

## Essential Oils in the Control of *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae)

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### ABSTRACT

*Ceratitis capitata* (Wiedemann) causes significant economic losses to Brazilian fruit crops. In this context, and given the need to develop sustainable strategies for controlling this pest, the study of the insecticidal potential of essential oils has gained worldwide prominence. Thus, this study aimed to evaluate the effectiveness of the essential oils of *Carapa guianensis* Aublet, *Cymbopogon winterianus* Jowitt, and *Eucalyptus citriodora* Hook in the mortality and emergence of *C. capitata* in two life stages. The research was conducted at the Entomology Laboratory of the Agricultural Sciences Center at the Federal University of Paraíba, Brazil. The essential oils used were derived from commercial formulations. For each type of oil, 3rd instar larvae and pupae were immersed in five concentrations. The action of the oils varied depending on the life stages in which they were applied, and their effects were more pronounced during the initial hours post-application. The use of *C. guianensis* oil, when applied during the pupal stage at a concentration of 37.6 mg.ml<sup>-1</sup>, resulted in 75% mortality of individuals (LD75). For larval applications, *E. citriodora* oil stood out, producing the highest larval mortality rates and greater persistence of action. The use of essential oils in the control of *C. capitata* proved to be a promising alternative, but attention should be given to the life stage and applied concentrations to optimize the success of this control method.

**Keywords:** Alternative control, Natural insecticide, Mediterranean fruit fly.

## Óleos Essenciais no Controle de *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae)

### RESUMO (PT-BR)

*Ceratitis capitata* (Wiedemann) é responsável por grandes prejuízos econômicos às frutíferas brasileiras. Neste sentido e dada a necessidade de desenvolver estratégias sustentáveis de controle desta praga, o estudo do potencial inseticida dos óleos essenciais tem ganhado destaque a nível mundial. Assim, o presente estudo teve como objetivo avaliar a eficiência dos óleos essenciais de *Carapa guianensis* Aublet, *Cymbopogon winterianus* Jowitt e *Eucalyptus citriodora* Hook na mortalidade e emergência de *C. capitata* em duas fases de vida. A pesquisa foi realizada no Laboratório de Entomologia do Centro de Ciências Agrárias da Universidade Federal da Paraíba, Brasil. Os óleos essenciais utilizados foram oriundos de formulações comerciais. Para cada tipo de óleo, as larvas e pupas de 3º instar foram imersas em cinco concentrações. A ação dos óleos diferiu para as fases da vida em que foram aplicados, bem como suas ações foram mais efetivas nas primeiras horas após a aplicação. A utilização do óleo de *C. guianensis*, quando aplicado na fase pupal na concentração de 37,6 mg.ml<sup>-1</sup> foi responsável por matar 75% dos indivíduos (DL75). Para aplicações na fase larval, o óleo de *E. citriodora* se destacou dos demais, gerando as maiores taxas de mortalidade de larvas e com maior persistência de ação. A utilização de óleos essenciais no controle de *C. capitata* mostrou-se uma alternativa promissora, porém deve-se atentar para o estágio de vida e concentrações aplicadas para otimizar o sucesso deste método de controle.

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**Palavras-Chaves:** Controle alternativo, Inseticida natural, Mosca-do-Mediterrâneo.

## Aceites Esenciales en el Control de *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae)

### RESUMEN

*Ceratitis capitata* (Wiedemann) es responsable de grandes perjuicios económicos a los frutales brasileños. En este sentido y dada la necesidad de desarrollar estrategias sostenibles para el control de esta plaga, el estudio del potencial insecticida de los aceites esenciales ha ganado protagonismo a nivel mundial. Así, el presente estudio tuvo como objetivo evaluar la eficacia de los aceites esenciales de *Carapa guianensis* Aublet, *Cymbopogon winterianus* Jowitt y *Eucalyptus citriodora* Hook sobre la mortalidad y emergencia de *C. capitata* en dos fases de vida. La investigación se realizó en el Laboratorio de Entomología del Centro de Ciencias Agrarias de la Universidad Federal de Paraíba, Brasil. Los aceites esenciales utilizados fueron provenientes de formulaciones comerciales. Para cada tipo de aceite, las larvas y pupas de 3º estadio fueron sumergidas en cinco concentraciones. La acción de los aceites varió según la fase de vida en la que fueron aplicados, así como también sus efectos fueron más efectivos en las primeras horas tras la aplicación. El uso del aceite de *C. guianensis*, cuando se aplicó en la fase pupal a la concentración de 37,6 mg.ml<sup>-1</sup>, fue responsable de la muerte del 75% de los individuos (DL<sub>75</sub>). Para aplicaciones en la fase larval, el aceite de *E. citriodora* se destacó sobre los demás, generando las mayores tasas de mortalidad larval y con mayor persistencia de acción. El uso de aceites esenciales en el control de *C. capitata* se mostró como una alternativa prometedora, sin embargo, se debe prestar atención a la etapa de vida y a las concentraciones aplicadas para optimizar el éxito de este método de control.

**Palabras clave:** Control alternativo, Insecticida natural, Mosca del Mediterráneo.

### 1. Introduction

Fruit farming is one of the main agricultural activities worldwide, with significant growth in the consumption markets for fresh fruits and industrial processing in recent years (Fachi et al., 2019). Brazil is the third-largest fruit producer globally, behind only China and India, with an annual production of about 43 million tons (Freitas et al., 2018) in an area of approximately 2 million hectares (Pio et al., 2019). The country stands out for producing both tropical and temperate fruits year-round, attributed to its territorial extension, geographical position, and edaphoclimatic conditions (Fachi et al., 2019).

Pests are one of the significant challenges for fruit producers worldwide. Among pest insects, frugivorous dipterans, particularly some species of the Tephritidae family, stand out (Oliveira et al., 2019; Lopes et al., 2023). Around 250 of the 4,000 known species in this family infest cultivated fruits, causing substantial economic losses (Bekker et al., 2019). Among Tephritidae, fruit fly species such as *Anastrepha* Schiner, *Ceratitis capitata* (Wiedemann), and *Bactrocera carambolae* Drew & Hancock are considered the most important fruit crop pests in Brazil, representing a severe threat to this productive sector (Malacrino et al., 2018).

*Ceratitis capitata* is the main fruit fly species in Brazil, requiring strict population control of this pest as a prerequisite for fruit exports to the United States and Japan (Leite et al., 2019). This species is highly invasive and polyphagous, attacking a wide variety of fruits (Sciarretta et al., 2019; Elqdhly et al., 2024), due to its adaptability, which allows it to occupy increasingly more niches in Brazilian territory (Leite et al., 2019).

The losses caused by *C. capitata* are primarily due to the reduced commercial yield of orchards, resulting from the damage incurred when larvae feed on the fruit pulp, devaluing its quality and inducing premature fruit drop. Secondary damage is also common, such as bacterial and/or fungal infections arising from openings created by the pest's oviposition (Asadi et al., 2019).

Control of *C. capitata* is commonly achieved through the application of insecticides, such as organophosphates and pyrethroids (Brilinger et al., 2019; Harter et al., 2024). However, with growing restrictions on chemical control in fruit crops and increased awareness of food safety (Paranhos et al., 2019), there is an urgent need to implement new control strategies as alternatives to chemical insecticides. These alternatives aim to protect the environment, reduce resistance, increase crop productivity, and lower high levels of toxic residues in food (Hernández-Cruz et al., 2019).

The use of natural insecticides is a safer control strategy compared to chemical products. Three forms are commonly used for insect control: extracts, vegetable oils, and essential oils (Hidayat et al., 2018). Essential oils are compounds found in many plant families and are characterized by low molecular weight, lipophilic nature (Rizzo et al., 2020), and a relatively narrow range of volatilities, mainly monoterpenes and sesquiterpenes (Niogret; Epsky, 2018; Sarri et al., 2024). These oils or their derived chemical compounds can attract beneficial insects or repel undesirable pests. For example, they can be toxic to eggs, larvae, pupae, and adults of various economically significant insect species (Oviedo et al., 2018).

The use of essential oils in controlling *C. capitata* has proven to be an efficient and promising technique (Papanastasiou et al., 2017; Oviedo et al., 2018; Alves et al., 2019), requiring further studies to analyze the efficacy of essential oils from other plant species against this pest. Thus, the present study aimed to evaluate the efficiency of essential oils from three plant species by analyzing their insecticidal action in controlling *Ceratitis capitata*.

## 2. Material and Methods

The experiments were conducted in two stages: in the first, the essential oils and the adjuvant Tween® were prepared by dilution in distilled water. In the second, the oil concentrations were applied to 3rd instar larvae and pupae of *C. capitata*.

Adult flies were kept in 20x30x20 cm cages covered with voile fabric, placed on trays containing water to collect eggs. The adults were fed daily with a 10% honey-water solution via a cotton swab placed on top of the cage during the oviposition period. Eggs collected were placed on the artificial diet in 20x10x5 cm plastic containers, which were kept in the laboratory. Egg infestation in the diet occurred 24 to 48 hours after collection. Approximately 10 days after infestation, the containers with larvae were transferred to 34x34x14 cm trays containing sterilized sand to obtain the pupae, which were later placed inside the cages to restart the rearing cycle.

Commercial formulations of vegetable oils with insecticidal activity were used for the mortality tests on *C. capitata*. The oils tested included andiroba (*Carapa guianensis* Aublet), citronella (*Cymbopogon winterianus* Jowitt), and eucalyptus (*Eucalyptus citriodora* Hook) at concentrations of 0 (control), 20, 40, 60, 80, and 100 mg.mL<sup>-1</sup>. For the control group, only distilled water and the Tween® adjuvant were used.

For each oil concentration, 50 fruit fly individuals were tested, with a total of 10 replicates consisting of 5 specimens each for both 3rd instar larvae and pupae. The insects were immersed in each concentration for 10 seconds. After treatment, larvae and pupae were transferred to Petri dishes containing 15 g of sterilized sand.

For 3rd instar larvae, the toxicity of the essential oils was evaluated at intervals of 6, 12, 24, 48, 72, 96, 120, and 144 hours post-application, by counting the number of dead larvae. For larvae that reached the pupal stage, emergence (number of males and females) was assessed from the 8th to the 10th day, considering that the minimum pupal development period for adult formation is 7 days.

Pupal assessment was conducted from the 7th to the 9th day after product application, the pre-established time for normal adult emergence. Pupal mortality was determined by the lack of adult emergence.

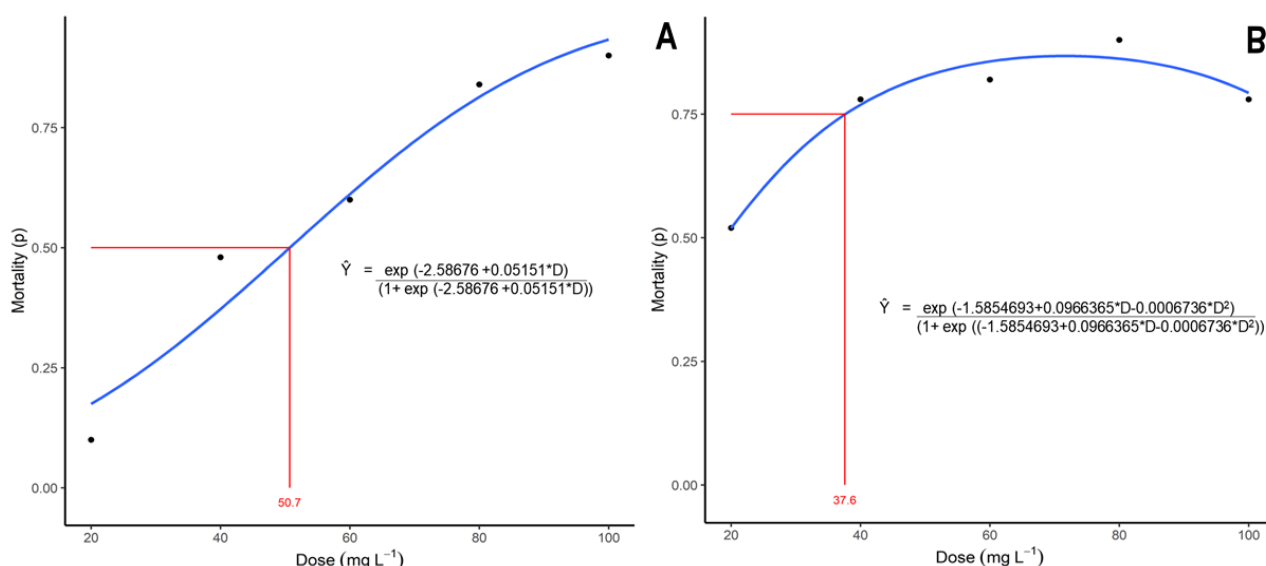
The experimental design was completely randomized, and the results of the trials were subjected to Probit analysis and variance analysis using the F-test. The software used was R (R Development Core Team, 2024).

### 3. Results

When applied directly to the pupae, a significant interaction ( $p \leq 0.0001$ ) was observed between the essential oils used and their respective concentrations on the mortality of *C. capitata* pupae. However, this effect was not observed with citronella oil, demonstrating its low efficacy for controlling this pest at this life stage.

An increase in the concentrations of eucalyptus oil showed a linear effect on pupal mortality, with the LD50 obtained at a concentration of  $50.7 \text{ mg.mL}^{-1}$  (Figure 1A), indicating the need for higher concentrations of this oil for effective control of *C. capitata*.

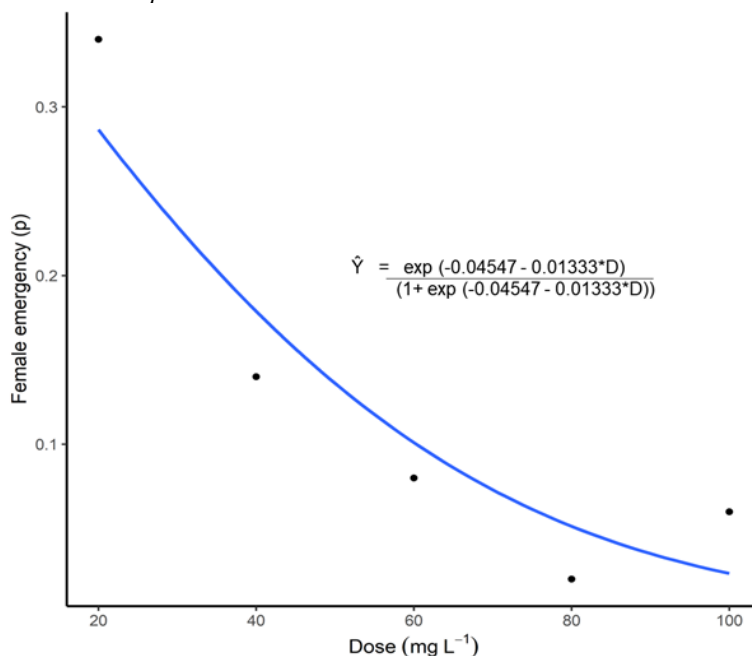
**Figure 1** - LD50 and mortality of *C. capitata* pupae treated with different concentrations of eucalyptus oil (A), and LD75 and mortality of *C. capitata* pupae treated with different concentrations of andiroba oil (B).



Source. Authors (2025).

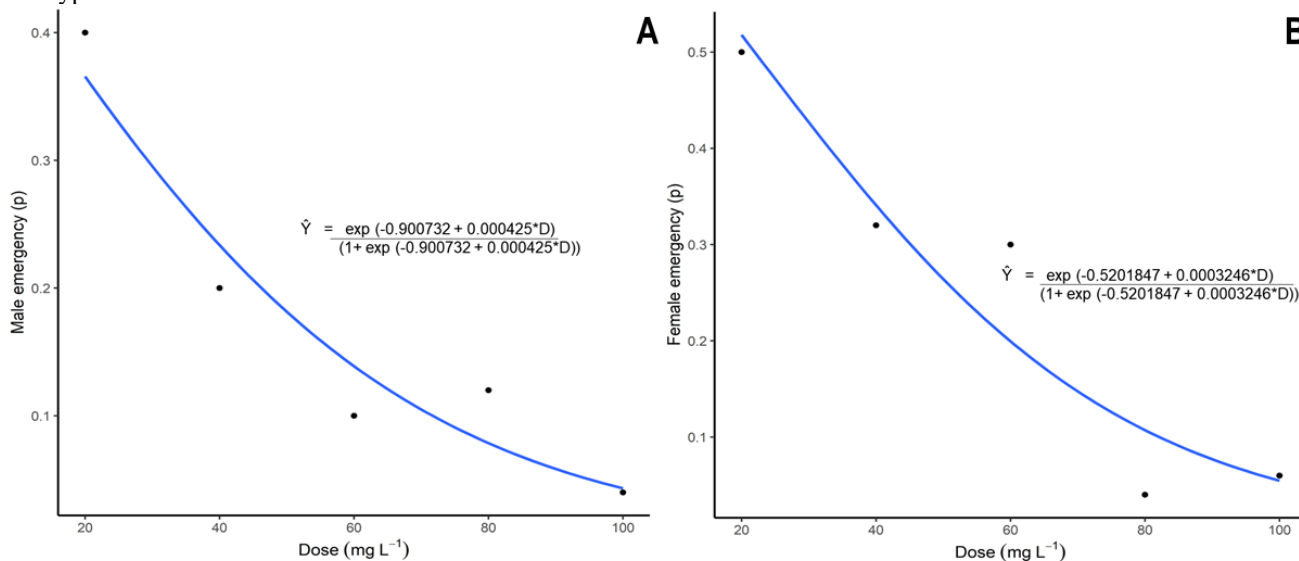
For the use of andiroba oil (Figure 1B), a quadratic effect of the concentrations on pupal mortality was observed. The concentration of  $37.6 \text{ mg.mL}^{-1}$  resulted in 75% mortality of the individuals (LD75).

Regarding andiroba oil, a significant effect ( $p \leq 0.001$ ) of the applied concentrations on the emergence of female *C. capitata* was observed. The increase in concentrations led to a reduction in the emergence of new female insects (Figure 2).

**Figure 2** - Emergence of female *C. capitata* individuals treated with different concentrations of andiroba oil.

Source. Authors (2025).

Eucalyptus oil, when applied during the pupal stage, resulted in a higher emergence of females compared to males (Figures 3A and 3B). However, both males and females showed a decrease in emergence as the concentrations increased, due to the high mortality caused by this oil. In contrast, citronella oil did not significantly affect the emergence of either female or male *C. capitata* individuals ( $p > 0.05$ ).

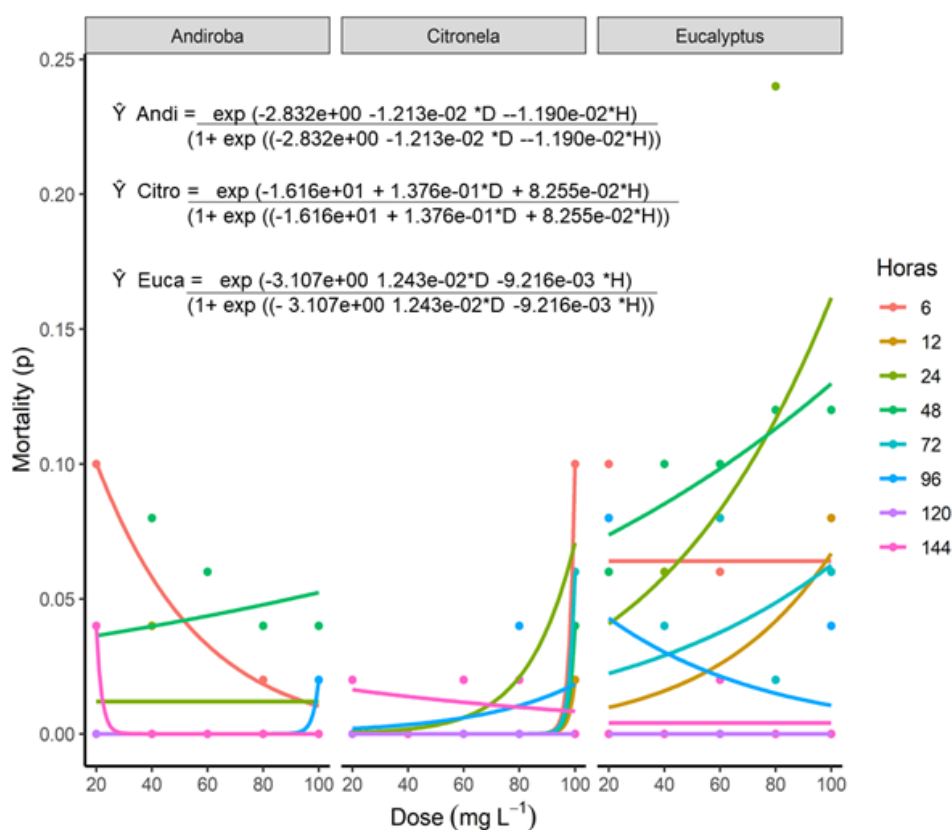
**Figure 3** - Emergence of male (A) and female (B) *C. capitata* individuals treated with different concentrations of eucalyptus oil.

Source. Authors (2025).

For Andiroba oil, the minimum concentration (20 mg.ml<sup>-1</sup>) over the first 6 hours showed the highest larval mortality (Figure 4). A similar result was observed with Citronella oil, where the highest mortality values were also observed during the first 6 hours, though these values were obtained with the maximum dosage (100 mg.ml<sup>-1</sup>). Conversely, in the 24-hour period with the maximum dosage, Eucalyptus oil exhibited the highest mortality, making it the most effective oil for pest control at this stage.

For Andiroba oil, the minimum concentration (20 mg.ml<sup>-1</sup>) over the first 6 hours showed the highest larval mortality (Figure 4). A similar result was observed with Citronella oil, where the highest mortality values were also observed during the first 6 hours; however, these values were obtained with the maximum dosage (100 mg.ml<sup>-1</sup>). Conversely, the 24-hour period with the maximum dosage resulted in the highest mortality for Eucalyptus oil, making this oil more effective for pest control at this stage.

**Figure 4** - Larval mortality of *C. capitata* treated in the 3rd instar with different concentrations of Andiroba, Citronella, and Eucalyptus oils over time.

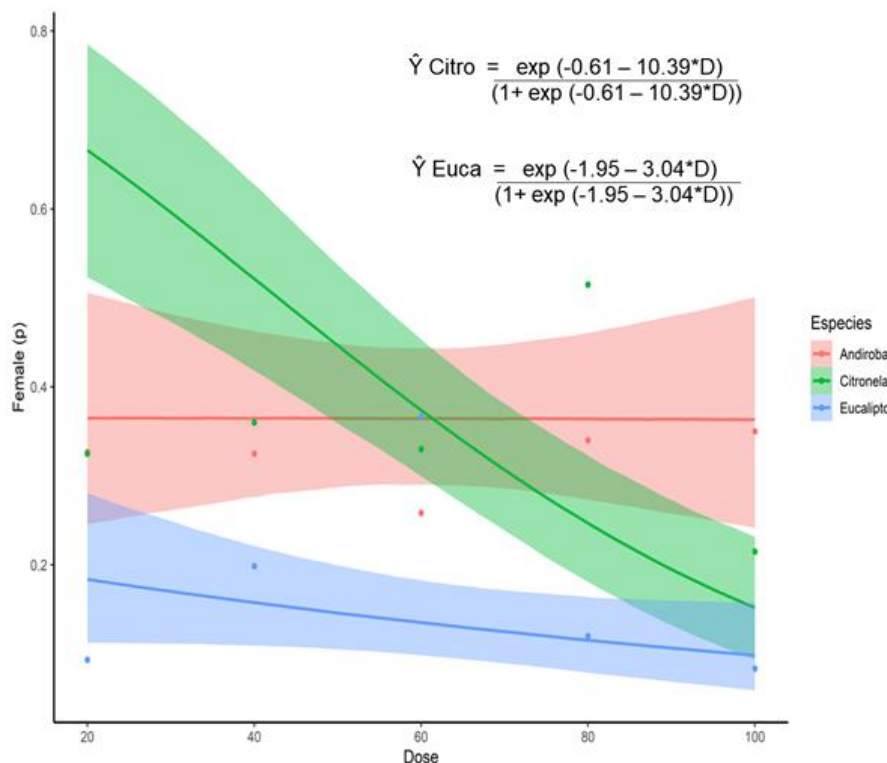


Source. Authors (2025).

For pupal mortality derived from previously treated larvae, no significant effect ( $p \leq 0.001$ ) was identified between the essential oils used and their respective concentrations on *C. capitata* mortality.

Regarding female emergence, a significant effect ( $p \leq 0.001$ ) was observed for citronella and copaíba oils. Increasing the concentrations of these essential oils decreases the probability of insect emergence of this sex (Figure 5), which represents a good control alternative since a lower number of female individuals will result in less oviposition inside the fruits, consequently leading to fewer fruits damaged by the pest.

**Figure 5** - Female insect emergence of *C. capitata* from pre-treated larvae with different concentrations of Andiroba, Citronella, and Eucalyptus oils.



For male emergence, as with female emergence, no significant effect ( $p > 0.05$ ) was observed for the essential oils analyzed.

#### 4. Discussion

In this study, we demonstrated that the essential oils of Andiroba, Citronella, and Eucalyptus have diverse effects on the emergence and mortality of larvae and pupae of *Ceratitis capitata*. This result is due to the great variability of chemical components present in these oils. Studies such as that of Nhan et al. (2020) show that citronella oil is mainly composed of citral (46.603%), limonene (34.983%), and geraniol (3.556%). Andiroba oil, on the other hand, predominantly contains three components:  $\alpha$ -humulene, bicyclogermacrene, and germacrene-D, which make up more than 50.00% of its composition (Zórtea et al., 2017). Eucalyptus essential oil predominantly contains Eucalyptol,  $\alpha$ -Pinene, and  $\beta$ -Pinene (Adak et al., 2020).

The high mortality of pupae caused by andiroba oil when applied to this structure is likely due to the high number of insecticidal components present in the oil, which, although not primary constituents, exhibit significant insecticidal action, such as triterpenes, coumarins, flavonoids, and linoleic acid (Sarria et al., 2011). Insecticidal effects of andiroba oil have also been reported for *Anastrepha fraterculus*, another economically significant fruit fly species (Nunes et al., 2015; Brilinger et al., 2019). Our results further indicate that the components present in andiroba oil have a greater ability to penetrate the protective layer of pupae, leading to higher mortality rates.

Citronella oil is well-known for its insecticidal effect (Saad et al., 2017; Brilinger et al., 2019), with its main constituent being a monoterpene, citral, which, although highly toxic (Kaur et al., 2019), has a rapid

volatilization rate (Silva et al., 2019), making it an inefficient alternative for application during the pupal stage.

The application of essential oils affects the growth, fertility, and development of insects (Sehari et al., 2018), which in this study can be confirmed by the influence on the emergence of female flies, caused by the application of eucalyptus and andiroba oils, and on male flies when eucalyptus oil was applied during the pupal stage. This negative influence on the emergence of new female flies is extremely beneficial from a biological control perspective, as female insects are responsible for ovipositing in fruits, causing significant economic damage (Hafsi et al., 2020).

When applied during the larval stage, essential oils exhibited different behaviors in terms of larval mortality over time, with andiroba and citronella oils showing pronounced effects mainly in the initial hours. This is due to the fact that most components of insecticidal oils have high volatility (Lee et al., 2015), with citronella oil exhibiting even more significant effects, where the highest mortality rates were observed in the first six hours with the maximum concentration, reinforcing the use of alternative application strategies, such as microencapsulation (Pavoni et al., 2020).

Eucalyptus oil exhibited a more persistent effect, with its potential increasing with higher concentrations applied. This is partly due to its composition, particularly Eucalyptol, which plays a significant role in its insecticidal action and also synergizes with other compounds present in the oil to enhance its effect (Dhakad et al., 2018). Additionally, the greater persistence of this oil contributes to a prolonged effect, resulting in higher mortality rates of subsequent stages of the pest (Bett et al., 2017).

Higher concentrations of citronella oil resulted in lower emergence of female *C. capitata*, differing from its application directly on pupae, where this oil did not show a significant effect. This indicates the significant variability in the action of these oils depending on the developmental stage at which application is made. It is important to note that increasing concentrations of essential oils evaluated did not affect the emergence of male *C. capitata* when these oils were applied from pre-treated larvae. Further studies evaluating the fertility of these insects are necessary, as, in addition to the direct effects of larval and pupal mortality, the generation of sterile males also constitutes an important control method for fruit flies (*Ceratitis capitata*) (Kouloussis et al., 2017; Suárez et al., 2019).

## 5. Conclusions

Andiroba oil, when applied during the pupal stage, achieves satisfactory mortality rates and can be used at lower concentrations ( $\leq 40$  mg).

The use of eucalyptus oil can be recommended for application during the larval stage of *Ceratitis capitata*.

The use of essential oils appears to be a promising and sustainable alternative for the control of *Ceratitis capitata*.

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