



## Phytochemical investigation, phenol content and allelopathic potential of *Croton heliotropiifolius* Kunth extract

### Investigação fitoquímica, conteúdo de fenol e potencial alelopático do extrato de *Croton heliotropiifolius* Kunth

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**ABSTRACT:** The present study involves phytochemical investigation, phenol content and allelopathic potential of *Croton heliotropiifolius* Kunth extract in the germination of *Digitaria insularis* (L.) Fedde. with extracts ethanolic and aqueous of *C. heliotropiifolius* leaf; was performed prospected secondary metabolites and Ultraviolet-visible (UV-visible) spectrophotometry tests for total phenols and flavonoids. The germination bioassay was performed in a BOD chamber (12L:12D) with plant extracts at 100, 50, 25 and 10%, with water as control against *D. insularis* seeds. The germination percentage (G%), germination speed index (IVG), abnormal seedlings (PAn%), root length and aerial part (CR and CPA) were evaluated. The extracts reduced the germination and development of the target species indicated by G% and IVG from the concentration of 10%. The same occurred with the variables PAn%, CR and CPA. In both extracts the presence of secondary metabolites was indicated, the phenol content obtained was 0.3422 mg EAG/g for the ethanolic extract and 0.375 mg EAG/g for the aqueous extract, for the flavonoids 0.1071 mg EQ/g in ethanolic extract and 0.0110 mg EQ/g in aqueous extract. The extracts demonstrated a reduction in germination and alteration in the physiology of *Digitaria insularis* (L.) Fedde seeds.

**KEYWORDS:** Extracts, Secondary Metabolites, Germination.

**RESUMO:** O presente estudo envolve investigação fitoquímica, conteúdo de fenol e potencial alelopático do extrato de *Croton heliotropiifolius* Kunth na germinação de *Digitaria insularis* (L.) Fedde. Com extratos etanólicos e aquosa da folha de *C. heliotropiifolius*; foi realizada prospecção de metabólitos secundários e Testes de espectrofotometria ultravioleta-visível (UV-visível) para fenóis totais e flavonóides. O bioensaio de germinação foi realizado em câmara BOD (12L: 12D) com extratos vegetais a 100, 50, 25 e 10%, tendo água como controle contra sementes de *D. insularis*. Foram avaliados a porcentagem de germinação (G%), índice de velocidade de germinação (IVG), plântulas anormais (PAn%), comprimento de raiz e parte aérea (CR e CPA). Os extratos reduziram a germinação e o desenvolvimento das espécies-alvo indicadas por G% e IVG a partir da concentração de 10%. O mesmo ocorreu com as variáveis PAn%, CR e CPA. Em ambos os extratos foi indicada a presença de metabólitos secundários, o teor de fenol obtido foi de 0.3422 mg EAG / g para o extrato etanólico e 0.375 mg EAG / g para o extrato aquoso, para os flavonóides 0.1071 mg EQ / g no etanólico extrato e 0.0110 mg EQ / g em extrato aquoso. Os extratos demonstraram redução na germinação e alteração na fisiologia das sementes de *Digitaria insularis* (L.) Fedde.

**PALAVRAS-CHAVE:** Extratos, Metabólitos secundários, Germinação.

## INTRODUCTION

Agriculture is a sector with high growth potential however, many crops of economic interest have been suffering from the presence of weeds, which affect their development, their damages restrict productivity and the quality of several crops, thus decreasing the producers profits (ŚLIWIŃSKI and SZCZEŚNIAK, 2012; ASHIQ and ASLAM, 2014; WANG et al., 2015; ERVIN and JUSSAUME, 2016; ESPEJO-GARCIA et al., 2020; SOTI et al., 2020).

Weeds have a rapid proliferation and a high degree of competitiveness for nutrients, water and light, directly interfering in the development of crops (RABÊLO et al., 2008; EKWEALOR et al., 2019; SOTI et al., 2020). There is a diversity of weeds that are very problematic for agriculture, such as *Digitaria insularis* (L.) Fedde, known as bitter grass, that intervene in the development of other crops (TABAGIO, 2008; RABÊLO et al., 2008).

In this way, there are different control methods commonly used in weeds such as manual, mechanical and chemical, however, among the disadvantages directed to these methods it is possible to mention, for example, high labor costs, the possibility of damaging natural resources and damage to living organisms in the environment, causing resistance to herbicides and leaving residues in food, among others (HAKANSSON, 2003; BLAIR et al., 2015; PERRY et al., 2009; MOSS, 2010; SCAVO et al., 2018).

To minimize the proliferation of weeds, and at the same time, reduce the use of chemical products, the use of appropriate alternative management methods is recommended, in order to mitigate weed interference and maintain the ecological, environmental and productive balance (JABRAN et al., 2015; FAVARETTO et al., 2018).

An investigated alternative is the use of allelopathy, which corresponds to the direct or indirect effect of one plant on another, through the production of chemical compounds (secondary metabolites) released into the environment capable of acting on the development of other plants (BAJWA, 2014; MUZELL- TREZZI et al., 2016; MOŹDŹEŃ et al., 2018). The allelopathic nature and/or bioherbicidal action of plants is related to their secondary metabolites, whose main groups are terpenes, alkaloids,

phenolic acids, lactones, coumarins, sesquiterpenes, phenols, tannins and flavonoids, among others (RICE, 1984; ZHANG et al., 2012; TAIZ and ZEIGER, 2013; JABRAN et al., 2015; FAVARETTO et al., 2018).

Euphobiaceae comprises a group of families of plant species that have a variety of plants with the most distinct activities already investigated, which it is possible to emphasize allelopathic activity (SISODIA and SIDDIQUI, 2010; GILANI et al., 2010; MARASCHIN-SILVA and AÇÚILA, 2006; MA et al., 2011; DE OLIVEIRA et al., 2020).

*Croton heliotropiifolius* Kunth (Euphobiaceae) popularly known as velame, velaminho, valame-de-cheiro and marmeleiro is distributed in northeastern Brazil, and belongs to the genus *Croton*, which includes plants rich in bioactive compounds (LUCENA, 2000; RANDAU et al., 2004; PAYO et al., 2001; SALATINO et al., 2007; COMPAGNONE et al., 2010; SILVA et al., 2010; SOUSA et al., 2014; SODRÉ e SILVA, 2015; CANELO et al. 2017).

The present research aimed to investigate the phytochemical composition of *Croton heliotropiifolius* (Euphobiaceae) extracts and their allelopathic potential on the germination of *Digitaria insularis* (L.) Fedde.

## MATERIALS AND METHODS

### *Plants samples*

The plant samples were obtained from *C. heliotropiifolius* leaves collected in the municipality of Arapiraca (latitude 09°45'09"S and longitude 36°39'40"W; average altitude 264 m), and seeds from *D. insularis* obtained at Rio largo (latitude 9°27'S, longitude 35°27'W, average altitude of 127m). The botanical identification was performed by the curator of the MAC herbarium at Institute of Environment (IMA) of Alagoas State - IMA, Rosângela Pereira de Lyra Lemos. Plants exsiccates were deposited under registration number 54392-IB/MAC (*Croton heliotropiifolius* Kunth) and 62206-IB/MAC (*Digitaria insularis* (L.) Fedde).

### ***Obtention of the extract***

The leaves of the plant were dried in an oven at 50 °C, and crushed in a knife mill to obtain the powder. Subsequently, the vegetable powder was sent to the appropriate procedures for obtaining the aqueous and ethanolic extracts.

Aqueous extract: 113g of the *C. heliotropiifolius* leaf powder (velame) was weighed and 1,7L of distilled water was added, and kept at 50°C for 5 min on a heating plate. After cooling, the material was filtered, transferred to 50 mL falcon tubes, then frozen in a freezer at -18°C for 72 hours and subsequently subjected to freeze drying (Model: MICRO MODULOYO-115, Serial No: 02J400021-1B), until obtaining the dry aqueous extract stored in an amber flask.

Ethanolic extract: For cold ethanol maceration, 50g of the powder was weighed and 2,8L of PA 99,5% ethyl alcohol was added. Every 24 hours for 6 days of extraction, the material was filtered and the solvent replaced. The collected filtrate was dried on a rotary evaporator (Brand: BUCHI Heating Bath B-490) to obtain the dry ethanolic extract stored in an amber flask.

Percentage yield extraction (R%): After obtaining each dry extract, percentage yield was calculated utilizing the following formula:  $R\% = (\text{mass of crude extract} / \text{mass of plant material}) \times 100$ .

### ***Laboratory germination bioassay***

The bioassay was conducted in a laboratory where seeds of *D. insularis* (n = 25) were distributed in Gerboxes with 9,0 cm in diameter, sterilized, and covered with two sheets of Germitest paper previously autoclaved. Then, 7 mL of each test solution of the respective aqueous and ethanolic extracts were added in concentrations of 1, 0 (100%); 0, 5 (50%); 0, 25 (25%) and 0, 1(10%) mg mL<sup>-1</sup>, using distilled water as control. The germinative bioassay was conducted in a germination chamber type BOD with a temperature of 25°C and photoperiod of (12L : 12D). The treatments were arranged in a factorial 2 (extracts) x 5 (concentrations) in a completely randomized design with four

replications. The evaluations were conducted daily with verification of the germination rate and germination speed index every 24 hours for ten days.

In the experiment, variables were evaluated as percentage of Germination (G %) using the formula:  $G \% = (N/A \times 100)$  where, N = Number of seeds germinated at the end of the test, A = Number of total seeds.

The Germination Speed Index (IVG) was obtained using the formula  $IVG = (G1/N1) + (G2/N2) + \dots + (Gn/Nn)$ , where G1, G2 ... Gn = Number of seedlings germinated each day, divided by N1, N2 ... Nn = number of days after sowing the first, second and last count (MAGUIRE, 1962). The percentage of abnormal seedlings (PA %) = (Percentage of abnormal seedlings/A) x100, where A = Number of seeds in the sample. Root length (CR) and Aerial part length (CPA).

#### *Statistical analysis*

For statistical analysis, the data obtained were subjected to analysis of variance and the means grouped by the Scott-Knott test at 5% probability using the Genes software (CRUZ, 2013).

#### ***Determination of total phenol content***

The determination of the total phenol content of the extracts was performed using the method of Folin-Ciocalteu (Scherer and Godoy, 2014), with some adaptations for its realization in 96-well microplates. Thus, in triplicate, test solutions were prepared from an aliquot of the methanolic solution of the extract (100µl), aqueous solution of Folin-Ciocalteu 1:11 (v/v) reagent (500µl) and aqueous Na<sub>2</sub>CO<sub>3</sub> solution (7, 5 %) (400µl), as white, the extract was replaced by methanol in the process. Then the solutions were vortex for 30 seconds and immediately after, an aliquot of each solution in triplicate (250 µl) was added to the wells of the microplate. The material was kept protected from light for 2 hours, with the end of the incubation time, the absorbance reading was performed at 740 nm, using a Ultraviolet-visible (UV-visible) spectrophotometry microplate reader (Thermo Scientific Multiskan Spectrum, Type1500, REF 51118750, SN 1500-634).

For the quantification, a calibration curve of gallic acid in the concentrations of 0,1 to 0,005mg mL<sup>-1</sup> was constructed. The results of the total phenol content were determined by interpolating the average absorbance of the samples, against the equation of the line obtained in the calibration curve of the gallic acid, and expressed in mg of EAG (gallic acid equivalent)/per g of the extract.

### ***Quantification of flavonoids***

The quantification of the flavonoid content in the samples was determined by a method described by Souza et al. (2011), with some adaptations being conducted in 96-well microplates. In each well (triplicate), an aliquot of the methanolic solution of the extract (200 µl) and methanolic solution of 2% aluminum chloride (100 µl) was added. The plate was kept in a dark environment for 30 minutes with absorbance reading at 420 nm, in an Ultraviolet–visible (UV–visible) spectrophotometry microplate reader (Thermo Scientific Multiskan Spectrum, Type1500, REF 51118750, SN 1500–634).

To determine the flavonoid content, a calibration curve was constructed at concentrations from 0.03 to 0.00125 mg mL<sup>-1</sup>. The results obtained were determined by interpolation of the absorbances mean of the samples on the equation of the straight line acquired by the quercetin calibration curve and expressed in mg of EQ (quercetin equivalent)/per g of the extract.

### ***Phytochemical prospecting***

To perform the phytochemical screening step was taken as basis the methodology proposed by Matos and Matos (1989), which was conducted with some adjustments in order to perform exploration of the following compounds: phenols, pyrogenic tannins, phobafenics tannins, anthocyanin and anthocyanidins, flavones, flavonols, xanthonas, chalcones, auronas, flavonoids, leucoanthocyanidins, catechins, flavonones, flavonols, xanthonas, steroids, triterpenoids and saponins.

## RESULTS

The Percentage Yield Extraction observed for the aqueous (Aqu) and ethanolic (Eth) extracts of *C. heliotropiifolius* leaves showed crude extract mass of 32,517g and 8,369g, respectively, with 28,7% and 16,7% percentage yield

Thus it was found that both analyzed extracts did not differ statistically in relation to the content of total phenols, however, in relation to the content of flavonoids the ethanolic extract differed statistically from the aqueous extract (Tab. 1).

**Table 1.** Content of total phenolics and flavonoids in *C. heliotropiifolius* leaf extract.

Extracts	Phenolic compounds mg EAG / g extract	Flavonoids mg EQ / g of the extract
Ethanol	0.3422a	0.1071a
Aqueous	0.3754a	0.0110b

Means followed by the same letter, in the column, do not differ statistically by the F test at the 5% probability level. **Source:** Research data (2021).

In order to analyze the allelopathic potential of the *C. heliotropiifolius* extracts, tests were performed that allowed results for germination percentage (%G), germination speed index (IVG), percentage of abnormal seedlings, root length and aerial part length, utilizing seeds of *D. insularis* exposed to each test solution (0; 10; 25; 50 and 100%) of both ethanolic and aqueous extracts.

In the results, it was observed that after the concentration of 10% the seeds suffered interference from the extracts in the germination that began to decrease, presenting an average value of 42% in the ethanolic extract and 69% in the aqueous extract. In general, all treatments affected the germination process, as there was a lower rate of germination percentage of treatments when compared to the control (Tab. 2).

The solutions of ethanolic extract showed average values of germination percentage that did not differ from each other, only in relation to the control. While in aqueous extract the solutions of 100 and 50% differed statistically from the solutions of 25, 10% and the control (Table 2). When analyzing the general average of the germination rate of the treatments between the extracts, it was observed that these differed from each other (Tab. 2).

**Table 2.** Average values of the germination percentage (%) of *Digitaria insularis* seeds submitted to *C. heliotropiifolius* leaf extracts.

Treatments	Ethanol extract (%)	Aqueous Extract (%)	Average
Control	100.00±0.00Aa	100.00±0.00Aa	100.00±0.00a
10%	42.00±11.55Ab	69.00±8.87Bb	55.550±17.30b
25%	44.00±14.24Ab	54.00±10.58Bc	49.00±12.78b
50%	37.00±11.94Ab	42.00±13.66Ad	39.50±12.18c
100%	33.00±6.83Ab	40.00±6.53Bd	36.50±7.23c
Overall Average	51.2000±26.9299A	61.0000±24.0657B	

Means followed by the same uppercase letter in the row and lowercase in the column do not differ statistically by the F and Scott-Knott test, respectively, at the level of 5% probability. **Source:** Research data (2021).

When analyzing the results regarding the germination speed index (IVG) variable, it was observed that the average values of IVG began to decrease in both extracts after the concentration of 10% (Tab. 3). Thus, it suggests that the lower the average IVG values, the greater the time that the seeds needed to germinate. The treatments of ethanolic extract demonstrated values that varied from 1.05 to 0.82; the results did not differ statistically from each other, only in relation to the control (Tab. 3).

**Table 3.** Average value of the germination speed index of bitter grass seeds submitted to *C. heliotropiifolius* leaf extracts.

Treatments	Ethanol extract	Aqueous Extract	Average
Control	2.50±0.00Aa	2.50±0.00Aa	2.5000±0.00a
10%	1.05±0.29Ab	1.725±0.22Bb	1.3875±0.43b
25%	1.10±0.36Ab	1.35±0.26Bc	1.2250±0.32b
50%	0.92±0.30Ab	1.05±0.34Ad	0.9875±0.30c
100%	0.82±0.17Ab	1.00±0.16Ad	0.9125±0.18c
Overall Average	1.2800±0.6732A	1.5200±0.6016B	

Means followed by the same uppercase letter in the row and lowercase in the column do not differ statistically by the F and Scott-Knott test, respectively, at the level of 5% probability. **Source:** Research data (2021).

While in the aqueous extract the solutions 100 and 50% presented mean values of IVG inferior to the other solutions and to the control, thus the solutions of 100 and 50% differed from the solutions of 25 and 10% and also from the control (Tab. 3).

In general, the mean values of IVG resulting from the test solutions of both extracts on the seeds of *D. insularis*, after the end of 10 days of monitoring, showed a



statistical difference between them, the ethanolic extract showed a result of a lower average than the aqueous extract.

In the bioassay it was also observed that the seeds of *D. insularis* exposed to the different test solutions of *C. heliotropiifolius* leaves extracts demonstrated the occurrence of abnormal seedlings in the 10% solution. The occurrence of abnormal seedlings was not observed in the control.

According to the results obtained, the 100% solution differed statistically from the other test solutions of 10, 25, 50% in both extracts and also from the control evaluated against *D. insularis* seeds (Tab. 4).

**Table 4.** Mean value of the percentage of abnormal seedlings of *Digitaria insularis* submitted to the extracts of the leaves of *C. heliotropiifolius*.

Treatment	Ethanol extract (%)	Aqueous Extract (%)	Average
Control	0.00±0.00Ac	0.00±0.00Ac	0.00±0.00c
10%	18.00±5.16Ab	18.00±5.16Ab	18.00±5.16b
25%	19.00±8.25Ab	20.00±5.66Ab	19.50±6.57b
50%	22.00±6.93Ab	22.00±5.16Ab	22.00±5.66b
100%	27.00±6.83Aa	29.00±5.03Aa	28.00±5.66a
Overall Average	17.20 ± 2.43A	17.80 ± 2.40A	

Means followed by the same uppercase letter in the row and lowercase in the column do not differ statistically by the F and Scott-Knott test, respectively, at the level of 5% probability. **Source:** Research data (2021).

The test solutions of the ethanolic extract demonstrated average values of abnormal seedlings that ranged from 18 to 27% ethanolic and 18 to 29% aqueous, without statistically difference in the values of the solutions between the extracts. The overall average percentage of abnormal seedlings of the test solutions in both extracts did not differ among themselves (Tab. 4).

The test solutions of *C. heliotropiifolius* leaf extracts also affected the length of the *D. insularis* root in the 10% solution (Tab. 5). The solutions of the ethanolic extract showed average values of root length that varied from 9.0743 to 8.2579mm, the results of the solutions from 10 to 100% were not statistically different from each other, only differed in relation to the control.

**Table 5.** Mean length of the root length of *Digitaria insularis* submitted to the extracts of the leaves of *C. heliotropiifolius*.

Treatments	Ethanol extract (mm)	Aqueous Extract (mm)	Average
Control	16.5754Aa	16.5754Aa	16.5754a
10%	9.0743Bb	14.1067Aa	11.5905b
25%	8.9642Bb	12.8893Aa	10.9268b
50%	8.5761Bb	12.6165Aa	10.5963b
100%	8.2579Bb	11.2671Aa	9.7625b
Overall Average	10.4136B	13.5225A	

Means followed by the same uppercase letter in the row and lowercase in the column do not differ statistically by the F and Scott-Knott test, respectively, at the level of 5% probability. **Source:** Research data (2021).

The solutions of the aqueous extract demonstrated values that ranged from 14.1067 to 11.2671mm, the average values of root length did not differ statistically from each other, neither in relation to the control (Tab. 5).

As the concentration in the test solutions of extracts increased, the root length decreased. In general, the overall average of both extracts solutions were statistically different, evidencing the ethanolic extract and the aqueous 10.4136mm 13.5225mm (Tab. 5).

Another variable analyzed was the length of the aerial part of *D. insularis*, where the average results presented by the solutions of the extracts ranged from 11.2952 to 9.6912mm in ethanolic and 13.4028 to 11.9486mm in water (Tab. 6). It was observed in the results that the length of the aerial part decreases as the concentration increases.

**Table 6.** Mean length of the aerial part of *Digitaria insularis* submitted to the extracts of the leaves of *C. heliotropiifolius*.

Treatments	Ethanol extract (mm)	Aqueous Extract (mm)	Average
Controle	13.9158Aa	13.9158Aa	13.9158a
10%	11.2952Bb	13.4028Aa	12.3490a
25%	10.6730Bb	13.3075Aa	11.9902a
50%	10.0675Bb	13.1691Aa	11.6183b
100%	9.6912Bb	11.9486Ab	10.8199b
Overall Average	11.1285B	13.1488A	

Means followed by the same uppercase letter in the row and lowercase in the column do not differ statistically by the F and Scott-Knott test, respectively, at the level of 5% probability. **Source:** Research data (2021).

In the ethanolic extract, the results presented by the 10 to 100% solutions did not differ statistically from each other, only in relation to the control, while the aqueous

extract of 100% solution differed from the other solutions and the control (Tab. 6). The general average of the solutions of each extract differed statistically, the ethanol extract 11,1285mm and aqueous extract 13,1488mm (Tab. 6).

## DISCUSSION

The flavonoid content of the ethanolic extract was higher than the aqueous one, in addition, there was evidence of the presence of some groups of secondary metabolites between the extracts. In scientific studies present in the literature with other plant species, the presence of metabolites was verified and their relationship with the studied of allelopathic action was suggested, highlighting some plant species such as, for example, *Flaveria bidentis* (L.) Kuntze (ZHANG et al., 2012); *Rhazya stricta* (ALQARAWI et al., 2018) *Plectranthus amboinicus* (Lour.) and *Ocimum basilicum* L. (EL-ROKIEK et al., 2018); *Amburana cearensis* (OLIVEIRA et al., 2020), *Fimbristylis miliacea* (L.) Vahl. (DA SILVA et al., 2020a), *Cyperus distans* L., *Cyperus laxus* Lam. and *Cyperus rotundus* L. (DA SILVA et al., 2020b).

The phenolic compounds have a series of effects on the physiology and biochemistry of the organism, affecting, for example, the cell wall, cell respiration, which compromises the permeability of the membranes and absorption of nutrients, affecting the growth of the target plant (LI et al., 2010; WEIN et al., 2004; IMATORI et al., 2013).

Compounds such as flavonols, flavones, flavanones, isoflavonoids and anthocyanins, among others present in a variety of plants, have multiple functions, which it is possible to highlight for the agricultural sector, the allelopathic action, important for the protection of crops against the action of invasive species (FIORENZA et al., 2016; SSALI et al., 2019). Studies demonstrate that phenolic acids and flavonoids have depressive effects, considering that they can affect the seedling germination and development, interrupting these processes (LI et al., 2010; FAN et al., 2010; LADHARI et al., 2018; SSALI et al., 2019).

According to the study performed by Pereira et al. (2019) several secondary metabolites have been indicated in the different fractions of the *Paspalum maritimum*

Trind extract, suggesting that the compounds identified may be related to the allelopathic potential conferred by the extract of this plant. Allelopathic studies of plant species are relevant because they act in reducing the spread of weeds in agricultural environments. Thus, many extracts that are sources of several metabolites become a promising source of natural herbicides, which tend to benefit many crops (JABRAN, 2017).

In this study, the investigation of the allelopathic potential of *C. heliotropiifolius* leaves extracts against the target plant *Digitaria insularis* under experimental laboratory conditions, demonstrated, from the evaluated parameters, interference in the germination processes and development of roots and aerial part. These parameters have also been the subject of studies in the literature for allelopathic activity with some plant species of Euphobiaceae family (SISODIA and SIDDIQUI, 2010; GILANI et al., 2010; DE OLIVEIRA et al., 2020).

Regarding the extracts allelopathic potential used in this study, the germination process and germination speed index (IVG) were analyzed, which comprise one of the parameters commonly used to assess allelopathic activity. In the present research, it is emphasized that the extracts reduced the germination and IVG of *Digitaria insularis* in the 10% solution. In the literature about targeting allelopathic action, these parameters were also evaluated in several extracts against target plants, such as, for example, *Flaveria bidentis* (L.) Kuntze on cotton seeds (*Gossypium herbaceum* L.) (ZHANG et al. 2012); extracts of *Rhazya stricta* on *Salsola villosa* (ALQARAWI et al., 2018) and extracts of *Plectranthus amboinicus* (Lour.) and *Ocimum basilicum* L. in the growth of *Phalaris minor* (EL-ROKIEK et al. 2018), *Fimbristylis miliacea* (L.) Vahl. inhibiting, *Emilia fosbergii* Nicolson (DA SILVA et al., 2020a).

A study conducted with *Sapindus mukorossi* Gaertn. demonstrated that the ethanolic extract of leaves prevents the growth of *Avena fatua* L. and *Amaranthus retroflexus* L. (MA et al. 2018). According to Mozden et al. (2018) when analyzing the extracts influence of *Galinsoga parviflora* Cav and *Oxalis Fontana* on the germination of *Raphanus sativus* var. radícula cultivars Krakowianka, Pódluga, Rowa showed an allelopathic action of the extracts, where germination decreased as the concentrations of the extracts increased.

The extracts of *C. heliotropiifolius* also induced the growth of abnormal seedlings in *D. insularis*, in the primary roots there was an aspect of deformation and atrophy in some cases tissue necrosis. It was also registered by the authors of this study that as the concentration of the extracts increases, consequently there is also an increase in the rate of abnormal seedlings of the target plant. The presence of abnormal seedlings was also reported in a study with the extract of *Eucalyptus urograndis* on the seeds of the target plants *Urochloa decumbens* and *Panicum maximum* (CARVALHO et al. 2015).

In the present study, *C. heliotropiifolius* extracts affected the development of the aerial and root part of *D. insularis*, progressively reducing the length when exposed to increasing concentrations. In studies with *Paspalum maritimum* Trind. from the extract of the aerial part at the concentration of 7,49 mg/mL and root at the concentration of 11,41 mg/mL, demonstrated an allelopathic effect affecting the germination and seedling growth of the target species *Lactuca sativa* L., *Digitaria insularis* (L.) Fedde., *Emilia coccinea* (Sims) G. Don and *Portulaca oleracea* L. (PEREIRA et al. 2019).

In studies conducted by Da Silva et al. (2019) the methanolic root extract of *Euphorbia heterophylla* L. at a concentration of 2,0 mg/mL inhibited germination by 100%, in addition to the growth of roots and aerial parts of target *Sorghum bicolor* and *Lactuca sativa* L. According to studies performed by Sisodia and Siddiqui (2010), they emphasized that the length of the root and aerial part of weeds *Melilotus alba* Medik., *Vicia sativa* L. and *Medicago hispida* Gaertn, decreased according to exposure to concentrations of 0,5; 1; 2 and 4% of *Croton bonplandianum*.

## CONCLUSION

It was concluded in the present study that the seeds of *D. insularis* are sensitive to the solutions of ethanolic and aqueous extracts of the *C. heliotropiifolius* leaf, emphasizing that the results of the ethanolic extract was highlighted when compared to the aqueous extract. Thus, it is suggested that *C. heliotropiifolius* leaf extracts are a potential source of compounds that act in weed control.

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