ISSN: 2230-9926

Available online at http://www.journalijdr.com

IJDR

*International Journal of Development Research*
*Vol. 09, Issue, 09, pp. 29868-29873, September, 2019*

REVIEW ARTICLE OPEN ACCESS

# HERBICIDE SELECTIVITY IN THE FORAGE PALM

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**ARTICLE INFO**

***Article History:***
Received 20th June, 2019
Received in revised form
13th July, 2019
Accepted 17th August, 2019
Published online 30th September, 2019

***Key Words:***
Chemical control; Weeds;
Nopalea cochenillifera;
Opuntia cochenillifera.

**ABSTRACT**

The objective of the present study was to assess the selectivity of five herbicides and their potential pre-emergent control over weeds in the forage palm crops. A completely randomized design was used to evaluate selectivity in triple factorial (2 x 5 x 7) considering varieties, herbicides and seasons assessed, respectively. In turn, the experiment to evaluate weed control was conducted with a completely randomized design with five treatments and one control with eight replications, yielding a total of 48 units. The herbicides tebuthiuron, atrazine and flumioxazin were selective for the *Nopalea cochenillifera* and *Opuntia cochenillifera* varieties at the doses used. On the other hand, the herbicides oxyfluorfen and ametryn caused severe phytotoxicity in both forage palm varieties. Tebuthiuron, atrazine, flumioxazin, oxyfluorfen and ametryn were considered efficient, controlling over 90% of the species present in the experiment. The efficacy of herbicides varies according with the infesting community, the age of the plant and the dose of the active ingredient applied.



**Citation: José Alex do Nascimento Silva, Jorge Luiz Xavier Lins Cunha *et al*. 2019.** "Herbicide selectivity in the forage palm", *International Journal of Development Research*, 09, (09), 29868-29873.

## INTRODUCTION

The forage palm originated from the central region of Mexico and spread across the world with colonization, having an important role as a food source for ruminants especially in semiarid conditions, which are common in the Brazilian Northeast region. This plant provides water, meeting greatly the needs of animals, in addition to representing a green food for animal nutrition (Neto *et al*., 2016). The high productive potential of this plant in conditions of high temperatures and low humidity can be attributed to its photosynthetic process. As a CAM (crassulacean acid metabolism) plant, the forage palm has the ability to fixate $CO_2$ during nighttime, avoiding a great loss of water due to transpiration. Thus, this plant is being used as an alternative to the adversities found in semiarid regions. In the Brazilian Northeast, the forage palm covers an area of more than 900 thousand hectares, which is the largest area covered by this cactus in the world (Donato *et al*., 2014).

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Despite its high adaptability in face of environmental conditions, the forage palm is similar to other crops in that its performance declines when in competition with weeds. This worsening in performance occurs especially due to the low cladode area index of some species, allowing weedy plants to surpass this crop regarding factors like water, light, nutrients and space (Kaur, S., Kaur, R., & Chauhan, 2018; De Santiago; Domínguez-Fernández; Cid & De Peña, 2018). Interferences caused by weedy plants originate from direct and indirect factors. Direct factors are represented by competition for natural resources, allelopathy and parasitism, while indirect factors include losses to the harvest, increased costs in treating the crop, and serving as a host for pests and diseases that damage the plants. Thus, knowledge on the weedy community allows the use of control techniques that avoid subsequent losses. However, there are no recommended herbicides in the control of these plants in forage palm crops (Hansen, Chatterjee, Gramig & Prischmann-Voldseth, 2018; Rao, Singh, Mahajan & Wani, 2018). Given the absence of molecules registered for forage palm, the objective of the present study was to assess the selectivity of five herbicides and their potential pre-emergent control over weeds.

## MATERIAL AND METHODS

First, the most used cultivars of forage palm in the Brazilian Northeast were identified, which were the cultivars *Opuntia cochenillifera* and *Nopalea cochenillifera* Salm Dyck. After identifying these cultivars, 20 cladodes of the *Opuntia cochenillifera* cultivar and 20 cladodes of the *Nopalea cochenillifera* Salm Dyck cultivar were planted. A single cladode was planted per vase with capacity of 5 kg. The substrate used was composed of three parts of soil and one of tanned manure. Both cultivars went through a 90-day period of initial development, during which only manual weeding was carried out weekly. After 90 days, five herbicide molecules were applied, which were: ametryn, atrazine, flumioxazin, oxyfluorfen and tebuthiuron. Two experiments were conducted to assess the selectivity of these molecules: one observing the selectivity of these molecules in the two cultivars used (*Opuntia* and *Nopalea*) and another experiment observing their weed control capacity (Table 1). Given the absence of recommended products for the control of weeds in forage palm crops, calculations were based on sugar-cane crops, applying the maximum dose of these molecules.

A backpack sprayer was used to apply the molecules with the aid of a spray hose with two 11002 flat-fan nozzles at a distance of 0.5 m between each other, spraying at a constant pressure (maintained using compressed $CO_2$) of 2.0 $kg.f^{2}$, yielding a carrier volume of 200 $L.ha^{-1}$. During application, experimental units were protected and separated to avoid carrier volume drift reaching other units. In addition to applying molecules to the two forage palm varieties, molecules were also applied to vases containing only the soil from the seed bank of the municipality of Jacaré dos Homens, state of Alagoas. This procedure was conducted using the same dosages in order to verify weed control. After applying the molecules, the plants were transported to a greenhouse where they were kept until the end of the experiment. During the study, the temperature in the greenhouse varied between 41.9 °C and 19 °C, while minimum and maximum humidity were 36.7 and 90%, respectively. Growth analyses were carried out, which included the variables: height, length, width, thickness and number of cladodes. Measurements were taken in millimeters using a ruler and a digital caliper. At the end of the study, the plants were gathered separately, identified and placed in a forced air oven at 65 °C for 72 hours to obtain dry matter. Digital images obtained throughout the experiment evaluations were used to assess phytotoxicity caused by herbicides.

Images were captured at a distance of 15 cm between the camera and the cladode analyzed and then processed using AFSoft software developed by the Embrapa Agricultural Instrumentation center. This software applies artificial intelligence to analyze leaf images based on pre-established color patterns, which allows the identification and quantification of areas affected by pests or damaged by diseases. Moreover, crop evolution can also be assessed by means of different patterns through this analysis. Regarding the experiment on weed control capacity by these molecules applied pre-emergence, the analysis included an evaluation of germination flow, quantification and identification of emerged seedlings, and observations on phytotoxic effects of the molecules on the plants. The experiment to assess the selectivity of herbicides applied to the forage palm was conducted with a completely randomized design with a triple factorial system (2 x 5 x 7), which regarded varieties, herbicides and seasons evaluated, respectively. In turn, the experiment to assess weed control was carried out with a completely randomized design with five treatments and one control with eight replications, yielding a total of 48 units. After data was collected from both experiments, they were submitted to a variance analysis using Sisvar software. Mean values were compared through Tukey's test at 5% probability.

## RESULTS

According to the variance analysis, there was no triple interaction among the factors evaluated nor was there a double interaction between herbicides and cultivars and season evaluated and cultivars. However, there was a significant interaction with 1% probability between herbicides and season evaluated, demonstrating that the molecules used can damage the forage palm according to the evolution period assessed (Table 2).

**Table 1. List of pre-emergence treatments applied to the forage palm**

| Treatment | Action mechanism | Dose |
|---|---|---|
| Tebuthiuron | Photosystem II inhib. | 2.4 $kg.a.i./ha^{-1}$ (1) |
| Atrazine | Photosystem II inhib. | 2.5 $kg.a.i./ha^{-1}$ |
| Ametryn | Photosystem II inhib. | 3.0 $kg.a.i./ha^{-1}$ |
| Oxyfluorfen | Protox inhib. | 480 $g.a.i./ha^{-1}$ |
| Flumioxazin | Protox inhib. | 125 $g.a.i./ha^{-1}$ |

Note. [1] Active ingredient per hectare

Given the interaction between herbicides and the seasons evaluated, each variety was analyzed separately. The damages observed in the *Opuntia cochenillifera* and *Nopalea cochenillifera* Salm Dyck varieties began eight days after applying herbicides, with a growing evolution over the period assessed for both varieties studied (Table 3 an 4). The highest levels of damage occurred 28 days after applying herbicides for both varieties studied, with ametryn and oxyfluorfen triggering symptoms of chlorosis and necrosis, respectively. During this same evaluation period, tebuthiuron, atrazine and flumioxazin did not cause damages to the plants analyzed. Oxyfluorfen is a protoporphyrinogen oxidase (protox) inhibitor, which leads to porphyrin deregulation in plants due to abnormal accumulation in cells, causing their death. This molecule presents a high residual effect in the soil and, associated with slow plant metabolism, can present both pre and post-emergence effects (Kim *et al*., 2014; Mantzos *et al*., 2014).

The pre-emergence activity of oxyfluorfen is conditioned to soil moisture during application. Permanence within the soil is high and depends on the dose applied. High doses of this molecule can cause the inhibition of electron transport and ATP synthesis (Phung & Jung, 2015; Wu *et al*., 2019). A dose of 480 $g.a.i./ha^{-1}$ of oxyfluorfen in the forage palm caused reddish-brown spots that evolved to necrosis in both varieties analyzed. Flumioxazin can be used either pre or post-emergence. This molecule is easily adsorbed in the soil and its main degradation pathway is through microbial activity, presenting low soil persistence and a half-life that can vary between 15 and 19 days. Flumioxazin is considered an important mechanism for the control of weeds because there are no known weedy plants that are resistant to this active ingredient (Bigot, Fontaine, Clément & Vaillant-Gaveau,

**Table 2. Mean square values and coefficient of variation (CV) obtained in the analysis of variance (ANOVA)**

| VS[1] | DF[2] | Mean square |
|---|---|---|
| Herbicides | 5 | 366.307** |
| Seasons | 6 | 47.822** |
| Cultivars | 1 | 0.008 ns |
| Herbicides x Seasons | 30 | 26.520** |
| Herbicides x Cultivars | 5 | 0.524ns |
| Seasons x Cultivars | 6 | 0.0165ns |
| Herbicides x Seasons x Cultivars | 30 | 0.071ns |
| Error | 252 | |
| CV(%) | 19.05 | |

Note. [1] Variation Source; [2] Degree of freedom; **Significant at 1% through Tukey's test; ns not significant.

**Table 3. Damages to the forage palm variety *Opuntia cochenillifera* at different evaluation moments after applying treatments containing herbicides. Rio Largo-AL, 2018**

| Treatment | Damage (days) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 4 | 8 | 12 | 16 | 20 | 24 | 28 |
| Tebuthiuron | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Atrazine | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Ametryn | 0.00 a | 0.00 a | 0.00 a | 52.4 b | 63.0 c | 64.7 b | 65.7 b |
| Oxyfluorfen | 0.00 a | 30.1 b | 30.6 b | 40.5 b | 44.4 b | 47.7 b | 51.0 b |
| Flumioxazin | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Control | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |

Note. In the columns, mean values followed by the same letters did not differ statistically by Tukey's test at 5% probability.

**Table 4. Damages to the forage palm variety *Nopalea cochenillifera* Salm Dyck at different evaluation moments after applying treatments containing herbicides. Rio Largo- AL, 2018**

| Treatment | Damage (days) | | | | | | |
|---|---|---|---|---|---|---|---|
| | 4 | 8 | 12 | 16 | 20 | 24 | 28 |
| Tebuthiuron | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Atrazine | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Ametryn | 0.00 a | 0.00 a | 0.00 a | 42.8 b | 54.1 b | 55.5 b | 58.8 b |
| Oxyfluorfen | 0.00 a | 26.6 b | 39.5 b | 46.7 b | 50.1 b | 50.7 b | 53.1 b |
| Flumioxazin | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |
| Control | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a |

Note. In the columns, mean values followed by the same letters did not differ statistically by Tukey's test at 5% probability.

**Table 5. Analysis of variance of the variables assessed in the *Opuntia cochenillifera* variety of the forage palm, Rio Largo- AL (2018)**

| Treatment | Height | Length | Thickness | Width | No. of cladodes | Dry matter |
|---|---|---|---|---|---|---|
| Tebuthiuron | 59.75 a | 26.69 ab | 15.52 a | 59.49 a | 3.25 a | 0.42 b |
| Atrazine | 65.25 a | 28.21 ab | 19.25 a | 62.65 a | 5.25 a | 0.42 b |
| Ametryn | 59.25 a | 17.01 b | 15.96 a | 55.29 a | 3.25 a | 0.33 b |
| Oxyfluorfen | 48.50 a | 24.74 ab | 14.52 a | 52.09 a | 5.75 a | 0.32 b |
| Flumioxazin | 51.00 a | 29.00 a | 14.39 a | 52.01 a | 6.25 a | 0.47 ab |
| Control | 71.50 a | 31.89 a | 19.84 a | 63.94 a | 7.25 a | 0.73 a |
| DMS | 23.26 | 11.56 | 7.57 | 31.75 | 4.36 | 0.30 |
| CV (%) | 17.10 | 19.17 | 19.88 | 24.07 | 36.78 | 29.53 |

Note. In the columns, mean values followed by the same letters did not differ statistically by Tukey's test at 5% probability; DMS = minimal significant difference; CV = coefficient of variation.

2007; Assunção *et al*., 2017: Arakawa, Otani, Iwashita & Yamazaki, 2017). Although flumioxazin and oxyfluorfen present non-systemic action and the same action mechanism, there was no kind of visual alteration in the cladodes when applying flumioxazin, maintaining the same pattern throughout the whole period evaluated. The difference in dose between these molecules may be one of the factors that led to damages to the plants, since maximum doses were used for both herbicides: 480 and 125 g.a.i./ha$^{-1}$ for molecules of oxyfluorfen and flumioxazin, respectively. Moreover, metabolization is one of the main factors that can trigger selectivity and it can act differently in face of each of these molecules, since oxyfluorfen is a diphenyl ether, while flumioxazin is a cyclohexenedicarboximide (Abe *et al*., 2018; Wu *et al*., 2019). Another factor that could have influenced the selectivity of these molecules is translocation within the plants. Since both herbicides present non-systemic action, the place of action becomes the contact area between the molecules and the plant and/or nearby areas (Park *et al*., 2018; Peragón & Amores-Escobar, 2018). Similar to the translocation of these molecules in the plant, the composition of inert ingredients in these herbicides can exercise great influence in selectivity given that they dilute the active ingredient, facilitating dispersion and penetration in the targeted organism (Mesnage, Defarge, Spiroux de Vendômois & Séralini, 2014). Although the forage palm presented high levels of necrosis, predominantly in the apex of the cladodes, which was where there was the most contact with oxyfluorfen molecules (51 and 53.1% for the *Opuntia cochenillifera* and *Nopalea cochenillifera* Salm Dyck varieties, respectively), the plant is able to recover from this damage, as observed from the unscathed emergence of new cladodes (table 5 and 6). Considering the herbicides that act as photosystem II inhibitors, a high level of chlorosis was observed when ametryn was applied to both varieties analyzed, with values of 65.7 and 58.8% for *Opuntia cochenillifera* and *Nopalea*

**Table 6. Analysis of variance of the variables assessed in the *Nopalea cochenillifera* Salm Dyck variety of the forage palm, Rio Largo-AL (2018)**

| Treatment | Height | Length | Thickness | Width | No. of cladodes | Dry matter |
|---|---|---|---|---|---|---|
| Tebuthiuron | 64.00 a | 26.36 a | 16.17 a | 75.72 a | 3.25 b | 0.52 a |
| Atrazine | 61.75 a | 27.35 a | 18.66 a | 79.01 a | 3.75 ab | 0.43 a |
| Ametryn | 53.75 a | 26.04 a | 15.80 a | 57.74 a | 2.75 b | 0.39 a |
| Oxyfluorfen | 58.00 a | 25.15 a | 14.87 a | 69.41 a | 2.75 b | 0.35 a |
| Flumioxazin | 60.25 a | 31.81 a | 16.37 a | 66.94 a | 3.50 ab | 0.48 a |
| Control | 63.25 a | 33.08 a | 17.67 a | 84.97 a | 6.50 a | 0.56 a |
| DMS | 32.63 | 14.43 | 10.96 | 34.36 | 3.35 | 0.39 |
| CV (%) | 23.60 | 22.19 | 28.74 | 20.68 | 38.85 | 36.72 |

Note. In the columns, mean values followed by the same letters did not differ statistically by Tukey's test at 5% probability; DMS = minimal significant difference; CV = coefficient of variation.

**Table 7. Weed control percentages according to the different treatments used on the forage palm. Rio Largo - AL, 2018**

| Treatment | *Merremia aegyptia* (L.) Urb. | *E. indica* (L.) Gaertn. | *Phyllanthus tenellus* Roxb. | *Bidens* sp. | *Dactyloctenium aegyptium* (L.) Willd |
|---|---|---|---|---|---|
| | | | 30 DAA[1] | | |
| Tebuthiuron | 100.0 a | 100.0 a | 100.0 a | 100.0 a | 100.0 a |
| Atrazine | 100.0 a | 100.0 a | 100.0 a | 100.0 a | 100.0 a |
| Ametryn | 100.0 a | 100.0 a | 94.00 a | 100.0 a | 100.0 a |
| Oxyfluorfen | 100.0 a | 100.0 a | 100.0 a | 100.0 a | 100.0 a |
| Flumioxazin | 94.00 a | 100.0 a | 100.0 a | 100.0 a | 100.0 a |
| Control | 0.0 b | 0.0 b | 0.0 b | 0.0 b | 0.0 b |

Note. [1] Days after application; in the columns, mean values followed by the same letters did not differ statistically by Tukey's test at 5% probability.

*cochenillifera* Salm Dyck, respectively. In turn, the molecules tebuthiuron and atrazine did not cause any damages, only darkened spots on the cladodes when they were applied. The difference of 0.5 kg.a.i./ha$^{-1}$ of ametryn in comparison with the other photosystem-inhibitor herbicides could have had an important role in the selectivity of these molecules in the cactus. This can be explained by the ability of the plant's cells to withstand certain amounts of compounds, eventually leading to an overflow when the amount surpasses this limit. Moreover, the high temperature in the greenhouse where the experiment was conducted may have contributed to the disruption of plasma membranes, increasing internal flow and allowing a greater number of molecules to enter the cell (Li *et al*., 2019; Mutlu *et al*., 2019). Following the same pattern as molecules that inhibit photosystem II, the composition of inert materials in the chemicals that inhibit protox also influences the selectivity of these molecules due to the dilution of the active ingredient, which facilitates penetration in the targeted organism (Guelfi *et al*., 2019). High levels of toxicity in these crops can lead to serious damages to development and biomass accumulation, thus triggering low yields and productivity. The low influence of the herbicides analyzed on the growth characteristics of the forage palm can be attributed to the plant's development stage. Despite its good adaptation to arid and semiarid regions, the cactus presents slow development. Thus, the influence of these molecules can gradually appear with the development of this crop (Pinheiro *et al*., 2014).

The main species of weeds that occurred in the experimental areas were *Eleusine indica* (L.) Gaertn., *Merremia aegyptia* (L.) Urb., *Phyllanthus tenellus* Roxb., *Bidens* sp. and *Dactyloctenium aegyptium* (L.) Willd. The most commonly occurring species in this study was *E. indica*. This highly competitive weed is a $C_4$ plant that presents quick development and is able to produce up to 40 thousand seeds per plant, which germinate with ease at temperatures alternating between 35 and 20 °C, common conditions in the Brazilian Northeast region (Chen *et al*., 2017; Zhang; Hall; McElroy; Lowe, & Goertzen, 2017). This species is more easily controlled when specimens are young, due to a thinner waxy cuticle and undifferentiated transport tissues. The protective layer of this plant becomes thicker and the transport tissues are differentiated as development progresses, which decreases the absorption of herbicide molecules. Another factor that makes this plant harder to control is that it is resistant to glyphosate (Yu *et al.,* 2015; Han *et al*., 2017), which is why it is the most common weed in glyphosate-resistant soybean crops (Cavalcante *et al.,* 2018; Takano, Oliveira Jr, Constantin, Silva & Mendes, 2018). In turn, *Bidens* sp. is one of the most serious weedy plants in annual crops. It presents high capacity for seed production (3,000 to 6,000) and is able to produce 3 to 4 annual generations, which allows a high rate of infestation; moreover, its seeds are able to survive 3 to 5 years when buried in soil and this species is a host for Meloidogyne and Pratylenchus nematodes (Brandel, 2004; Tsai *et al*., 2008). *Bidens pilosa* is able to excrete allelochemicals that hinder the germination and growth of potentially competing plants and release these compounds in the atmosphere through the lixiviation of its leaves and other aerial parts and through other pathways that include volatile emissions, root exudate and litter (Deba, Xuan, Yasuda & Tawata, 2008; Budumajji, Raju, & Jacob, 2018).

*Merremia aegyptia* (L.) Urb. is a plant species of the Convolvulaceae family and can develop in various crop systems (Lakshminarayana & Raju, 2018). This weed presents aggressive branching behavior, with a single plant occupying an area of up to 4 to 5 m$^2$, becoming the dominating species in that area by either suppressing or killing smaller plants (De Lima, Linhares, Liberalino Filho & Neto, 2007; Azania, A., Azania, C., Pavani & Cunha*,* 2003). Freitas *et al*. (2009) reported a decrease in the number of weedy plants related to competition with dominant species that occupy a larger physical space, surpassing the others. The use of herbicides is one of the most common methods for weed control. In the present study, the use of these molecules was efficient, with control values above 90% for all compounds applied. In the *Opuntia* variety, only a single *Phyllanthus tenellus* Roxb. occurred but the plant showed symptoms of intoxication and was unable to conclude its physiological cycle. Similar behavior was observed for *Merremia aegyptia* (L.) Urb., which was unable to conclude its physiological cycle and, 30 days after the molecules were applied, there were no new seedlings over the follow-up period of up to 90 days after treatment. (Table 7). However, although all herbicides presented effective control over weedy plants, the herbicides oxyfluorfen and ametryn caused damages to the forage palm.

**Conclusion**

No difference was observed in the selectivity of herbicides for the *Opuntia* and *Nopalea* varieties of the forage palm of the state of Alagoas. Herbicides tebuthiuron, atrazine and flumioxazin were selective for these varieties at the recommended doses. Oxyfluorfen and ametryn caused severe phytotoxicity in both varieties of the plant studied. Tebuthiuron, atrazine, flumioxazin, oxyfluorfen and ametryn were efficient and controlled over 90% of the species present in this experiment. The efficacy of herbicides varies according to the weedy community and the dose of a.i. applied.

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