



## **Castor Bean Cake: A Promising Alternative for Sustainable Spider Mite Management**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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### **ABSTRACT**

*Tetranychus urticae* is an important phytophagous and cosmopolitan pest, affecting several cultures worldwide, such as papaya, strawberry, cotton, beans and soybeans. Chemical pesticides for mite control pollute the environment, poison humans and select resistant mite populations. *Ricinus communis* cake is a by-product of biodiesel production, containing compounds, such as ricinin and some fatty acids, described as toxic to arthropods. The objective of this work was to evaluate the castor bean cake acaricide potential on the spider mite, *T. urticae*. *Ricinus communis* dried seeds were pressed for oil extraction. Subsequently, the cake obtained after the oil extraction was ground in a knife mill to produce a fine powder. After, 0.5, 1.0, 1.5, 2.0, 2.5 and 3.0 grams of castor bean cake powder were mixed into 100 mL of Tween 80 aqueous solution 0.05 % v v<sup>-1</sup>, stirred (30 minutes, at 25 °C) and left to rest (20 minutes) to decant the solid particles. Finally, the supernatant (castor cake extract) was separated from the solid part by simple filtration. For direct application, 6 mL of castor cake extract were sprayed onto 10 *T. urticae* females dispersed on a 4 cm diameter *Canavalia ensiformis* leaf. For indirect application, 10 *T. urticae* females were placed in discs leaf, previously immersed in castor bean extract. Ten replicates were used for each treatment, evaluating

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the mortality parameter as a time function (24, 48 and 72 hours) for both application forms. The obtained data were submitted to linear regression analysis. *T. urticae* mortality did not show a dose-dependent behavior as a function of the castor bean cake extract concentrations for both forms of application (direct and indirect). Castor bean cake extract at 2.5 % w v<sup>-1</sup> caused the highest mortality *T. urticae* females treated directly or indirectly (95 or 88 %, respectively) than this extract in the other evaluated concentrations. The average mortality of *T. urticae* females treated directly or indirectly with this extract at the different concentrations evaluated were 88.4 and 77.7 %, respectively. The results showed that the castor bean cake extract, applied directly or indirectly in *T. urticae*, caused high mortality of females of this mite, being a promising alternative for the spider mite sustainable management.

**Keywords:** *Tetranychus urticae*; alternative control; castor bean cake.

## 1. INTRODUCTION

The spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae), is an invasive species with a polyphagous habit reported in more than 1.100 hosts, being considered one of the main agricultural pests in the world [1]. This mite is found in regions with warm and humid climate, occurring mainly during the hottest periods of the year [2]. Its occurrence is reported throughout the Brazilian territory, hosting crops of economic importance, such as, cotton, beans, papaya, corn, strawberry, soybean, tomato, grape and ornamental plant species [3].

*Tetranychus urticae* infestations are established between the ribs of the abaxial side of the leaves and are characterized by the formation of a large amount of web to protect their eggs and colonies. This mite has a phytophagous habit, causing yellowing, necrosis and perforation of the leaf blade, reducing the plant's photosynthetic capacity [4].

The papaya crop is among the most affected culture by the spider mite attack. In this culture, the high populations of spider mite cause the premature leaf fall, decreasing the plant's productivity. In addition, the leaf fall exposes the fruits to the direct action of the sun's rays, causing harming product quality [4]. Brazil is the fourth largest cotton producer in the world, being the second main exporter of this fiber, with around 2.3 thousand tons harvest in the 2020/2021 [5]. In addition, Brazil is an important producer and exporter of papaya, being Espírito Santo the most productive State, with 403.278 tons in 2019 [6]. Given the importance of these cultures, *T. urticae* becomes a threat to the Brazilian and Espírito Santo economy, it is necessary to search for alternatives to control this pest.

Synthetic compounds, such as Abamectin and Bifenthrin, are used to control agricultural mites, however, causing spider mite resistant populations selection due to its high reproductive potential [7–9]. Furthermore, synthetic acaricides have high residual levels, being toxic to the environment and humans [10–12].

Plant derived formulations have been studied for pest management, being advantageous for their low toxicity to the environment and human. *Moringa oleifera* L seed extract at different stages of maturation showed acaricide activity on *T. urticae* [13]. Furthermore, *Ricinus communis* L. (Euphorbiaceae) seed oil showed efficiency in the red coffee mite [*Oligonychus ilicis* (McGregor)] control, indicating the potential of this plant for mite alternative control.

Castor bean, *R. communis*, is a plant species with little known insecticide and acaricide potential, being used mainly as a source of oils extracted from its seeds. Castor bean seeds contain toxic proteins, such as protein inhibitors that act on  $\alpha$ -amylase and other glycosyl hydrolases [14–16]. The castor bean cake is a by-product of *R. communis* seeds oil extraction described for having insecticide action against *Aegypti larva* [12].

The objective of this work was to evaluate the acaricide potential of castor bean cake against *T. urticae*.

## 2. MATERIAL AND METHODS

The experiments were carried out at the Federal Institute of Education, Science and Technology of Espírito Santo (IFES) – Campus Itapina (IFES-Campus Itapina). All experimental units were conditioned in acclimatized chambers at the temperature 25 ± 1°C, relative humidity 70 ± 10% and photophase for 12 hours.

*Tetranychus urticae* breeding was established in *Canavalia ensiformis* leaves without phytosanitary treatment, cultivated in pots placed in cages made with anti-aphid mesh, kept in a greenhouse.

## 2.1 Castor Cake Extracts Production

Dried fruit seeds were collected at the IFES-Campus Itapina and subjected to oil extraction. Subsequently, the castor bean cake from the oil extraction was ground in a knife mill to obtain a fine powder.

For aqueous extraction, 0.5, 1.0, 1.5, 2.0, 2.5 and 3.0 grams of castor bean cake powder was mixed into 100 ml of Tween 80 aqueous solution 0.05% v v<sup>-1</sup>, stirred (thirty minutes at 25°C), stirred (30 minutes, at 25°C) and left to rest (20 minutes) to decant the solid particles. The supernatant was separated from the solid part by simple filtration using a polyester filter.

## 2.2 Bioassay

*Canavalia ensiformis* leaves were washed with distilled water, dried on filter paper and placed in gerbox-type plastic boxes. Afterwards, 10 females of *T. urticae* were transferred to 4 cm diameter *C. ensiformis* leaf discs.

Two types of tests were carried out: direct and indirect application of the castor bean cake extract on *T. urticae*.

### 2.2.1 Direct application of castor bean cake extract on *T. urticae*

The direct application of castor bean extract on *T. urticae* was carried out at 25 ± 20 °C, 70 ± 10 % U.R. and 12-hour photophase. Castor bean cake extract at concentrations of 0.5, 1.0, 1.5, 2.0, 2.5 and 3.0, respectively, was applied to *C. ensiformis* leaf discs (4 cm in diameter) placed in Petri dishes (10.0 x 1.2cm) containing 10 *T. urticae* females. Moistened cotton was added around the *C. ensiformis* leaf discs to prevent mites from escaping and to keep from turgidity of the leaf. The experiment was installed in a completely randomized design with 7 treatments (concentrations) and 10 replications. The experiment was carried out in a 7 x 10 factorial (7 concentrations and 10 replicates), with each Petri dish being a replicate. The mortality of individuals was evaluated 24, 48 and 72 hours after spraying. The data obtained were submitted to linear regression analysis.

The control consisted of discs sprayed with the Tween<sup>®</sup> 80 adhesive spreader solution in distilled water at a concentration of 0.05 % (v v<sup>-1</sup>). 6 ml of each solution were applied to the plates containing the mites using a Potter tower, under a pressure of 15 psi.

### 2.2.2 Indirect application of castor bean cake extract on *T. urticae*

The indirect application of castor bean extract on *T. urticae* was carried out at 25 ± 2 °C, 70 ± 10 % U.R. and 12-hour photophase.

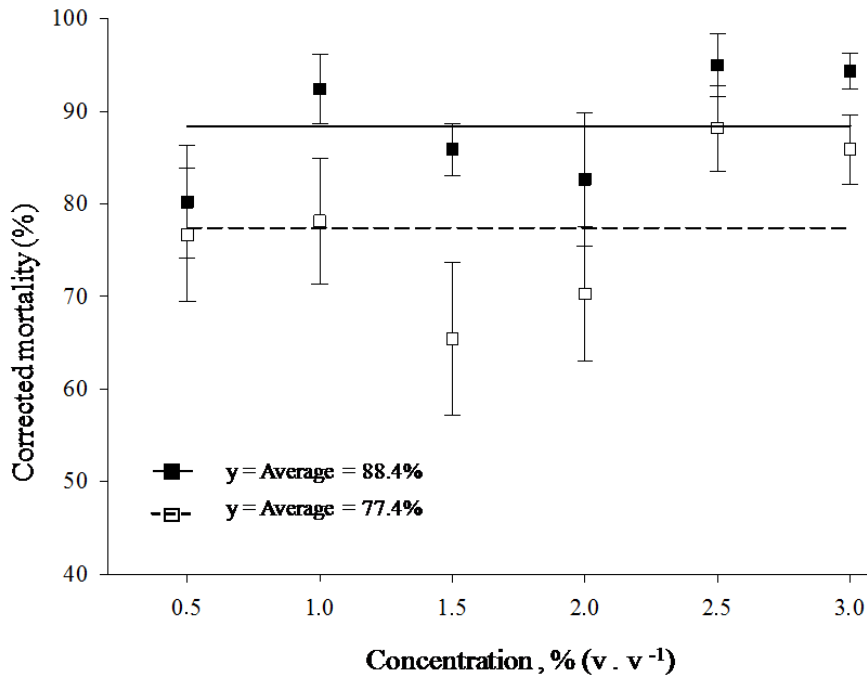
For the indirect application of castor bean cake extract in *T. urticae* females, *Canavalia ensiformis* leaf discs were immersed for 5 seconds in castor bean cake extracts at concentrations of 0.5; 1.0; 1.5; 2.0; 2.5; 3.0, respectively. Control treatment consisted of discs previously immersed in a Tween<sup>®</sup> 80 adhesive spreader solution in distilled water at a concentration of 0.05 % (v v<sup>-1</sup>). After immersion, the discs were placed on paper towels to remove excess liquid and dry. Afterwards, the discs were placed in Petri dishes with moistened cotton on the bottom and sides of the disc to maintain the turgidity of the leaf and prevent the individuals from escaping. The experiment was carried out in a 7 x 10 factorial (7 concentrations and 10 replicates), with each Petri dish being a replicate. The mortality of the individuals was evaluated 24, 48 and 72 hours after spraying the extract. The data obtained were submitted to linear regression analysis.

## 3. RESULTS AND DISCUSSION

There was no significant interaction between the application form and the castor bean cake extract concentration to cause *T. urticae* females' death ( $F_{5, 108} = 0.573$ ;  $P = .05$ ).

*T. urticae* mortality treated directly or indirectly with castor bean extract was not directly proportional to the concentration of this extract. Castor bean cake extract at 2.5 % w v<sup>-1</sup> caused the highest mortality of *T. urticae* treated directly and indirectly (95 and 88 %, respectively) than other evaluated concentrations of this extract (Fig. 1).

The average mortality of *T. urticae* treated directly or indirectly with castor bean cake extract under the different concentrations evaluated were 88.4 and 77.7 %, respectively.



**Fig. 1. Mortality corrected in relation to the control (Abbott, 1925) of *Tetranychus urticae* females treated with castor bean cake at different concentrations, by means of direct (■) and indirect (□) application, after three days. Temperature of  $25 \pm 1$  °C, RH  $70 \pm 10$  % and photophase of 12 hours**

Castor bean cake extract being toxic against *T. urticae* agrees with the report of castor bean oil causing acaricidal effect on *Oligonychus ilicis* (McGregor, 1917) (Acari: Tetranychidae), with LC50 of 1.07 % [17]. Castor bean seed oil applied indirectly on *Diaphania nitidalis* (Stoll) (Lepidoptera: Pyralidae) causing 92 % mortality, demonstrated the ability to intoxicate arthropods by the digestive route [11], corroborating with the observed for castor cake extract indirect application on *T. urticae* in this study. Furthermore, it was demonstrated that RcTI, a protease competitive inhibitor with 14 kDa present in castor bean cake, caused 91 % inhibition of midgut proteases from *Aedes aegypti* larvae, agreeing with the observed for *T. urticae* treated with castor bean cake in this work [12].

Detoxification enzymes in the mites' digestive tracts justifies the lower *T. urticae* mortality under indirect application than under direct application of castor bean extract [18]. Castor bean cake extract causing higher *T. urticae* mortality under direct application than with that indirect is justified by toxins acting as fumigants and entering the insect's body through its tracheal system, leading this organism to death [19]. The

castor bean cake generated after oil extraction has high ricinin (alkaloid toxic against arthropods) content, justifying the observed for the *T. urticae* treatment with this cake extract [20]. The high mortality caused by castor bean cake extract on *T. urticae* agrees with reports of this cake presenting high content of ricin, a toxic protein against insects, humans and animals. Ricin catalyzes the 28S rRNA alteration in the ribosome, inhibiting the protein synthesis, leading to cell death [21]. The results for *T. urticae* castor bean cake extract treatment corroborate those of *Anopheles stephensi* (Liston), *Culex quinquefasciatus* (Say) and *Aedes albopictus* (Skuse) (Dip.: Culicidae) treated with acetonic extract of powdered seeds of this plant leading to 100 % mortality of their larvae [22].

Castor bean cake extract applied directly or indirectly to *T. urticae* proved to be effective in causing mortality of this mite, being a promising way for the sustainable management of the spider mite.

#### 4. CONCLUSION

The castor bean cake extract applied directly and indirectly on *T. urticae* caused average mortality

of up to 95 and 88 %, respectively, being a promising way for the spider mite sustainable management.

## DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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