

Bioactivity of the organic extracts of *Cnidoscolus urens* (L.) Arthur (Euphorbiaceae) on the cabbage-caterpillar

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Abstract

The aim of the current study was to identify the phytochemical profile of several different fractions of the ethanolic extract taken from the cansanção-nettle leaves, as well as assess the bioactivity on *Ascia monuste orseis*. After gathering the plant samples, the species was identified, and the organic extracts were collected after processing was completed in the laboratory. The fractions obtained were as follows: hexane (Hex-F), chloroform (CHCl₃-F), ethyl acetate (AcOEt-F) and methanolic (MeOH-F). In the context of the insect, the biological parameters evaluated included the incubation period, egg viability, length of the larval stage and mortality, pupal viability and weight, number of defective adults and leaf consumption. The extracts revealed a large percentage of anthraquinones, coumarins, anthracene derivatives, terpenes and steroids and water-soluble tannins. All the organic extracts were confirmed to exert a negative effect on the embryonic, larval, pupal and adult phases of the cabbage-caterpillar, implying that the chloroform fraction induced 100% mortality in this pierid. It was also possible to verify that all the fractions also affected the feeding behavior of the cabbage-caterpillar, decreasing the leaf consumption by the larvae during the organic extract treatments.

Keywords: *Ascia monuste orseis*, botanical insecticides, secondary metabolites, cansanção-nettle

Introduction

Ascia monuste orseis (Lepidoptera: Pieridae), popularly known as the cabbage-caterpillar, is one of the principal pests on brassicaceae. The most frequently employed pest management method, in agriculture, in Brazil, is the chemical method (Bortoli et al., 2014). However, such unsystematic applications of these agrochemicals have resulted in several difficulties, especially the production of arthropod-plague populations impervious to the main available active principles (Trindade et al., 2015). The urgent need to counter such issues has prompted the development of alternative and integrated control methods, and the use of

botanical extracts is an effective one. According to the findings from some studies the botanical extracts directly interrupt the behavioral and developmental activities of insects and mites, exerting a negative influence on the various stages of their biological cycles (Lovatto, 2012; Ribeiro et al., 2016; Macagnan et al., 2016).

As the coatinga biome is rich in biodiversity with favorable ecophysiological conditions, it carries a huge potential for bioactive secondary compounds to be produced. Therefore, a variety of plant species have been subjected to testing for insecticidal activity.

The genus *Cnidoscolus* (Euphorbiaceae) has caught the attention of the scientific world,

Received: 23 February 2017
Accepted: 24 July 2018

mostly for the many biological actions it can perform (Valenzuela Soto et al., 2015; Jaramillo et al., 2015; Nunes et al., 2016). The plant extracts from various species of this genus have also been reported to induce alterations in the biological cycles of the arthropod-pests (Boiça Junior et al., 2013; Carvalho et al., 2014; Gomes et al., 2014; Candido & Bezerra, 2015).

An analysis of the dead insects of genus *Cnidoscopus* have revealed that anthocyanins, anthraquinones, coumarins, flavonoids, steroids, lignans, saponins, tannins, terpenoids, xanthines and alkaloids are present in the different plant parts (Obichi et al., Morais et al., et al., 2016, and Paula et al., 2016).

Investigations conducted on the secondary constituents of these botanical extracts enables the identification of new substances that can act as potential insecticides, while fulfilling the requirements of effectiveness, safety and selectivity (Silva et al., 2014). According to Corrêa & Salgado (2011) the botanical extracts reveal bioactivity which is directly linked to the synergistic action of this metabolite complex.

In order to extract, separate, purify and identify the secondary constituents of the various plant extracts, several eluents and chromatographic techniques can be employed for clearer understanding of the systematic bioactivity (Cechinel Filho & Yunes, 1998; Lima Neto et al., 2015). It was Chirinos et al., (2007) who highlighted that for the secondary substances to be extracted, the chemical nature of the compounds and, particularly, the composition of the extracting solvent selected for it, need to be evaluated.

From the preliminary studies it is evident that the ethanolic extracts from nettle-fatigue leaves exhibit insecticidal activity on the cabbage-caterpillar (Carvalho Neto et al., 2017). In this context, this study aimed at conducting laboratory bioassays to establish the phytochemical profile and estimate the bioactivity of the individual fractions of this extract on specific biological features of the cabbage-caterpillar.

Material and methods

From February / 2014 to January / 2016,

all the experiments were performed in the Laboratory of Apiculture and Entomology of the Agricultural Sciences Campus - CCA and the Organic Chemistry Laboratory of the Federal University of the São Francisco Valley - Univasf. All the tests were maintained in an air-conditioned room, at $25 \pm 2^\circ$ C temperature and $60 \pm 10\%$ relative humidity, with a 12-hour photophase.

The *C. urens* plant species were gathered from the community during February / 2014. Access was duly registered in the National System of Genetic Heritage Management and Associated Traditional Knowledge - SisGen, as No. A9B58BC. When the collection was being done, this region had received no rainfall for the last three years prior. The professionals of the Ecology and Environmental Monitoring Center - NEMA of UNIVASF, performed the botanical identification from an *exsicata* deposited in the herbarium of the Brazilian Agricultural Research Company - Embrapa Semiárido / CPATSA (Virtual Herbarium of Flora and Fungus - INCT-HVFF) under fall No. HTSA 6294.

To process the vegetal material and draw out the crude ethanolic extract and its organic fractions, the nettle-fatigue leaves were oven-dried under circulating air at 40° C, until they achieved constant weight. In the next step they were processed in a knife mill, to produce a dry powdered sample of the vegetable material. The material then passed through an exhaustive maceration system, with the extracting liquid (ethanol 95% Laboratory Grade) being renewed every 72 hours. This extract was evaporated in a rotary evaporator at 50° C under reduced pressure, to remove the solvent. The extractions were thus performed seven times to acquire the ethanolic leaf extract (EPS) sample. The fractions were obtained by subjecting the ethanolic leaf extract (EPS) to vacuum liquid chromatography (CLV) utilizing silica gel 60 as the stationary phase, and the solvents hexane, chloroform, ethyl acetate and methanol in increasing polarities as the mobile phase. Each fraction was then evaporated in a rotary evaporator at 50° C under reduced pressure, to remove the solvent. Eight such extractions were done to gain each fraction of the EPS. Once the solvent was evaporated, the fractions of hexane (Hex-F),

chloroform (CHCl₃-F), ethyl acetate (AcOEt-F) and methanolic (MeOH-F) were obtained. One leaf ethanolic extract (FES) sample and fraction were separated for phytochemical studies.

The *A. monuste orseis* specimens were collected and the stock was maintained from the postures gathered in the urban gardens in Petrolina - PE and Juazeiro - BA. Once the postures were transported to the laboratory they were conditioned in paper lined trays, moistened with distilled water. They were placed in an air-conditioned room to raise caterpillars of known ages for bioassay use. The caterpillars were transferred, as soon as they emerged, to polyethylene cages (45.7 x 32.6 x 28.0 cm) provided with openings on the top and the side surfaces lined with voile-type tissue. They were left undisturbed until the pupal stage, and were fed everyday on organically raised cabbage. The adults which emerged were transferred to an outdoor screened cage (2.00 x 2.00 m), which enclosed kale plants raised in pots, as a base for the oviposition. The adults were fed on 10% honey solution, which was changed daily to prevent fungal growth. Cabbage leaves bearing the postures were cut and transferred to the laboratory, where a few of them were used in the experiments and the remaining were stored to maintain the stock.

The classes of the secondary constituents present in the EPS and fractions were identified using phytochemical screening by applying the samples on an Analytical Thin Layer Chromatography (CCDA) plate using a capillary. Elution was then performed in specific solvent systems or standards for each molecule group. The chromatate plates thus obtained were developed in a dark chamber under UV radiation (\square) at wavelengths (\square) of 254 and 366 nm, followed by reactive staining and heating, as cited by Wagner & Bladt (1996). A total of 14 classes of secondary metabolites were investigated, viz., alkaloids, anthocyanins, anthraquinones, flavonoids, coumarins, anthracene derivatives, lignans, mono-, sesqui-, and diterpenes, naphthoquinones, saponins, water-soluble tannins, condensed tannins, xanthines, triperpenes and steroids. Depending upon the pigmentation intensity of the substances

in the plates compared to the specific eluent and developer systems, the evaluation criteria were established. Here, (-) is the absence of the chemical constituent, (+) refers to the presence of the constituent in its lowest concentration, (++) is its presence in moderate concentration, and (+++) is its availability in higher concentration.

For the bioactivity experiments involving the effect of the organic extracts on *A. monuste orseis*, in keeping with the findings from the earlier studies using aqueous and ethanolic extracts of different nettle-fatigue parts (Carvalho Neto et al., 2017), the conclusion drawn was that the organic extracts will need to be used at 2% concentration (w / v) in all the bioassays. To apply the treatments, the organic extracts were first dissolved in acetone and then in distilled water, in proportions required to achieve 2% concentration. Regarding the utilization of acetone in the organic extracts, two controls, one using distilled and sterilized water (ADE) and the other with a combination of distilled water plus acetone, were included in all the tests. This was done to quantify the effect exerted only by the extract on the insects, in the bioassays.

In the experiment dealing with the effect of the organic extracts on the *A. monuste orseis* embryonic stage, the leaf cuttings bearing the postures (1 - 4 hours) were immersed for one minute in the different treatments. After excess moisture had evaporated, the cuttings were transferred to moistened, filter paper-lined petri dishes. Every day, the number of caterpillars hatched on each plate were counted, and the incubation period (days) and egg viability (%) recorded. The completely randomized experimental design was adopted, with six treatments and four replicates, with each replicate containing 200 eggs. Data were submitted to the analysis of variance and, when significant effect was attained, the means were compared by the Scott-Knott test, at 5% probability. To suit the statistical analysis, the data were transformed into $\sqrt{(x + 1.0)}$.

In the experiment focused on the influence exerted by the organic extracts on the *A. monuste orseis* development, the leaf disks (\varnothing 6 cm) were immersed for one minute in the different treatments. Once the excess moisture had evaporated, the disks were moved

to plastic containers (Ø 14 cm and 10 cm high) lined with moistened filter paper and provided with a voile-coated lid. On each disk, ten newly hatched caterpillars (between 0 and 12 hours of age) drawn from the stock-rearing batch were placed. Every day, the containers were cleaned, and fresh filter paper put in. These treated disks were offered to the caterpillars for 24 hours, after which time, all the treated disks were disposed of and the caterpillars were fed on untreated cabbage disks. These disks were replaced daily, and repeated until the caterpillars reached the pupal stage. As the caterpillars underwent transformation into pupae, they were weighed and placed individually in plastic containers (Ø 14 cm and 10 cm high) provided with a 'voile'-covered lid and left undisturbed until the adults emerged. The newly emerged adults were maintained in the deformation assessment vessels and the biological parameters listed were determined, viz., larval mortality (%), larval stage duration (days), pupal viability (%), pupal weight (mg) and number of defective adults (%). The completely randomized experimental design was adopted with six treatments and five replicates, and each replicate included ten caterpillars. Data were submitted to the analysis of variance and, when a significant effect was encountered, the means were compared by the Scott-Knott test, at 5% probability. To suit the statistical analysis, the data were transformed into $\sqrt{x + 1.0}$.

In the experiment considering the way the organic fractions affect the foliar consumption in the *A. monuste orseis* larvae, the identical extracts, treatments and procedures employed in the earlier test were used. Thus, the 10-day old caterpillars were individually picked and placed on the treated leaf disks. Then, 24 hours later, the images were recorded of the remaining leaf area of the disks after the caterpillars had fed on them. The leaf area was determined using the free access software ImageJ. The completely randomized experimental design adopted included six treatments and ten replicates, and each replicate was represented by one caterpillar. Data were submitted to the analysis of variance and, whenever a significant effect was noted, the means were compared using the

Scott-Knott test, at 5% probability.

Results and discussion

The phytochemical study (Table 1) facilitated identification of the secondary compounds in the different organic fractions of the nettle-fatigue and their probable actions on the cabbage-caterpillar, like decrease in mobility, larval growth and development, decline in pupal weight, pupal non-viability, morphological alterations in the wings, defective pupae, differences in pupal size, larval mortality, extension of the phase and incubation period, as well as incomplete ecdysis in the larval and pupal phases. These effects can be attributed to the presence of substantial and possible synergies of the chemical components viz., anthraquinones, coumarins, anthracene derivatives, mono-, sesqui- and diterpenes, triterpenes and steroids, as well as water-soluble tannins.

Peixoto-Sobrinho et al., (2012) observed similar compounds in *C. urens* in their investigation of the chemical constituents present in the shoots and roots of the cansanção-nettle, like the coumarins, mono- and diterpenes, tannins, triterpenes and steroids. In their evaluation of the phytochemical profile of the ethanolic extract and fractions (Hex, Hex / AcOEt and AcOEt) of *C. urens*, Souza (2014) reported the presence of tannins, steroids, mono-, sesqui-, di- and triterpenes.

Investigations performed through the phytochemical exploration of the various botanical extracts and the influence the secondary compounds exerted on different pest arthropods, specifically in terms of their biology and behavior, have been extensively discussed. Thus far, however, no study has been done on the kale caterpillar as the target insect. Andrade Filho et al., (2010) identified the presence of hydrolysable tannins, saponins, reducing sugars and triterpenes and the way they affect the development and mortality of *Bemisia tuberculata* Bondar (Hemiptera: Aleyrodidae). Cavalcante et al., (2006) also confirmed the effect of the tannins and alkaloids on the mortality and alterations in the fertility criteria for *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae). Restello et al., (2009) reported terpenes present

Table 1. Chromatographic phytochemical profile of the ethanolic leaf extract (EEF) from *C. urens* and the hexane (Hex-F), chloroform (CHCl₃-F), ethyl acetate (AcOEt-F) and methanol fractions ⁽¹⁾

Secondary Metabolite Class	EEF	Hex-F	CHCl ₃ -F	AcOEt-F	MeOH
Alkaloids	-	-	-	-	-
Anthocyanins	-	-	-	-	-
Anthraquinones	++	-	++	+	-
Flavonoids or Phenolic Compounds	++	-	+	+	++
Cumarins	-	-	+++	-	-
Anthracene Derivatives	+++	-	+++	+	-
Lignans	-	-	-	-	-
Mono-, sesqui- and diterpenes	+++	+++	+++	+	-
Naphthoquinone	++	-	+	-	-
Saponins	-	-	-	-	-
Water-soluble tannins	+++	-	++	++	-
Condensed tannins	-	-	-	-	-
Hatins	-	-	-	-	+
Triterpenes and steroids	+++	+++	+++	++	-

⁽¹⁾ (-) absence of constituent, (+) presence of constituent, (++) moderate presence of constituent, (+++): high presence of constituent.

in *Tagetes patula* demonstrating insecticidal effects on the *S. zeamais* adults (Coleoptera: Curculionidae). Silva et al., (2004) established that the tannins exert a larvicidal effect in *Aedes aegypti*. Thus, in the context of the 100% mortality observed in the *A. monuste orseis* larvae for the chloroform fraction treatment, the terpenes and steroids present in this fraction of the extract revealed the insecticidal potential of these components on the cabbage-caterpillar.

From the results shown in Table 2, the organic extracts, barring the ethyl acetate phase (AcOEt-F), affected the *A. monuste orseis* during the incubation phase, by lengthening its embryonic phase. Connected with the egg viability, the extracts showed no effect. This was attributed most likely to the presence of a lipid layer within the chorion, which is able to retain the toxic substances, thus restricting them from reaching the embryo (Machado et al., 2007).

Table 2. Bioactivity of the organic extracts present in the *A. monuste orseis* embryonic stage. (25 ± 2 ° C, 60 ± 10% RH and 12-hour photophase) ⁽¹⁾

Treatment ⁽²⁾	Period of incubation (days)	Viability (%)
Test. 1 (ADE)	4.31 ± 0.07 b	100 ± 0.00
Test. 2 (ADE + Acetone)	4.22 ± 0.23 b	100 ± 0.00
Hex-F	4.61 ± 0.21 a	100 ± 0.00
AcOEt-F	4.01 ± 0.25 b	100 ± 0.00
CHCl ₃ -F	4.94 ± 0.24 a	100 ± 0.00
MeOH-F	4.73 ± 0.17 a	100 ± 0.00
CV (%)	3.84	-

⁽¹⁾ For the statistical analysis, the data were transformed as $\sqrt{x + 1.0}$. Means followed by the same letter, in the columns, do not differ by Scott-Knott's test, with the probability at 5%. ⁽²⁾ ADE - Distilled and Sterilized Water; Organic Fractions of the Ethanolic Leaf Extract (EEF), hexane (Hex-F), chloroform (CHCl₃-F), ethyl acetate (AcOEt-F) and methanolic (MeOH-F).

The negative effect exerted by the botanical extracts of different species and plant structures on lepidoptera has been extensively researched. Biermann (2009) in his assessment of the influence exerted by ten extracts on the *A. monuste orseis* eggs identified that the tobacco extract from *Nicotiana tabacum* revealed activity during the embryonic stage of the cabbage caterpillar, impeding the hatching of 100% of the larvae. The ovicidal effects caused by a variety of organic extracts on lepidoptera is

well recognized and been recorded by several authors (Alves et al., 2012; Freitas et al., 2014 and 2015; and Magrini et al., 2015) and the results have indicated a negative influence on both the length of the incubation phase, with this stage being extended, and the egg viability, restricting the larvae from hatching.

The organic extracts were observed to induce larval mortality in *A. monuste orseis* at concentrations above 60%, for all the fractions (Table 3). The bioactive influence exerted by

the chloroform fraction (CHCl₃-F), inducing 100% mortality and noted at 10 days of age, was very different from the other extracts and controls. The manner that the chloroform fraction behaved both in larval mortality and during the embryonic phase of the *A. monuste orseis* suggests that

the bioactivity of the botanical extracts can demonstrate different actions depending on the biological cycle of the insects. The lack of any mortality in the control with acetone indicates that the choice of this solvent for the solubilization of the organic extracts did not hinder their effects.

Table 3. Biological aspects (mean ± SEM) of *A. monuste orseis* fed with cabbage disks treated with organic extracts of *C. urens*. (25 ± 2° C, 60 ± 10% RH and 12-hour photophase) ⁽¹⁾

Treatment ⁽²⁾	Mortality (%)	Duration of the Larval Phase (days)	Weight of pupa (mg)	Pupal Viability (%)	Defective Adults (%)
Test 1 (ADE)	0 ± 0.00c	16.42 ± 0.50a	0.26 ± 0.01a	100 ± 0.00 a	2.0 ± 3.54 b
Test 2 (ADE + Acetone)	0 ± 0.00c	17.08 ± 0.19a	0.26 ± 0.01a	100 ± 0.00 a	4.0 ± 3.54 b
Hex-F	82.0 ± 16.71b	9.94 ± 6.57a	0.13 ± 0.09b	18.0 ± 16.71b	14.0 ± 13.54 a
AcOEt-F	76.0 ± 3.54b	15.4 ± 0.71a	0.18 ± 0.02b	24.0 ± 3.54 b	20.0 ± 6.77a
CHCl ₃ -F	100.0 ± 0.00a	-	-	-	-
MeOH-F	68.0 ± 23.36b	12.9 ± 5.89a	0.12 ± 0.06b	32.0 ± 23.36 b	24.0 ± 15.68 a
CV (%)	15.34	26.63	2.92	27.86	56.36

⁽¹⁾ The data were transformed as $\sqrt{x + 1.0}$ to suit the statistical analyses. Means followed by the same letter in the columns do not differ from each other by the Scott-Knott test at 5% probability. ⁽²⁾ ADE - Distilled and Sterilized Water; Organic Fractions of Ethanol Leaf Extract (EEF), hexane (Hex-F), ethyl acetate (AcOEt-F), chloroform (CHCl₃-F), and methanolic (MeOH-F).

The effects exerted by the organic extracts from the plants belonging to genus *Cnidocolus* on a variety of biological features of the pest arthropods have been documented. However, while very few reports exist on the insecticidal activity of *C. urens*, there are no studies on the influence that these botanical extracts from this species has on the cabbage-caterpillar. Candido et al., (2013) confirmed that the organic extract of *C. phyllacanthus* exerted a larvicidal effect in the control of *Aedes aegypti*. Numa et al., (2015) in their investigation on the acaricidal action of certain organic extracts, corroborated the consequences of the ethanolic extract of *C. aconitifolius* leaves on the mortality of the adult *Tetranychus urticae* Koch females. Cruz et al., (2012) reported the termiticidal effect of the powdered root of *C. urens* on the *Nasutitermes* sp. adults.

From the results it is also evident that when the effects of the hexane (Hex-F), ethyl acetate (AcOEt-F) and methanolic (MeOH-F) fractions on the *A. monuste orseis* larval, pupal and adult phases were compared with the controls, they did not differ from each other. It was also noted that these organic extracts did not alter the length of the larval phase of the cabbage-caterpillar when compared with the controls. As all the larvae of the chloroform fraction (CHCl₃-F) treatment succumbed showing 100% mortality, the other

biological aspects of this treatment could not be assessed. These findings clearly demonstrate the insecticidal potential of the CHCl₃-F fraction of *C. urens* on the cabbage-caterpillar. It can be certified that in all the treatments the pupae showed reduction in weight, when compared with the witnesses. During the experiment, the fifth instar larvae and pupae from treatments with the extract, revealed incomplete ecdysis - deformations, and produced above 60% of pupal infeasibility in all the treatments. As for the adults, all the extracts induced different degrees of morphological alterations in the wings in comparison to the control, in the range of 14 to 24%.

Investigations into the bioactivity of the hexane, ethyl acetate, chloroform and methanolic fractions drawn from a variety of plant extracts have revealed that caterpillars raised on an artificial diet treated with different extracts register a decrease in the pupal weight, larval size, feed conversion efficiency, embryonic development, morphometric alterations in the adults, defective pupae, differences in the pupal sizes obtained, larval mortality, flawed adults with retention of the frontal carapace, deformed wings, inhibited growth and incomplete ecdysis (Matos et al., 2006; Cunha et al., 2008; Ribeiro et al., 2015).

All the organic extracts were able to

decrease the leaf consumption by *A. monuste orseis*. The ethyl, hexane and chloroform acetate fractions induced a greater than 70% decline in foliar feeding, while the methanolic fraction caused a reduction of over 50% (Table 4). There is

the possibility of a repellent and phagdeterrante effect exerted by the organic extracts of *C. urens* on the feeding habits of the cabbage-caterpillar. However, specific studies in the future are necessary for these parameters to be analyzed.

Table 4. Leaf consumption during 24 hours of *A. monuste orseis* fed on the cabbage leaf disks treated with the organic extracts of *C. urens* (25 ± 2 ° C, $60 \pm 10\%$ RH and 12-hour photophase) ⁽¹⁾

Treatment ⁽²⁾	Consumption (%)
Test. 1 (ADE)	98.51 ± 0.00 c
Test. 2 (ADE + Acetona)	98.73 ± 0.00 c
Hex-F	20.26 ± 19.20 a
AcOEt-F	28.82 ± 11.70 a
CHCl ₃ -F	16.3 ± 8.63 a
MeOH-F	49.77 ± 3.15 b
CV (%)	25.20

⁽¹⁾ The means followed by the same letter in the columns do not differ from each other by the Scott-Knott test at 5% probability. ⁽²⁾ ADE - Distilled and Sterilized Water; Organic Fractions of Ethanol Leaf Extract (EEF), hexane (Hex-F), ethyl acetate (AcOEt-F), chloroform (CHCl₃-F), and methanolic (MeOH-F).

The fact that the plant extracts, mostly in the aqueous form negatively affect the foliar consumption in the *A. monuste orseis* caterpillars, had been recorded even earlier by several other authors (Medeiros & Boiça Junior, 2005; Medeiros et al., 2007; Mata & Lomonaco, 2013). The actions of the organic extracts (hexane, ethyl acetate, chloroform, ethanolic and methanolic fractions) on the dietary behavior of other lepidoptera have also been explored, and a marked decline in leaf litter consumption on treated substrates was reported (Andrade et al., 2016; Freitas et al., 2014; Alves et al., 2012). However, it is striking that there are no scientific reports that have reported the effect of organic extracts from species belonging to genus *Cnidocolus* on the cabbage-caterpillar.

Although the *C. urens* extracts demonstrate promising effects on *Ascia monuste orseis*, further studies are essential to gain greater understanding of their bioactivity in other biological facets of the kale and the ways they affect the non-target beneficial organisms.

Conclusions

All the fractions (hexane, ethyl acetate, chloroform and methanolic) of *C. urens* exert negative influences on the embryonic, larval, pupal and adult phases of *A. monuste orseis*, disturbing the developmental cycle of this insect.

All fractions of the nettle-fatigue extract were found to affect the feeding behavior of the cabbage-caterpillar negatively, thus decreasing

the leaf consumption of the larvae during the treatments.

In the *A. monuste orseis* larvae the ingestion of the chloroform fraction induced 100% mortality.

The organic extracts from *C. urens* are composed of chemical components belonging to the secondary metabolite classes, which include the anthraquinones, coumarins, anthracene derivatives, mono-, sesqui-, di- and triterpenes, steroids and water-soluble tannins.

Acknowledgment:

The authors express their gratitude to the Center for Studies and Research in Agroecology Sertão Agroecológico: NUPESA (referred to as 81/2013 - Univasf / CNPq / MDA / SAF / MEC / MCTI / MAPA) and Embrapa Semiárid Nucleus of Agroecology (referred to as 38/2014 - Embrapa / CNPq / MDA) for the financial support and scholarship granted to them.

References

- Alves, D.S., Carvalho, G.A., Oliveira, D.F., Sâmia, R.R., Villas-Boas, M.A., & Côrrea, A.D. 2012. Toxicity of copaiba extracts to armyworm (*Spodoptera frugiperda*). *African Journal of Biotechnology* 11: 6578-6591.
- Andrade Filho, N.N.D., Roel, A.R., Porto, K.R.D.A., Souza, R.O., Coelho, R.M., Portela, A. 2010. Toxicity of aqueous extract of *Anacardium humile* leaves on *Bemisia tuberculata*. *Ciência Rural* 40: 1689-1694.
- Andrade, J.V., Alcassio, T.P., Saldanha, F.G.,

- Toscano, L.C., Dias, P.M. 2016. Efeito antialimentar de extratos hidroalcoólicos de *Melia azedarach* (Meliaceae) sobre *Helicoverpa armigera* (Lepidoptera: Noctuidae). *Revista de Agricultura Neotropical* 3: 39-43.
- Biermann, A.C.S. 2009. *Bioatividade de Inseticidas Botânicos sobre Ascia monuste orseis* (Lepidoptera: pieridae). 72f. (Dissertação de Mestrado em Agronomia) - Universidade Federal de Santa Maria, Santa Maria, Brasil.
- Boiça Júnior, A.L., Janini, J.C., de Souza, B.H.S., Rodrigues, N.E.L. 2013. Efeito de cultivares de repolho e doses de extrato aquoso de nim na alimentação e biologia de *Plutella xylostella* (Linnaeus) (Lepidoptera: Plutellidae). *Bioscience Journal* 29: 22-31.
- Bortoli, S.A.D., Carvalho, J.D.S., Vacari, A.M., Goulart, R.M. 2014. Consumo foliar da traça-das-crucíferas em couve e brócolis tratados com sinigrina. *Arquivos do Instituto Biológico* 81: 264-271.
- Candido, L.P., Cavalcanti, M.T., Beserra, E.B. 2013. Bioactivity of plant extracts on the larval and pupal stages of *Aedes aegypti* (Diptera, Culicidae). *Revista da Sociedade Brasileira de Medicina Tropical* 46: 420-425.
- Candido, L.P., Beserra, E.B. 2015. Repellent activity of *Cnidocolus phyllacanthus* Mart. and *Ricinus communis* L. extracts against *Aedes aegypti* L. oviposition behavior. *Biotemas* 28: 105-112.
- Carvalho, G.S., Silva, L.S., Silva, L.B., Almeida, M.L.S., Pavam, B.E., Peres, M.T.L.P. 2014. Mortalidade e comprometimento do desenvolvimento de *Zabrotes subfasciatus* Boh. (Coleoptera: Chrysomelidae), induzido pelo extrato de sangra d'água *Croton urucurana* Baill (Euphorbiaceae). *Comunicata Scientiae* 5: 331-338.
- Carvalho Neto, M.F., Gonçalves-Gervásio, R.C.R., Araújo, E.C.C., Almeida, J.C., Guimarães, A.L. 2017. Phytochemical profile of cansanção-nettle extracts and bioactivity on the-cabbage-caterpillar. *Pesquisa Agropecuária Brasileira*. 53: 841 – 848.
- Cavalcante, G.M., Moreira, A.F.C., Vasconcelos, S.D. 2006. Potencialidade inseticida de extratos aquosos de essências florestais sobre mosca-branca. *Pesquisa Agropecuária Brasileira* 41: 9-14.
- CechinelFilho, V., Yunes, R.A. 1998. Estratégias para a obtenção de compostos farmacologicamente ativos a partir de plantas medicinais. Conceitos sobre modificação estrutural para otimização da atividade. *Química nova* 21: 99-105.
- Chirinos, R., Rogez, H., Campos, D., Pedreschi, R., Larondelle, Y. 2007. Optimization of extraction conditions of antioxidant phenolic compounds from mashua (*Tropaeolum tuberosum* Ruiz & Pavón) tubers. *Separation and Purification Technology* 55: 217-225.
- Corrêa, J.C.R., Salgado, H.R.N. 2011. Atividade inseticida das plantas e aplicações: revisão. *Revista Brasileira de Plantas Mediciniais* 13: 500-506.
- Cruz, C.S., Medeiros, M.B., Gomes, J.P., Sousa, F.C. 2012. Uso de plantas em pó seco com propriedades termiticida sobre a mortalidade de cupins arbóreos. *Revista Verde de Agroecologia e Desenvolvimento Sustentável* 7: 01-05.
- Cunha, U.S.D., Vendramim, J.D., Rocha, W. C., Vieira, P.C. 2008. Bioatividade de moléculas isoladas de *Trichilia pallida* Swartz (Meliaceae) sobre *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). *Neotropical entomology* 37: 709-715.
- Freitas, A.F., Pereira, F.F., Formagio, A.S.N., Lucchetta, J.T., Vieira, M.C., Mussury, R.M. 2014. Effects of methanolic extracts of *Annona* species on the development and reproduction of *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae). *Neotropical Entomology* 43: 446-452.
- Freitas, A.F., Formagio, A.S., Pereira, F.F., Lucchetta, J.T., Vieira, M.D.C. 2015. Effect of extracts of *Trichilia silvatica* C. DC., on development and reproduction parameters of *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae). *African Journal of Biotechnology* 13: 2041-2049.
- Gomes, L. M.A., Lima-Saraiva, S.R.G., Andrade, T.M.D., Silva, J.C., Diniz, T.C., Barreto, V.N.S., Silva Almeida, J.R.G. 2014. Antinociceptive activity of the ethanolic extract from barks and leaves of *Cnidocolus quercifolius* (Euphorbiaceae) in mice. *Journal of Young Pharmacists* 6: 64-69.
- Jaramillo Jaramillo, C.G., García Mir, V., Cuesta Rubio, O., Campo Fernández, M., García Simón, G. 2015. Fitoquímica preliminar, actividad antioxidante e hipoglucemiante de extractos de hojas de *Cnidocolus aconitifolius* (Mill.) IM Johnst (chaya). *Revista Cubana de Farmacia* 49: 0-0.
- Lima Neto, G.A., Kaffashi, S., Luiz, W., Ferreira, W., Dias da Silva, Y.S.A., Pazin, G., Violante, I. 2015. Quantification of secondary metabolites and antimicrobial and antioxidant activities of some medicinal plants from the Cerrado of the Mato Grosso. *Revista Brasileira de Plantas Mediciniais* 17: 1069-1077.
- Lovatto, P.B. 2012. A interação co-evolutiva entre insetos e plantas como estratégia ao manejo agroecológico em agroecossistemas sustentáveis. *Interciência* 37: 657-663.

- Macagnan, R., Macagnan, R., Werner, F., Rego, B.E.F., Barp, E.A. 2016. Eficácia de extratos vegetais no controle de *Spodoptera frugiperda* (JE SMITH, 1797) em milho. *Biosaúde* 14: 74-80.
- Machado, L.A., Silva, V.B., OLIVEIRA, M.D. 2007. Uso de extratos vegetais no controle de pragas em horticultura. *Arquivos do Instituto Biológico* 69: 03-106.
- Magrini, F.E., Specht, A., Gaio, J., Girelli, C.P., Miguez, I., Heinzen, H., Cesio, V. 2015. Antifeedant activity and effects of fruits and seeds extracts of *Cabralea canjerana* (Vell.) Mart.(Meliaceae) on the immature stages of the fall armyworm *Spodoptera frugiperda* (JE Smith)(Lepidoptera: Noctuidae). *Industrial Crops and Products* 65: 150-158.
- Mapeli, N.C., Santos, R.H.S., Casali, V.W.D., Cremon, C., Longo, L. 2010. Tabelas de fertilidade e esperança de vida de *Ascia monuste orseis* Latreille (Lepidoptera: Pieridae) alimentadas com couve-manteiga homeopatizada. *Revista Agrarian* 3: 82-193.
- Marangoni, C., Moura, N.F., Garcia, F.R.M. 2013. Utilização de óleos essenciais e extratos de plantas no controle de insetos. *Revista de Ciências Ambientais* 6: 92-112.
- Mata, R.F.F., Lomonaco, C. 2013. Toxicidade, deterrência e repelência de extratos aquosos de *Cabralea canjerana* ssp. *polytricha* (a. juss.) penn. (Meliaceae) sobre o curuquerê-da-couve *Ascia monuste orseis* (godart) (Lepidoptera: pieridae). *Revista Árvore* 37: 361-368.
- Matos, A., Nebo, L., Calegari, E.R., Batista-Pereira, L., Vieira, P., Fernandes, J., Rodrigues, R.R. 2006. Atividade biológica de extratos orgânicos de *Trichilia* sp (Meliaceae) sobre *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae) em dieta artificial. *BioAssay* 1: 1-6.
- Medeiros, C.A.M., Boiça Júnior, A.L. 2005. Efeito da aplicação de extratos aquosos em couve na alimentação de lagartas de *Ascia monuste orseis*. *Bragantia* 64: 633-641.
- Medeiros, C.A.M., Boiça Júnior, A.L., Robles Angelini, M. 2007. Efeito sub-letal de extratos vegetais aquosos de *Azadirachta indica* A. Juss. e *Sapindus saponaria* L. sobre aspectos biológicos de *Ascia monuste orseis* (Latreille)(Lepidoptera: Pieridae) em couve. *Boletín de Sanidad Vegetal Plagas* 33: 27-34.
- Morais, N., Oliveira Neto, F.B., Melo, A., Bertini, L., SILVA, F., ALVES, L. 2016. Phytochemical screening and antioxidant potencial evaluation of *Cnidocolus phyllacanthus* (Müll. Arg.) Pax & K. Hoffm from Apodi-RN. *Revista Brasileira de Plantas Medicinai* 18: 180-185.
- Numa, S., Rodríguez, L., Rodríguez, D., Coy-Barrera, E. 2015. Susceptibility of *Tetranychus urticae* Koch to an ethanol extract of *Cnidocolus aconitifolius* leaves under laboratory conditions. *SpringerPlus* 4: 1-10.
- Nunes, F.R.S., Dias, H.M.C., Cavalcante, G.M. 2016. Investigação das atividades antioxidante e antimicrobiana de duas espécies arbóreas ocorrentes no bioma caatinga. *Estação Científica* 6: 81-90.
- Obichi, E.A., Monago, C.C., Belonwu, D.C. 2015. Effect of *Cnidocolus aconitifolius* (Family Euphorbiaceae) aqueous leaf extract on some antioxidant enzymes and haematological parameters of high fat diet and Streptozotocin induced diabetic wistar albino rats. *Journal of Applied Sciences and Environmental Management* 19: 201-209.
- Paredes, P.F.M., Vasconcelos, F.R., Paim, R.T.T., Marques, M.M.M., Morais, S.M., Lira, S.M., Guedes, M.I.F. 2016. Screening of bioactivities and toxicity of *Cnidocolus quercifolius* Pohl. *Evidence-Based Complementary and Alternative Medicine* 2016: 1-9.
- Paula, A.C., Melo, K.M., Silva, A.M., Ferreira, D.A., Monte, F.J., Santiago, G.M., Silva, T.G. 2016. Constituintes químicos e atividade citotóxica de *Cnidocolus phyllacanthus*. *Revista Virtual de Química* 8: 231-241.
- Sobrinho, T.J.D.S.P., Saraiva, A.M., Almeida, D.M., Tavares, E.A., Pisciotano, M.N.C., Amorim, E.L.C. 2012. Phytochemical screening and antibacterial activity of four *Cnidocolus* species (Euphorbiaceae) against standard strains and clinical isolates. *Journal of Medicinal Plants Research* 6: 3742-3748.
- Pessoa, E.B., Almeida, F.A.C., Melo Silva, L.M. 2015. Bioatividade de três extratos de plantas no controle do *Z. abrotes subfasciatus* (Boh.). *Revista Cubana de Plantas Medicinales* 20: 0-0.
- Restello, R.M., Menegatt, C., Mossi, A.J. 2009. Efeito do óleo essencial de *Tagetes patula* L.(Asteraceae) sobre *Sitophilus zeamais* Motschulsky (Coleoptera, Curculionidae). *Revista Brasileira de Entomologia* 53: 304-307.
- Ribeiro, L.D.P., Santos, M.S., Gonçalves, G.L.P., Vendramim, J.D. 2015. Toxicity of an acetogenin-based bioinsecticide against *Diaphorina citri* (Hemiptera: Liviidae) and its parasitoid *Tamarixia radiata* (Hymenoptera: Eulophidae). *Florida Entomologist* 98: 835-842.
- Ribeiro, L.D.P., Ansante, T.F., Vendramim, J.D. 2016. Effect of ethanolic extract from *Annona mucosa* seeds on development and feeding behavior of *Spodoptera frugiperda*. *Bragantia*

75: 322-330.

Silva, H.H.G.D., Silva, I.G.D., Santos, R.M.G.D., Rodrigues Filho, E., Elias, C.N. 2004. Larvicidal activity of tannins isolated of *Magonia pubescens* St. Hil. (Sapindaceae) against *Aedes aegypti* (Diptera, Culicidae). *Revista da Sociedade Brasileira de Medicina Tropical* v: 396-399.

Silva, L., Carvalho, G., Silva, L., Pavan, B., Peres, M., Santos Almeida, M.L. 2014. Mortalidade e comprometimento do desenvolvimento do caruncho do feijão (*zabrotes subfasciatus*), induzido pelo extrato de sangra d'água (*croton urucurana*). *Comunicata Scientiae* 5: 331-338.

Souza, A.J. 2014. *Estudo fitoquímico e atividade biológica in vitro de Cnidoscolus urens L. (Arthur) (EUPHORBIACEAE). 99f. Dissertação (Mestrado em Recursos Naturais do Semiárido) - Universidade Federal do Vale do São Francisco, Petrolina, Brasil.*

Trindade, R.C.P., Marques, I.M.R., Xavier, H.S., Oliveira, J.D. 2000. Extrato metanólico da amêndoa da semente de nim e a mortalidade de ovos e lagartas da traça-do-tomateiro. *Scientia Agricola* 57: 407-413.

Trindade, R., Ferreira, E., Gomes, I., Silva, L., Sant'ana, A.E.G., Broglio, S., Silva, M. 2015. Extratos aquosos de inhame (*Dioscorea rotundata* Poir.) e de mastruz (*Chenopodium ambrosioides* L.) no desenvolvimento da lagarta-do-cartucho-do-milho *Spodoptera frugiperda* (JE Smith, 1797). *Revista Brasileira de Plantas Mediciniais* 17: 291-296.

Valenzuela Soto, R., Morales Rubio, M.E., Verde Star, M.J., Oranday Cárdenas, A., Preciado-Rangel, P., Antonio González, J., Esparza-Rivera, J.R. 2015. *Cnidoscolus chayamansa* hidropónica orgánica y su capacidad hipoglucemiante, calidad nutraceutica y toxicidad. *Revista Mexicana de Ciencias Agrícolas* 6: 815-825.

Wagner, H., Bladt, S. 2001. *Plant drug analysis*. 2ª ed. New York: Springer, 384p.

Wiesbrook, M.L. 2004. Natural indeed: Are natural insecticides safer and better than conventional insecticides? *Illinois Pesticide Review* 17: 1-8