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Johnatan Jair de Paula Marchiori Federal University Rural of Rio de Janeiro, Seropédica, Rio de Janeiro, Brazil

Anderson Mathias Holtz Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Ana Beatriz Mamedes Piffer Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Ronilda Lana Aguiar Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Jéssica Mayara Coffler Botti Department of Entomology, Federal University of Viçosa, Viçosa, Minas Gerais, Brazil

Mayara Loss Franzin Department of Agriculture, Nestlé Brazil, Várzea de Baixo, São Paulo, Brazil

Vinicius de Souza Oliveira Federal University of Espírito Santo - Center for Agricultural Sciences and Engineering, Alegre, Espírito Santo, Brazil.

Patrícia Soares Furno Fontes Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Bruna de Oliveira Magnani Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Matheus de Paula Gomes Campus Itapina, Federal Institute of Espírito Santo, Colatina, Espírito Santo, Brazil

Corresponding Author: Johnatan Jair de Paula Marchiori

Federal University Rural of Rio de Janeiro, Seropédica, Rio de Janeiro, Brazil

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Aqueous extract of chili pepper in the management of the pink hibiscus mealybug (Hemiptera: Pseudococcidae)

Johnatan Jair de Paula Marchiori, Anderson Mathias Holtz, Ana Beatriz Mamedes Piffer, Ronilda Lana Aguiar, Jéssica Mayara Coffler Botti, Mayara Loss Franzin, Vinicius de Souza Oliveira, Patrícia Soares Furno Fontes, Bruna de Oliveira Magnani and Matheus de Paula Gomes

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Abstract

Maconellicoccus hirsutus (Hemiptera: Pseudococcidae) is a polyphagous pest, attacks several crops of economic importance, including cocoa and coffee. Due to its rapid dissemination, studies are needed to develop management programs to combat this pest, as few studies seek the effectiveness of alternative products such as plant extracts, which can be promising in its control. Therefore, this work aimed to evaluate the efficiency of the aqueous extract of the fruits of chili pepper Capsicum frutescens (Solanaceae) in the management of the pink hibiscus mealybug. The tests were carried out in acclimatized chambers regulated at a temperature of 25 ± 1 °C, relative humidity of $70 \pm 10\%$, and a 12hour photophase. The treatments consisted of five extract concentrations: 0.0, 2.5, 5.0, 7.5, and 10% (weight/volume). Coffee leaves were submerged in the aqueous solution of pepper extract and mounted in Petri dishes with agar solution to transfer the mealybugs. Each treatment consisted of 10 repetitions with 10 young mealybugs each. The experiment was evaluated 24, 48, and 72 h after the procedure. Data were submitted for analysis of variance and regression test ($p \le 0.05$). Lethal concentration (LC50) was estimated using Probit analysis. The mortality of mealybugs increased with the increase in extract concentrations, with mortalities greater than 70% from the lowest concentration. The data fit the Probit model, with χ^2 of 1.0478 (p > 0.05) and a curve slope of 1.7799. The LC50 was estimated at 0.96%. Thus, the aqueous extract of *Capsicum frutescens* fruits is promising for the management of *M. hirsutus*.

Keywords: Maconellicoccus hirsutus, Capsicum frutescens, alternative pest control, integrated pest management

Introduction

The pink hibiscus mealybug, *Maconellicoccus hirsutus* (Green) (Hemiptera: Pseudococcidae), is a species originally from South Asia that quickly spread throughout the world ^[1, 2]. Worldwide, *M. hirsutus* has been found attacking more than 350 species, including fruit, vegetables, ornamentals, coffee, and cotton ^[3, 4, 5]. The species was first reported in Brazil in 2010 in the State of Roraima, and since then it has spread to other Brazilian states bringing economic losses to the affected regions. In 2013, the pest was detected in the cocoa crop in the regions of Bahia and Espírito Santo in the municipality of Linhares - ES ^[5].

The pink hibiscus mealybug is a polyphagous pest, it can cause severe damage and in high populations, it can lead the plant to death. This is because, when feeding, *M. hirsutus* injects toxic saliva into plants. The content injected by the pink hibiscus mealybug saliva can cause the malformation of leaves and fruits, promote the wilting of the apical branches and cause infested flowers and fruits to fall $^{[6, 7]}$.

Although established in Brazil, few control methods are available for the pink hibiscus mealybug. Currently, only the microbiological product *Cryptolaemus montrouzieri* Mulsant (Hemiptera: Coccinellidae) is registered in the information bank on agrochemicals and related products registered with the Ministry of Agriculture for the control of *M. hirsutus*^[8]. Due to the scarcity of products recommended for controlling this pest species, many producers end up applying synthetic chemicals not recommended for the crop to alleviate the damage caused by the organism. However, the indiscriminate use of these products can promote the emergence of

new resistant populations, cause mortality of natural enemies present in the environment, the intoxication of the surrounding fauna, and pollution of water bodies ^[9].

As an alternative to synthetic chemical control, research related to the use of extracts and substances obtained from plants has demonstrated satisfactory effectiveness in pest control ^[10, 9, 11]. Among the plants with acaricidal effects, peppers, belonging to the Solanaceae family, have a variety of alkaloid compounds and saturated or unsaturated fatty acids. These substances are produced in glands located in the placenta of the fruits, where the seeds are inserted and have an insecticidal effect ^[12, 13]. Thus, this work aimed to evaluate the

insecticidal effect of the aqueous extract of *Capsicum frutescens* fruits on *M. hirsutus*.

Materials and Methods

Local Site

The experiment was carried out at the Agricultural Entomology and Acarology Laboratory of the Federal Institute of Education, Science, and Technology of Espírito Santo - Campus Itapina (IFES-Campus Itapina), located in the municipality of Colatina, with coordinates of 19°29'52.7" S 40 °45'38.5" W (Fig. 1).

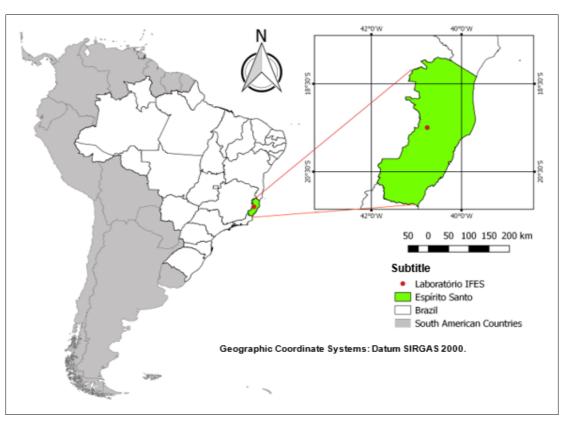


Fig 1: Map of the place where the experiments were carried out

Breeding of the pink hibiscus mealybug

Individuals of *M. hirsutus* were collected in cocoa plantations infested with this insect, in a rural property located in the district of Colatina, ES. Some specimens were sent for identification at the Center for Scientific and Technological Development in Phytosanitary Management (Nudemafi), at the Federal University of Espírito Santo (UFES), in Alegre, ES.

The breeding technique adopted was the same used by Sanches & Carvalho^[14]. In the initial infestation of pumpkins, individuals collected in the field on infested host plants were used. After establishing the initial colony, the mealybug multiplication process began. When it was necessary to replace the pumpkins with new ones, they were removed from the institute's experimental area, where chemical products are not used to grow the crop. They were placed in contact with the infested ones for approximately two hours. The proximity of the fruits favors the transfer of newly hatched pink hibiscus mealybug nymphs to the new fruit due to their mobility at this stage.

Obtaining Aqueous Extracts of Chili Pepper Fruits

For the preparation of the extracts, chili pepper fruits were

collected in their reproductive phase, in the Experimental Area of the IFES-Campus Itapina, in the Horticulture sector. After this procedure, the fruits were placed to dry in an oven with forced air circulation at a temperature of 40°C for 72 hours. After drying, they were ground using a knife mill to obtain a fine powder.

Bioassays

To obtain the aqueous extract of *C. frutescens*, the crushed plant material powder (10 g) was transferred to an Erlenmeyer flask (100 mL) containing distilled water and Tween[®] 80 adhesive spreader (0.05%), to obtain 100 mL of the 10% (w/v) starting solution. This solution was maintained under homogenization in a transverse shaker (240 rpm) for a period of 24 hours. After this period, the mixture was filtered with voile fabric and transferred to a volumetric flask, and the volume was checked to 100 mL.

The experimental units were composed of Petri dishes (10.0 x 1.2 cm) on coffee leaf disks of approximately 4 cm in diameter, as described by Holtz *et al.* ^[15]. The discs were immersed in the solution for 30 seconds, at different concentrations corresponding to the different treatments (0, 2.5, 5.0, 7.5, and 10.0%). The leaf disks were fixed to the

Petri dish, with a layer of 0.5 cm of agar-agar solution and solid Vaseline around the disk to prevent the insects from escaping, and the plates were sealed with transparent PVC film. Each treatment consisted of 10 repetitions with 10 mealybugs in the young stage each, totaling 100 insects per treatment. Subsequently, they were stored in a B.O.D (Biochemical Oxygen Demand), to preserve and maintain a constant ideal temperature, where they were removed and evaluated daily, for three days. Distilled water and adhesive spreader Tween® 80 (0.05) were used as a control. The insecticidal effect was evaluated 24, 48, and 72 hours after immersion.

The experiment was conducted in a completely randomized design. Data were corrected using the Abbott formula ^[16] and

subsequently submitted to analysis of variance and regression test ($p \le 0.05$), using the SISVAR software. To estimate the lethal concentration (LC50), Probit regression analysis was performed using the Polo Pc program.

Results and Discussion

According to the linear function of the regression test, there was an increase in the mortality of the pink hibiscus mealybug with the increase in the concentration of the aqueous extract of chili pepper (Fig. 2). The data fit the Probit model, showing a chi-square of 1.0478. The slope of the concentration-mortality curve was 1.7799%. The LC50 was estimated at 0.96%, that is, 0.96 g/100 mL would cause mortality in at least 50% of the population (Table 1).

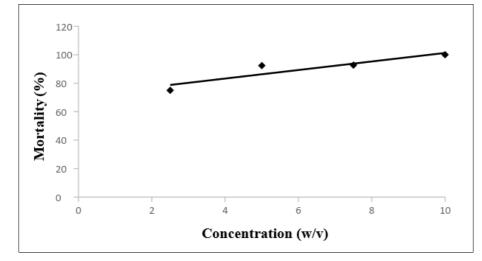


Fig. 2: Mortality (%) of Maconellicoccus hirsutus in different concentrations of Capsicum frutescens fruit aqueous extract.

Table 1: Concentration-response of chili pepper fruit extract on thepink hibiscus mealybug Maconellicoccus hirsutus. Temp.: $25 \pm 1^{\circ}$ C,RH 70 \pm 10% and 12 h of photophase.

		LC503 (IC4 a 95%) (g/100ml)			
303	$1,7799 \pm 0,4786$	0,96 (0,19 – 1,63)	2	1,0478	0,306
¹ Number of insects used in the test; ² Curve slope \pm standard error;					
³ Lethal concentration; ⁴ Confidence interval at 95% probability;					
⁵ degrees of freedom; ⁶ Chi-square test; and ⁷ P value.					

The values found demonstrated a high insecticidal potential of the aqueous extract of *C. frutescens* in the control of *M. hirsutus* because even at low concentrations of the extract, mortalities were higher than 70%.

This mortality rate can be attributed to the secondary compounds present in the *C. frutescens* extract. Several studies indicate the presence of alkaloids, tannins, esters, glycosins, saponins, flavonoids, phenols, and other components in fruits of the genus Capsicum. A significant presence of capsaicin is also observed, representing between 50 and 70% of the total capsaicinoids, the substance responsible for its pungency in pepper fruits ^[17, 18, 19].

Compounds such as alkaloids, phenols, and esters can act in multiple locations and express biocidal, developmental retardant of insects, repellent, and anti-food activity in pest organisms ^[20, 21]. In this way, GAUTAM *et al.* ^[22] isolated purified phenolic compounds from *Acacia nilotica* against the caterpillar *Spodoptera litura* Fabricius (Lepidoptera: Noctuidae) and observed inhibition of egg emergence, prolongation of the period of larval, pupal and total insect development, as well as observed mortality rates above 60% for adult individuals.

Other compounds such as tannins and saponins can reduce organisms' growth rate, affect feed efficiency and protein digestibility, and degrade their intestinal cells of them ^[23]. In a study carried out by Farahat *et al.* ^[24], a delay in the larval development of *Culex pipiens* L. mosquitoes (Diptera: Culicidae) subjected to treatment with tannic acids was observed, which suggests the effectiveness of these compounds in controlling insect pests.

In addition to the secondary compounds of chili pepper extract, other factors can act on the performance of insect pests, such as the extract extraction methodology, the extracted plant part, and the vegetative stage of the plant. In a study conducted by Izah ^[19], the crude, ketone, and ethanoic extracts of *C. frutescens* var. *minima* were evaluated in larvae of *Anopheles gambiae* (Diptera: Culicidae). The results showed that the ethanolic extract resulted in higher rates of larval mortality, suggesting that this extraction method may allow the extraction of greater amounts of secondary compounds from pepper fruits. This information is important for developing more effective and sustainable pest control strategies.

Also, different parts of the plant may contain varying amounts of specific compounds. In a study carried out by Faleiro *et al.*^[25], the mortality of *Atta laevigata* ants (Hymenoptera: Formicidae) was evaluated using ethanol extracts from different plant parts of *Eugenia dysenterica* (Mart.) (Myrtaceae). Results indicated significant mortality rates for all extracts of different plant parts, however, the highest mortality rate was observed for the extract of flowers and leaves. It is possible that the higher concentration of metabolites in these parts of the plant contributed to the mortality of the organisms. This information is important for the development of more effective pest control strategies using plant extracts.

In the present study, as fruits and seeds were used, probably there were higher concentrations of secondary compounds since they are part of the plant responsible for the reproduction and permanence of the species, therefore it needs greater protection of the plant's defenses. The degree of maturation of the fruits is another factor that can influence together with the part of the plant used from the pepper plants in the composition of the extracts and cause the mortality of mealybugs ^[23]. For example, the physiological changes in metabolites that occur during plant development affect their nutritional and health-promoting properties during vegetative development ^[26].

Finally, as the coffee leaf discs were submerged in the pepper extract (indirect application of the extracts), mealybugs are likely intoxicated through the digestive tract. Piffer *et al.* ^[27] studied the effect of the interaction between castor oil (*Ricinus communis*) and physic nut oil (*Jatropha curcas*) on the aphid *M. persicae* in direct (spraying) and indirect (immersion) application forms, insect mortality rates of up to 84% were observed in the study when they were exposed to the indirect treatment method.

Despite the promising results, it is necessary to identify more precisely the substances present in the fruit with insecticidal effects, to evaluate their sublethal effects, in addition to carrying out tests in fields with different concentrations to evaluate the viability of the method.

Conclusion

The aqueous extract of the fruit of C. *frutescens* has insecticidal potential to control M. *hirsutus* at all concentrations tested in the laboratory.

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