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# Chemical control of glyphosate-resistant volunteer maize[1]

## *Controle químico de milho voluntário resistente ao herbicida glyphosate*

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**Abstract -** With the introduction in national agriculture of corn hybrids resistant to glyphosate herbicide, weed management, especially grasses, it was favored. However, volunteer maize plants coming from grains lost in mechanized harvesting can cause significant losses in crops subsequently sown, which has often been soybeans. Thus, the aim of this study was to evaluate the effect of different herbicides in controlling volunteer maize plants resistant to glyphosate herbicide. The experiment was conducted in the Brazilian municipality of Nova Xavantina, MT, between July and September 2014, in an area with a center pivot-type irrigation system. Grains of the F2 generation of AG 8061 VT PRO2® hybrid were used for assessments. The experimental design was randomized blocks with four replications. The treatments consisted in the application of thirteen different herbicides: haloxyfop-p-methyl 54 g a.i. ha$^{-1}$; tepraloxydim 87.5 g a.i. ha$^{-1}$; cyhalofop-butyl 247.5 g a.i. ha$^{-1}$; fluazifop-p-butyl 156.25 g a.i. ha$^{-1}$; sethoxydim 207 g a.i. ha$^{-1}$; fenoxaprop-p-ethyl 96.25 g a.i. ha$^{-1}$; chlorimuron-ethyl 15 g a.i. ha$^{-1}$; imazethapyr 100 g a.i. ha$^{-1}$; carfentrazone-ethyl 45 g a.i. ha$^{-1}$; fomesafen 237.5 g a.i. ha$^{-1}$; saflufenacil 35 g a.i. ha$^{-1}$; flumioxazin 60 g a.i. ha$^{-1}$ a and paraquat 450 g a.i. ha$^{-1}$, in the V3 growth stage of maize and a control without application. Herbicides paraquat, haloxyfop-p-methyl, tepraloxydim, cyhalofop-butyl, fluazifop-p-butyl, sethoxydim, fenoxaprop-p-ethyl and imazethapyr were efficient in controlling maize plants resistant to glyphosate herbicide. These herbicides did not show satisfactory efficiency: chlorimuron-ethyl, carfentrazone-ethyl, fomesafen, saflufenacil and flumioxazin.

**Keywords:** *Zea mays*; herbicides; weeds

**Resumo -** Com a introdução na agricultura nacional de híbridos de milho resistentes ao herbicida glyphosate, o manejo de plantas daninhas, principalmente, de gramíneas, foi favorecido. No entanto, plantas voluntárias de milho oriundas dos grãos perdidos na colheita mecanizada podem causar perdas significativas na cultura semeada subsequente, que frequentemente tem sido a soja. Desse modo, o objetivo do trabalho foi avaliar a eficiência de diferentes herbicidas no controle de plantas voluntárias de milho resistente ao herbicida glyphosate. O experimento foi conduzido no munícipio de Nova Xavantina–MT, entre os meses de julho e setembro de 2014, em área com sistema de irrigação do tipo pivô central. Foi utilizado para as avaliações grãos da geração F2 do híbrido AG 8061 VT PRO2®. O delineamento experimental utilizado foi o de blocos casualizados, com quatro repetições. Os tratamentos consistiram da aplicação de treze diferentes herbicidas, haloxyfop-p-methyl 54g i.a. ha$^{-1}$; tepraloxydim 87,5g i.a. ha$^{-1}$; cyhalofop-butyl 247,5g i.a ha$^{-1}$; fluazifop-p-butyl 156,25g i.a ha$^{-1}$; sethoxydim 207g i.a ha$^{-1}$;fenoxaprop-p-ethyl 96,25g i.a ha$^{-1}$; chlorimuron-ethyl 15g i.a ha$^{-1}$; imazethapyr 100g i.a ha$^{-1}$; carfentrazone-ethyl 45g i.a ha$^{-1}$;

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fomesafen 237,5g i.a $ha^{-1}$; saflufenacil 35g i.a $ha^{-1}$; flumioxazin 60g i.a $ha^{-1}$a e paraquat 450g i.a $ha^{-1}$, no estádio V3 do milho e uma testemunha sem aplicação. Os herbicidas paraquat, haloxyfop-p-methyl, tepraloxydim, cyhalofop-butyl, fluazifop-p-butyl, sethoxydim, fenoxaprop-p-ethyl e imazethapyr foram eficientes no controle de plantas de milho resistentes ao herbicida glyphosate. Não apresentaram eficiência satisfatória os herbicidas, chlorimuron-ethyl, carfentrazone-ethyl, fomesafen, saflufenacil e flumioxazin.

**Palavras-chaves:** *Zea mays*; herbicidas; plantas daninhas

## Introduction

Brazil is one of the largest world producers and exporters of maize, having produced, in the 2013/2014 harvest, 79.9 million tons (CONAB, 2014). Maize is grown in Brazil at two times during the harvest: in the spring/summer, called first crop, and the second one in the summer/fall (late harvest), representing 60% of the total maize area in Brazil according to data from CONAB [Companhia Nacional de Abastecimento (National Supply Company)] (2014). Most of the late harvest area is sown in dryland and in succession to a summer culture, often soybeans (IEA, 2014).

In maize production in Brazil, it is possible to highlight the use of genetically modified hybrids resistant to glyphosate herbicide. These plants are resistant to this herbicide due to a genetic modification performed by insertion of the CP4 gene, derived from a bacterium of the *Agrobacterium* genus, providing a change in the 5-enolpyruvylshikimate-3-phosphate (EPSP) synthase enzyme, which makes it insensitive to this herbicide (Madsen and Jensen, 1998; Trezzi et al., 2001). There is also the GA21 event, insertion of the EPSPs enzyme insensitive to glyphosate in maize plant by a biolistic process, making it tolerant to the herbicide (Spencer et al., 2000).

The emergence of maize volunteer plants resulting from grains that were lost in harvest after the maize harvest is common (Beckett et al, 1988). According to Tabile et al. (2008), the main causes that determine the losses in mechanical harvesting of grains are: poor soil preparation, inadequate planting timing, plants spacing and density, inadequate cultivars, occurrence of invasive plants, delayed harvest incorrect grain moisture, harvester displacement speed, lack of operator training, inadequate regulation, poor machinery conservation status and lack of losses monitoring.

The volunteer maize plants or *tiguera* may damage subsequent crops, resulting in significant losses (Albrecht Jr. et al., 2013). Marquardt et al. (2012) report losses in soybean yield ranging from 10 to 41%, with emergence from 0.5 to 16 volunteer maize plants per square meter, respectively.

Volunteer maize plants originated from materials with resistance to glyphosate have the complexity in their chemical control significantly increased. According to Maciel et al. (2013), in a rotation/succession system in which glyphosate-resistant maize appears as a volunteer plant, the combined application of glyphosate with herbicides inhibiting ACCase enzyme (Acetyl Coenzyme A carboxylase) may be an option for both the management predating direct seeding and in control in postemergence after installation of glyphosate-resistant soybean crops. However, results found by Barroso et al. (2010) show variability in the effective control of these herbicides according to the invasive species, that is, all herbicides of these groups do not have the same efficiency in the control of all grass species.

In the application of herbicides for the control of volunteer maize plants resistant to glyphosate, most likely other weed species will be present, requiring their control. In the event of infestation of broad-leaved plants with tolerance or resistance to glyphosate, such as dayflower (less often known as widow's tears)

(*Commelina* sp.), coat buttons (or tridax daisy) (*Tridax procumbens*), false buttonweed (*Spermacoce latofolia*) and horseweed (*Conyza* sp.), among others, the addition of ACCase inhibiting herbicides will not produce effects that are synergistic or complementary to glyphosate. Resulting from this, the evaluation of herbicides with latifolicide action on volunteer RR maize plants is critical in the search for options that promote the expansion of the weeds spectrum that can be controlled.

The aim of this study was to evaluate the effect of different herbicides in controlling volunteer maize plants resistant to glyphosate herbicide.

## Material and Methods

The research was conducted between July and September 2014, in an area with a center pivot-type irrigation system, on the farm Fazenda Roberta located in the Brazilian municipality of Nova Xavantina, MT, having as geographical coordinates: south latitude 15°05’25’’, west longitude 52°51’56’’ and altitude of 560 metros. The soil of the experimental area is classified as averaged textured eutrophic yellowish-red latosol.

Maize sowing (AG 8061 VT PRO2®) was mechanically performed and grains derived from the harvest of the first generation of this hybrid (F2) were used, simulating the grains derived from losses in mechanized harvesting. The spacing used was 0.50 m between rows, with a final population of 70,000 plants per hectare, plot size of 5 m x 3 m, and total area of 15 m². No fertilization was performed, and no kind of seed treatment with fungicides or insecticides either.

The experimental design was randomized blocks with four replications and 14 treatments, being 13 herbicides of several mechanisms of action: ACCase inhibitors (haloxyfop-p-methyl 54 g a.i. $ha^{-1}$; tepraloxydim 87.5 g a.i. $ha^{-1}$; cyhalofop-butyl 247.5 g a.i. $ha^{-1}$; fluazifop-p-butyl 156.25 g a.i. $ha^{-1}$; sethoxydim 207 g a.i. $ha^{-1}$ and fenoxaprop-p-ethyl 96.25 g a.i. $ha^{-1}$), ALS (acetolactate synthase enzyme) inhibitors (chlorimuron-ethyl 15 g a.i. $ha^{-1}$; imazethapyr 100 g a.i. $ha^{-1}$), PROTOX (protoporphyrinogen oxidase) inhibitors (carfentrazone-ethyl 45 g a.i. $ha^{-1}$; fomesafen 237.5 g a.i. $ha^{-1}$; saflufenacil 35 g a.i. $ha^{-1}$ and flumioxazin 60 g a.i. $ha^{-1}$), PSN (photosynthesis) inhibitor (paraquat 450 g a.i. $ha^{-1}$) and a control without application.

The herbicide applications were performed in the V3 maize growth stage using a knapsack sprayer pressurized by $CO_2$ with a 2 m boom with four 110-02 XR fan-type spray nozzle tips, spaced 0.50 m, with a spray volume equivalent to 150 liters per hectare.

Weather conditions monitored during the herbicides application were: average temperature: 29 ºC; average relative humidity of air: 60%; and average wind speed: 4 km/h.

The variables assessed were: plants height (cm) at 2, 7, 15, 30 and 45 DAA, randomly sampling 10 plants present in the floor area; control visually assessed at 2, 7, 15, 30 and 45 days after application (DAA) using a percent scale, where 100% mean all the dead plants and 0% means no kind of symptom; and shoot dry matter weight (g) at 45 DAA.

The data collected were tabulated and submitted to analysis of variance and analyzed by the F test ($p < 0.05$), and the averages of the significant variables were grouped by the criteria established by Scott-Knott at 5% probability.

## Results and Discussion

Analysis of variance showed significance for all variables evaluated at all times. In the first evaluation conducted two days after herbicide application (DAA), the herbicides inhibiting ACCase, haloxyfop-p-methyl, tepraloxydim, cyhalofop-butyl, fluazifop-p-butyl, sethoxydim, fenoxaprop-p-ethyl, and the herbicides inhibiting ALS, chlorimuron-ethyl and imazethapyr, caused a slight height reduction in plants resistant to glyphosate, significantly different from the control (Table 1).

In the next analysis (7 DAA), besides these herbicides reported, paraquat herbicide also provided a height reduction in maize plants resistant to glyphosate, and at 15 DAA it was not possible to quantify the height of maize plants resistant to glyphosate in the plots where paraquat was used due to the high degree of decomposition of plants. Also in this assessment, herbicides stood out: haloxyfop-p-methyl, tepraloxydim and fenoxaprop-p-ethyl, which lead to drastic decreases in the glyphosate-resistant maize plants height. The results of the fourth review (30 DAA) showed the impossibility of measuring the glyphosate-resistant maize plants height in plots that received treatments with haloxyfop-p-methyl, tepraloxydim, fluazifop-p-butyl, sethoxydim and fenoxaprop-p-ethyl.

**Table 1.** Glyphosate-resistant maize plants height after applying herbicides of different mechanisms of action in the V3 growth stage. Nova Xavantina, MT. 2014.

| Treatments | Plant height (cm) | | | | |
|---|---|---|---|---|---|
| | 2 DAA | 7 DAA | 15 DAA | 30 DAA | 45 DAA |
| Control | 52.00 a | 64.50 a | 85.25 a | 127.00 c | 143.25 a |
| Haloxyfop-p-methyl | 41.75 b | 52.00 b | 8.75 d | 0.00 f | 0.00 e |
| Tepraloxydim | 36.75 b | 42.00 b | 3.75 d | 0.00 f | 0.00 e |
| Cyhalofop-butyl | 39.25 b | 53.00 b | 48.25 c | 3.75 f | 0.00 e |
| Fluazifop-p-butyl | 44.00 b | 54.75 b | 50.50 c | 0.00 f | 0.00 e |
| Sethoxydim | 41.25 b | 58.50 b | 48.25 c | 0.00 f | 0.00 e |
| Fenoxaprop-p-ethyl | 40.50 b | 54.00 b | 3.75 d | 0.00 f | 0.00 e |
| Chlorimuron-ethyl | 43.25 b | 52.50 b | 68.00 b | 114.50 d | 108.50 c |
| Imazethapyr | 44.50 b | 56.50 b | 50.25 c | 52.25 e | 47.50 d |
| Carfentrazone-ethyl | 50.50 a | 75.50 a | 87.00 a | 135.50 b | 130.75 b |
| Fomesafen | 45.50 a | 69.50 a | 83.50 a | 129.75 c | 128.75 b |
| Saflufenacil | 48.25 a | 80.50 a | 92.00 a | 124.00 c | 136.50 a |
| Flumioxazin | 47.75 a | 71.75 a | 89.00 a | 145.75 a | 139.75 a |
| Paraquat | 47.25 a | 48.00 b | 0.00 d | 0.00 f | 0.00 e |
| CV (%) | 11.21 | 12.68 | 14.80 | 8.85 | 9.83 |

DAA = days after application. Means followed by the same letter do not differ by the Scott-Knott test ($p \leq 0.05$).

In the analysis of the last assessment of glyphosate-resistant maize plants height it was possible to see that all herbicides inhibiting ACCase showed maize growth inhibition in such a way as to make it impossible to obtain values for plant height. The ACCase inhibitors cause inhibition of this enzyme, resulting in blocking the lipids synthesis in susceptible plants (Burke et al., 2006). From this, harmful effects begin to compromise the cell wall formation, especially in growing regions (Schneider, 2011; Nalewaja et al., 1994). The symptoms observed in these treatments were growth stoppage and yellowing leaves, as described by DeFelice et al. (1989).

Among ALS inhibitor herbicides, imazethapyr was more effective in reducing glyphosate-resistant maize plants height. The reduction imposed by the action of this herbicide on glyphosate-resistant maize plants final height was approximately 67%, based on the height of plants present in the plots that had not received herbicide applications. None of PROTOX inhibiting herbicides (carfentrazone-ethyl, fomesafen, saflufenacil and flumioxazin) were effective in reducing glyphosate-resistant maize plants height, although carfentrazone-ethyl and fomesafen have provided significant reductions compared to the control, however small in magnitude. Inhibitors of PROTOX, an enzyme related to chlorophyll synthesis, cause accumulation of protoporphyrinogen IX in cell cytoplasm (Daylan et al., 1997) which, in the presence of light, react to produce singlet oxygen, a substance that degrades the cell membrane (Devine et al., 1993).

In assessing control held at 2 DAA, paraquat herbicide caused symptoms of high phytotoxicity to maize plants resistant to glyphosate (75%), confirming to be a fast-acting herbicide on maize plants in the V3 growth stage (Table 2). This herbicide is an acceptor of self-oxidized electrons in photosystem I which, in the presence of light, reacts, causing the depletion of NADPH (nicotinamide adenine dinucleotide phosphate-oxidase) and inhibition of carboxylation, in addition to yield, after interaction with oxygen, superoxides that promote the destruction of chloroplasts membranes by peroxidation (Fujii et al., 1990; Preston et al., 1991).

**Table 2.** Control and shoot dry matter weight (DMS) of maize plants after application of herbicides of different mechanisms of action in the V3 growth stage. Nova Xavantina, MT. 2014.

| Treatments | Control (%) | | | | | | | | | | DMS/plant (g) | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 2 DAA | | 7 DAA | | 15 DAA | | 30 DAA | | 45 DAA | | 45 DAA | |
| Control | 0.00 | c | 0.00 | g | 0.00 | e | 0.00 | e | 0.00 | e | 110.936 | b |
| Haloxyfop-p-methyl | 5.75 | b | 83.75 | b | 98.75 | a | 100.00 | a | 100.00 | a | 8.785 | f |
| Tepraloxydim | 7.25 | b | 63.75 | c | 97.50 | a | 100.00 | a | 100.00 | a | 6.775 | f |
| Cyhalofop-butyl | 6.50 | b | 52.50 | d | 87.50 | b | 95.00 | a | 100.00 | a | 9.533 | f |
| Fluazifop-p-butyl | 9.50 | b | 60.00 | d | 95.00 | a | 100.00 | a | 100.00 | a | 1.058 | f |
| Sethoxydim | 11.25 | b | 77.50 | b | 95.00 | a | 98.75 | a | 100.00 | a | 9.576 | f |
| Fenoxaprop-p-ethyl | 9.75 | b | 71.25 | c | 97.50 | a | 100.00 | a | 100.00 | a | 8.956 | f |
| Chlorimuron-ethyl | 0.75 | c | 3.75 | g | 20.00 | d | 53.75 | c | 55.00 | c | 42.685 | e |
| Imazethapyr | 0.75 | c | 5.00 | g | 17.50 | d | 83.75 | b | 88.75 | b | 13.124 | f |
| Carfentrazone-ethyl | 7.75 | b | 13.75 | f | 15.00 | d | 6.25 | e | 7.50 | d | 97.747 | c |
| Fomesafen | 10.00 | b | 33.75 | e | 42.50 | c | 42.50 | d | 13.75 | d | 83.725 | d |
| Saflufenacil | 0.00 | c | 4.00 | g | 7.50 | e | 3.75 | e | 12.50 | d | 123.718 | a |
| Flumioxazin | 9.25 | b | 13.75 | f | 17.50 | d | 7.50 | e | 7.50 | d | 114.270 | b |
| Paraquat | 75.00 | a | 98.75 | a | 100.00 | a | 100.00 | a | 100.00 | a | 3.227 | f |
| CV (%) | 28.11 | | 13.82 | | 9.84 | | 8.46 | | 8.89 | | 14.81 | |

DAA = days after application. Means followed by the same letter do not differ by the Scott-Knott test ($p \leq 0.05$).

The other herbicides, except for chlorimuron-ethyl, imazethapyr and saflufenacil, promoted injuries to maize plants resistant to glyphosate that were mild but significant compared to the control. In the next analysis (7 DAA), symptoms in maize plants resistant to glyphosate herbicide that received paraquat herbicide continued to evolve, reaching 98.75% of control. Also in this assessment, herbicides haloxyfop-p-methyl and sethoxydim had control of maize plants resistant to glyphosate, 83.75 and 77.50% control, respectively. Paraquat herbicide leads to death of all glyphosate-resistant maize plants at 15 DAA, or 100% control. Effectiveness of graminicides haloxyfop-p-methyl, tepraloxydim, fluazifop-p-butyl, sethoxydim and fenoxaprop-p-ethyl has also stood out in this assessment. Besides paraquat, which had already shown full control of maize plant resistant to glyphosate at 15 DAA, other four herbicides, haloxyfop-p-methyl, tepraloxydim, fluazifop-p-butyl and fenoxaprop-p-ethyl, have also eliminated all glyphosate-resistant maize plants at 30 DAA (100% control). Maciel et al. (2013) have noted that haloxyfop-R herbicide in doses of 25, 50 and 62 g $ha^{-1}$ was effective in controlling maize plants resistant to glyphosate in the developmental stages of V5 and V7. Among PROTOX inhibitors, no herbicide has shown, up to this assessment (30 DAA), satisfactory control of maize plants resistant to glyphosate, and the higher control values were obtained from the application of fomesafen (42.50%).

The final control evaluation, conducted at 45 DAA (Table 2), revealed that, besides paraquat, all ACCase inhibitor herbicides promoted control of all maize plants resistant to glyphosate. No differences were observed in the control of maize resistant to glyphosate herbicides between the two groups of ACCase inhibitors, cyclohexanediones and riloxifenoxipropionates, only small differences in control speed between these herbicides.

Despite being classified at a level below these herbicides that had total control of maize plants resistant to glyphosate, imazethapyr herbicide promoted final control of 88.75%, enough, for example, for it to be registered with the MAPA [Ministério da Agricultura, Pecuária e Abastecimento (Ministry of Agriculture, Livestock and Supply)] for that type of use. The performance presented by imazethapyr was higher compared to the other herbicide of the same mechanism of action, chlorimuron-ethyl, which obtained final control of 55.0%, i.e., its single use in the evaluated dose was unsatisfactory. The ALS enzyme inhibitor herbicides caused, as main visual symptoms in maize plants resistant to glyphosate, growth stoppage and internerval chlorosis on the leaf edge.

The accumulation of shoots dry matter of maize plants resistant to glyphosate at 45 DAA dramatically varied according to the applied herbicide (Table 2).

Herbicides that promoted the greatest levels of reduction in dry matter weight of maize plants resistant to glyphosate were: haloxyfop-p-methyl, tepraloxydim, cyhalofop-butyl, fluazifop-p-butyl, sethoxydim, fenoxaprop-p-ethyl, imazethapyr and paraquat. These results confirm the report by Soares et al. (2010), who have observed an efficient control of glyphosate-resistant volunteer maize in V5/V6 in five different locations with the application of: clethodim (84; 108), sethoxydim (184; 230), tepraloxydim (80; 100), clethodim + fenoxaprop-p-ethyl (40+40; 50+50), fluazifop-p-butyl (125; 188), haloxyfop-methyl (50; 62), and all doses are presented in g a.i. $ha^{-1}$. Chlorimuron-ethyl, despite not being classified in this group of higher efficiency, promoted reduction in dry matter weight of maize plants resistant to glyphosate of about 61.5%, showing that it is a non-selective herbicide for maize, but that it does not have an effective control over this species. Reduced dry matter accumulation of maize plants resistant to glyphosate was also observed with the use of fomesafen and carfentrazone-ethyl, yet very mildly.

Despite paraquat herbicide having been effective in controlling maize plants resistant to glyphosate, it is not compatible in combination with glyphosate. This may limit its use for this purpose because, usually, at the time of desiccation management applications, other weed species are present in the area, which would induce the need to include glyphosate in the application. Moreover, paraquat herbicide has demonstrated efficacy in controlling horseweed biotypes resistant to glyphosate herbicide and common in agricultural areas in the period where desiccation takes place in pre-seeding. Another point to be observed is that paraquat herbicide has no long-distance translocation (systematism), which could affect its action if the maize plants are in the later stages of growth. If the maize plants emerge during the soybean crop cycle, paraquat can not be used either because it has no selectivity to this crop. Thus, other herbicides such as ACCase inhibitors will be extremely important in the areas of production of maize and soybeans resistant to glyphosate, especially when there is the presence of sourgrass (*Digitaria insularis*) biotypes resistant to glyphosate herbicide. On the other hand, if there is the presence of broad-leaf plants tolerant to glyphosate along the volunteer maize plants, the addition of imazethapyr herbicide may provide greater gains in overall control of weeds.

## Conclusions

Herbicides paraquat, haloxyfop-p-methyl, tepraloxydim, cyhalofop-butyl, fluazifop-p-butyl, sethoxydim, fenoxaprop-p-ethyl and imazethapyr are efficient in controlling maize plants resistant to glyphosate, applied up to the V3 growth stage.

As for herbicides chlorimuron-ethyl, carfentrazone-ethyl, fomesafen, saflufenacil and flumioxazin, they have no efficient control of maize plants resistant to glyphosate in the V3 growth stage.

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