

Insecticide activity of *Copaifera langsdorffii* oil in *Aphis craccivora* (Hemiptera: Aphididae)

Actividad insecticida del aceite de *Copaifera langsdorffii*
en *Aphis craccivora* (Hemiptera: Aphididae)

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SUMMARY

In this study, the insecticide activity of *Copaifera langsdorffii* oil in the control of *Aphis craccivora* was evaluated under laboratory conditions. A completely randomized experimental design was used, with five concentrations of 2 %, 4 %, 6 %, 8 % and 10 %, blank test treatment (distilled water + Tween®) and control (Orthene®). In the evaluation, adults of *Aphis craccivora* and *Gliricidia sepium* were exposed to the application of 1 mL of the water mixture with different concentrations of oil. Mortality (%) was the variable used for the analysis, being evaluated at 4, 24, 48 and 72 hours after application of the treatments through the analysis of variance and determination of the median lethal concentration (LC₅₀). It was observed that with the increase in concentrations there was increase in the mortality of *Aphis craccivora*, with higher mortality in the concentration of 10 % in the periods of 4 and 24 hours (88 % and 90 %) and from 6 %, with mortality ranging from 80 to 94 % (after 48 hours) and 92 to 98 % (after 72 hours). The LC₅₀ calculated at times of 4, 24, 48 and 72 hours was: 6.16 %, 4.65 %, 4.22 % and 2.99 %, respectively. Copaiba oil has insecticidal activity and concentration of 6 % can be indicated for the control of *Aphis craccivora*.

Key words: copaiba, insect pest, aphids, natural product.

RESUMEN

En este trabajo se evaluó la actividad insecticida del aceite de *Copaifera langsdorffii* en el control de *Aphis craccivora*, en condiciones de laboratorio. Se utilizó un diseño completamente aleatorizado, con cinco concentraciones 2 %, 4 %, 6 %, 8 % y 10 %, tratamiento control (agua destilada + Tween®) y control (Orthene®). En la evaluación, se recolectaron adultos de *Aphis craccivora* en *Gliricidia sepium*, y se expusieron a la aplicación de 1 mL de la mezcla de agua con las diferentes concentraciones del aceite. La mortalidad (%) fue la variable utilizada para el análisis, realizado a las 4, 24, 48 y 72 horas después de la aplicación de los tratamientos, mediante análisis de varianza y determinación de la concentración letal mediana (CL₅₀). Se observó que con el aumento de las concentraciones se produjo un incremento en la mortalidad de *Aphis craccivora*, presentándose una mayor mortalidad (88 % y 90 %) en la concentración del 10 % en los períodos de 4 y 24 horas y del 6 % con una mortalidad que varió de 80 a 94 % (después de 48 horas) y 92 a 98 % (después de 72 horas). La CL₅₀ calculada a las 4, 24, 48 y 72 horas fue del 6,16 %, 4,65 %, 4,22 % y 2,99 %, respectivamente. El aceite de copaiba tiene actividad insecticida y una concentración del 6 % está indicado para el control de *Aphis craccivora*.

Palabras clave: copaiba, insectos-plaga, pulgones, producto natural.

INTRODUCTION

Natural products from plants may be of potential interest against insects, as they do not present significant adverse effects on environment, among other damages that a chemical insecticide could cause (Lagunes y Rodríguez 1991). Thus, due to these factors, the demand for alternative pesticides has become increasing for pest insect control.

Studies involving alternative forms of aphid control have been developed, such as those that use essential oils, and have been shown to be efficient for the control of some aphid species, for example, orange oil in the control of aphid *Hyadaphis foeniculi* Passerini (Lopes *et al.* 2009) and essential oil of *Ageratum conyzoides* L. for the control of *Macrosiphum euphorbiae* (Thomas) (Soares *et al.* 2011). Among the compounds derived from plants, essential oils stand out in the control of insects (Isman 2000),

and among essential oils, *Copaifera langsdorffii* Desf oil. (Fabaceae) has high potential for this purpose, being used by native peoples for many years, presenting use also in folk medicine. *Copaifera langsdorffii*, popularly known as copaiba, is a tree of paramount importance, potential source of inhibiting compounds to be used as antimicrobials in treatments for humans and animals and conservation of food (Pieri *et al.* 2012).

Aphids are among the groups of insect pests that attack both agricultural and ornamental crops and forests (Hemiptera: Aphididae). Aphids attack various crops and cause significant damage due to intense sap suction. Strategies for the control of aphids, as well as other insect pests in agricultural and forest crops, have been increasingly limited, making alternative systems necessary to maintain agricultural productivity with as little environmental impact as possible (Altieri 2004). Therefore, researchers have increasingly sought alternatives to conventional insecticides through the use of natural insecticides, without causing damage to biodiversity, environment and human health. In addition, natural insecticides have proved to be a more economical, ecologically beneficial and biodegradable alternative (Regnault-Roger 1997).

In this context, the use of copaiba oil can be a promising alternative for aphids control in agroecological systems, especially in food-plant crops that are frequently attacked by this group of insects, having the possibility of control with a natural product, biodegradable and produced by small farmers, thus promoting increased income for oil producers. Hence, this study aimed at evaluating the insecticidal activity of *C. langsdorffii* oil in the control of *Aphis craccivora* Koch (Hemiptera: Aphididae), under laboratory conditions.

METHODS

The study was conducted in the Center for Research and Technology of the Southern Amazon (CEPTAM) of the State University of Mato Grosso - UNEMAT, in Alta Floresta, MT. The adult aphids of *A. craccivora* were collected from natural infestation in the forest species *Gliricidia sepium* (Jacq.) Kunthe Walp. (Fabaceae), and the attack was mainly verified in the flowers of plants of this species located in Alta Floresta-MT. Individuals with an approximate size of 2 mm, corresponding to the adult state, were selected. Flowers infested by *A. craccivora* were collected and the aphids were carried to the laboratory for the bioassay installation.

The concentrations (treatments) evaluated were: 2 %, 4 %, 6 %, 8 % and 10 %. A blank test treatment was performed, which consisted of distilled water with Tween® 20 at the concentration of 0.5 % and the insecticide Orthene® 750 BR (organophosphate) as a reference insecticide. Tween® 20 was used to homogenize the mixture of water and oil.

The bioassay was conducted in Petri dishes, and in each of them, 10 adult insects were placed on a leaf of the species *G. sepium* containing in its petiole a piece of cotton soaked in water to avoid leaf dryness during the evaluation period of the experiment and double filter paper under the leaf, also to avoid dryness.

With the use of a thin-tipped brush, the adult aphids were placed in the Petri dish on the sheet and, afterwards, the different concentrations of the oil were sprayed with the aid of a manual spray, and a milliliter of the solution was sprayed on the aphids. Subsequently, the Petri dishes containing the aphids were placed in a biochemical oxygen demand chamber (B.O.D), with temperature of 25 °C and photoperiod of 12 hours. Evaluations were performed at 4, 24, 48 and 72 hours after the application of the treatments. The mortality of adults of *A. craccivora* was quantified with the aid of a magnifying glass, and the aphid was considered dead when, touched by a thin brush, showed no movement.

The experiment was conducted in a completely randomized experimental design, with five replications, and each experimental unit consisted of ten aphids. Data were analyzed by the Shapiro-Wilk normality test and were submitted to a variance analysis. Means were compared by the Scott Knott test ($P < 0.05$), using the ExpDes.pt package (Ferreira *et al.* 2013) of version 3.4 of the R software (R Development Core Team 2017). Lethal concentrations (LC_{50}) were calculated for each period evaluated by a Probit analysis, using the ecotoxicology package (Gamma 2015) of software R, with mortality correction performed by the Abbott formula (1925) [Equation 1]:

$$M_{corr} = \frac{MT - MC}{100 - MC} * 100 \quad [\text{Equation 1}]$$

where: M_{Corr} = corrected mortality (%), MT = treatment mortality (%) and MC = control mortality (%).

RESULTS

The average accumulated mortality of *A. craccivora* submitted to concentrations of 2, 4, 6, 8 and 10 % of copaiba oil in the periods of 4, 24, 48 and 72 hours after application can be observed in table 1. It was verified that copaiba oil presented an insecticide effect for *A. craccivora* in all concentrations evaluated, except at concentrations of 2 % and 4 % of the first evaluation (4 hours) (table 1).

It was observed that at the concentration of 10 % there was higher mortality of adult aphids of *A. craccivora* in the periods of 4 h and 24 h with mortality of 88 % (4 h), reaching 90 % (24 h), not differing significantly from the reference treatment. It was also verified that in the period of 4 hours the mortality was above 46 % from the concentration of 6 %. In the observations at 48 h and 72 h, there was no significant difference in concentrations from 6 %, with mortality from 80 to 94 % and 92 to 98 %, respecti-

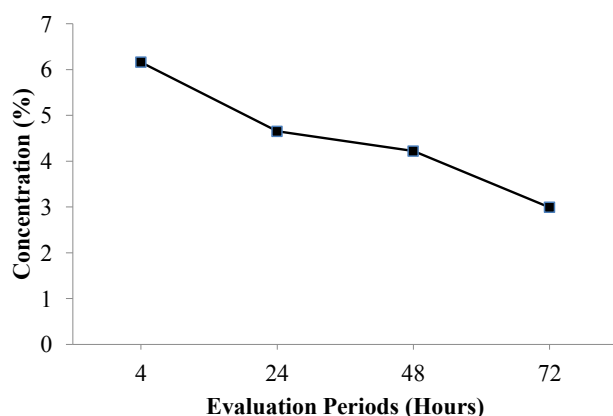
Table 1. Cumulative average percentage of dead adults of *A. craccivora* and relative efficiency, in corrected percentage of Abbott, with copaiba oil.Porcentaje medio acumulado de adultos muertos de *A. craccivora* y eficacia relativa, en porcentaje corregido de Abbott, con aceite de copaiba.

Treatment	Mortality (%)			
	4h	24h	48h	72h
Water + Tween20® 0,5 %	0C	4D	10C	24D
2 %	14C	20C	26B	46C
4 %	18C	24C	34B	70B
6 %	54B	74B	80A	92A
8 %	46B	82B	94A	96A
10 %	88A	90A	90A	98A
Reference insecticide	88A	100A	100A	100A
CV(%)	34.89	24.3	17.67	15.34

Means followed by the same capital letter in the column do not differ statistically from each other by the Scottknot test ($P < 0.05$).Las medias seguidas de la misma letra mayúscula en la columna no difieren estadísticamente entre sí por la prueba de Scottknot ($P < 0,05$).

vely, not differing from the reference treatment (chemical insecticide).

It was noted that LC_{50} calculated for copaiba oil in the 4-hour time interval was 6.16 %. It is observed that even after a short period of exposure of *A. craccivora* to the oil, the mortality of individuals was above 50 % already in the first evaluation (figure 1). LC_{50} , calculated for the 24-hour interval, was 4.65 %, and after 48 hours LC_{50} was 4.22 %, demonstrating that as the exposure period increased there was a reduction in the median lethal concentration (LC_{50}), with the lowest LC_{50} of 2.99 % in the 72-hour period.

**Figure 1.** LC_{50} values for the different periods validated after application of Copaiba oil in *A. craccivora*.Valores LC_{50} para los diferentes periodos validados tras la aplicación de aceite de Copaiba en *A. craccivora*.

DISCUSSION

Lima *et al.* (2008), testing essential oil compositions of *Illicium verum* L. and *Cymbopogon citratus* DC., in the aphid *Brevicoryne brassicae* L., verified that the essential oils of both botanical species have a repellent effect at concentrations of 0.1 % and 0.5 %, respectively. Thus, some essential oils even in smaller concentrations already have insecticidal activity for certain insects, including aphids. As verified in this study that after 24 h the lowest concentration had an effect on the mortality of *A. craccivora* when compared to the blank test.

Coitinho *et al.* (2006), evaluating different oils in the control of *Sitophilus zeamais* Motsch., observed that copaiba oil caused a reduction of 96.4 % in the emergence and 87.7 % in the repellency of adults of *S. zeamais*. According to these authors, repellency is an important property to be considered in pest control with vegetable oils in general, because the higher the repellency, the lower the infestation, resulting in the reduction or suppression of oviposition and, consequently, the number of insects emerged.

According to Freire *et al.* (2006), the oils of *Carapa guianensis* Aubl. or *C. langsdorffii* can be used for the treatment of phorid-infested hives (Diptera: Phoridae), and can also be used to prevent infestation. These authors studying the oils mentioned in the control of phorids, observed that when the oil of *C. guianensis* or *C. langsdorffii* mixed with pollen was used as substrates, they demonstrated a repellent effect, inhibiting up to 100 % of the oviposition.

The insecticide activity of copaiba oil may be related to compounds already isolated or detected, which demonstrate insecticide properties. Among these, the one that was

more studied and was active in a larger number of trials was β -caryophyllene, which is described as repellent in literature (Keeler and Tu 2011, Jacobson 2008). Furthermore, on the compounds, essential oils in general have monoterpenes and phenols as main constituents, and monoterpenes affect both the biology and behavior of various insects (Isman 2000, Viegas Junior 2003).

Essential oils can act in several ways: (i) by obstructing the spiracles, thus causing the insect to die from asphyxiation; (ii) by acting on digestive enzymes; (iii) in neurological function; and (iv) in the insect tegument (Soares *et al.* 2011). However, there is also an additional effect caused by the obstruction of olfactory receptors, interfering in the location of hosts, thus affecting the behavior and resulting in repellency for adult aphids and nymphs (Butler Junior *et al.* 1989, Larew and Locke 1990, Vincent *et al.* 2003).

Copaiba oil showed higher effect (with LC_{50} of 4.22 % after 48 hours) when compared with hydroethanolic extract of *Annona montana* Macfad in a study conducted by Bandeira *et al.* (2017), which obtained LC_{50} of 7.69 % after 48 hours of application, also on *A. craccivora*, in a greenhouse.

Other studies have evidenced the efficiency of vegetable oils in insect control as in research developed by the authors Furtado *et al.* (2005), who evaluated the larvicidal activity of essential oils against *Aedes aegypti* L. (Diptera: Culicidae). These authors observed that the essential oil of *Vanillosmopsis arborea* Baker induced the highest larvicidal activity, with LC_{50} of 15.91 mg/ml. Besides this, other species also presented larvicidal potential, such as *Lippia sidoides* Cham., which presented LC_{50} of 45.49 mg/ml, *Cymbopogon winterianus* Jowitt essential oil with LC_{50} of 54.69 mg/ml, and essential oil of *Ageratum conyzoides* L. with LC_{50} of 61.55 mg/ml. Fazolin *et al.* (2007) found LC_{50} value of 1.51 ml for *Tanaecium nocturnum* (Barb. Rodr.) Bur. & K. Shum in the control of *Tenebrio molitor* L., evidencing those vegetable oils are efficient in controlling different insect pests.

The use of this natural product can provide an alternative for aphids control, as it allows the control of this group of insect pests, without being harmful to the environment and in an efficient way.

CONCLUSIONS

Copaiba oil showed insecticidal activity in *A. craccivora*, leading to mortality above 92 % at concentrations equal to or higher than 6 % after 72 hours of application, thus proving to be efficient for the control of adults of *A. craccivora* in laboratory tests.

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Received: 29.03.21

Accepted: 10.03.22

