness, hindering the penetration of herbicides (LIMA et al., 2011).

For the evaluation at 14 DAA, the results demonstrated that the herbicides clethodim, haloxyfop and fluzifop applied alone or mixed with glyphosate provided control levels above 98% for V₂-V₃ and V₄-V₅ phenological stages, except for the alone spraying of sethoxydim in V₄-V₅ stage where the control was only 60% (Table 1). Higher control in V₆-V₈ phenological stage was obtained with the application of haloxyfop alone reaching 95% of the control and for the mixtures of glyphosate with fluzifop and haloxyfop where the control was 91% and 92% at 14 DAA, respectively (Table 1). In corn, the inhibition of ACCase can occur in two catalytic subunits (ACCase I and II) with approximately 80% of the total activity of the enzyme located on subunit I that shows higher sensitivity to inhibition, in particular, to the aryloxyphenoxypropionates group (PRADO et al., 2000).

At 21 DAA, except for clethodim, all treatments showed control levels greater than 98% for applications until the V₅ phenological stage (Table 1). Similar results were obtained for the control of volunteer corn with ACCase enzyme inhibitors where the effectiveness was higher for application in initial stages and lower than five leaves (CHAHAL et al., 2014). Furthermore, a

strong reduction in the control levels of clethodim was evidenced for the V₆-V₈ phenological stage where the control was 23% and 72% when applied alone or mixed with glyphosate, respectively (Table 1). In the V₆ stage corn, the growing point of the plant is above the soil surface, which occurs simultaneously with the beginning of floral differentiation and increased lignification of the cellular wall. This might result in low uptake of the herbicide and reduced control of volunteer corn plants (MARQUARDT; JOHNSON, 2013). Similar results were reported for late applications in *Digitaria insularis* where the reduction of control was linked to the decreased absorption, increase of metabolism and detoxification of the molecule (CARVALHO et al., 2012). Moreover, the increase of herbicide effectiveness observed for fluzifop and haloxyfop may have occurred due to higher absorption and translocation, since these molecules are formulated as esters and may penetrate the cell more easily (NAYLOR, 2002) and/or, possible synergic effects when mixed with glyphosate.

CONCLUSIONS

The volunteer corn was highly competitive on NA 5909 Roundup Ready soybean with a yield reduction of 17% for each plant per m² while the ETL ranged from 0.14 to 0.78 plants m⁻² depending of the parameter used.

The control of ACCase inhibitor herbicides applied alone or mixed with glyphosate was higher at the V₂-V₃ phenological stage.

The reduction of control occurred for the late applications where control levels above 87% were only obtained for the herbicides fluzifop and haloxyfop alone or mixed with glyphosate for the V₆-V₈ phenological stage at 14 and 21 DAA.

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RESUMO: O uso sucessivo de culturas "Roundup Ready" pode dificultar o manejo de plantas voluntárias originadas de sementes perdidas durante a colheita. Na soja, plantas de milho voluntário podem apresentar elevada interferência e causar redução da produtividade dependendo da sua densidade. O objetivo do estudo foi quantificar o nível de dano econômico (NDE) na soja em função da competição de milho voluntário e avaliar o controle químico em diferentes estádios fenológicos de desenvolvimento. Os experimentos de NDE e controle químico foram conduzidos a campo, em delineamento inteiramente e blocos casualizados com uma e três repetições, respectivamente. As variáveis analisadas foram a produtividade e o NDE em função da competição das diferentes populações de milho voluntário (zero; um; dois; quatro; seis; oito; 10; 12; 16; 20; 24 e 32 plantas m⁻²) e o controle químico com herbicidas inibidores da enzima

acetil CoA carboxilase (ACCCase) isolados ou misturados com o glifosato em diferentes estádios fenológicos de desenvolvimento (V₂-V₃; V₄-V₅ e V₆-V₈) que foram avaliados aos sete, 14 e 21 dias após a aplicação (DAA). Os resultados demonstraram maior potencial competitivo do milho voluntário onde a presença de uma planta m⁻² reduziu a produtividade da soja em 17%. O NDE variou de 0,14 a 0,78 plantas m⁻² e o controle do milho voluntário deve ser realizado em populações baixas. O uso de herbicidas inibidores da ACCCase isolados ou misturados com o glifosato proporcionaram controle superior 85% no estágio fenológico V₂-V₃ independente do período avaliado. A eficácia de todos os herbicidas decresceu com o atraso da aplicação com um nível de controle acima de 87%, no estágio fenológico V₆-V₈, obtido apenas para os herbicidas fluzazifope e haloxifope isolado ou em mistura com o glifosato aos 14 e 21 dias após aplicação.

PALAVRAS-CHAVE: Herbicidas alternativos. Competição. Nível de dano. *Glycine max* *Zea mays*

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