Artículo científico

Acaricidal activity of the oil from *Jatropha curcas* L. seeds on larvae of *Rhipicephalus (Boophilus) microplus* (Canestrini, 1887) (Acari: Ixodidae)▲

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Abstract

In order to evaluate the acaricidal activity of the oil from *Jatropha curcas* L. seeds, with different storage periods, against larvae of the tick *Rhipicephalus (Boophilus) microplus* (Strain: Cayo Coco), this research was conducted at the Pastures and Forages Research Station Indio Hatuey. Oils with different storage periods (1 and 4 years) were evaluated. The oil was obtained from the mature, sun-dried and shelled fruits, of the Cape Verde provenance, collected in the Paraguay Farm, Guantánamo. A complete randomized experimental design was used with 17 treatments and 10 repetitions per treatment, from the different evaluated concentrations for the oils (0,5; 1,75; 2,5; 5 and 10 mg/mL), the solvent (Tween-80) and the negative (distilled water) and positive controls (Butox® Deltamethrin) used. For the *in vitro* studies the larval immersion techniques were applied. The negative control (distilled water) did not show acaricidal activity on the larvae (2,07 ± 0,40 %); a similar performance was observed in the solvent concentrations. The oil showed a marked larvicidal action in the two storage periods, with significant differences ($p<0,05$) among the experimental treatments, the negative control and the solvent, with values higher than 93 %. The best results were for the concentrations of 10 mg/mL oil 2014 (97,72 ± 0,58 %) and of 10 and 5 mg/mL oil 2017 (98,99 ± 0,33 and 98,11 ± 0,30 %, respectively), without significant differences with regards to Butox® (97,83 ± 0,45 %). The results showed that the oil from the *J. curcas* L. seed has acaricidal activity for the evaluated concentrations against larvae of the tick *R. (B.) microplus*, without significant differences between the storage periods and with values that exceeded 90 % of efficacy.

Keywords: fruit, mortality, parasites, storage

Introduction

The tick [*Rhicephalus (Boophilus) microplus*] is considered the most important ectoparasite for cattle husbandry, affecting up to 80 % of the population of this species in the world due to its capacity to adapt to the most varied ecological conditions (Aguilar-Tipacamú *et al*., 2016). So far chemical acaricides constitute the main control method for these parasitoses. Nevertheless, this strategy has become inefficacious in some regions due to the emergence and accelerated development of resistance to the active principle, in tick populations (Higa *et al*., 2016).

In this context, plants have proven to be an alternative method for the control of insects and mites due to the secondary metabolites produced as a defense mechanism (Adenubi *et al*., 2016; Arceo-Medina *et al*., 2016). The essential or non-essential oils present in plant species are being widely used because of their therapeutic, bactericidal, fungicidal and insecticidal activity (Barros *et al*., 2019). Among them are those from *Jatropha curcas* L. seeds, whose utilization in traditional medicine and veterinary medicine has been reported in Asia, Africa and Latin America. Considering the above-mentioned facts, the objective of this research was to evaluate the acaricidal activity of the *J. curcas* fruit oil, with different storage periods, against the tick *R. (B.) microplus*.

Materials and Methods

Location. The studies were conducted at the Pastures and Forages Research Station Indio Hatuey (EEPFIH), located in the Perico municipality, Matanzas province, Cuba.

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Treatments and experimental design. A complete randomized experimental design was used with 17 treatments and 10 repetitions per treatment, from the different evaluated concentrations for the oils (0,5; 1,75; 2,5; 5 and 10 mg/mL), the solvent (Tween-80) and the negative and positive controls used. The evaluated treatments were:

T1. Butox® (Deltamethrin) (Positive control)
T2. Distilled water (Negative control)
T3. Tween-80 0,5 mg/mL (Negative control)
T4. Tween-80 1,75 mg/mL (Negative control)
T5. Tween-80 2,5 mg/mL (Negative control)
T6. Tween-80 5 mg/mL (Negative control)
T7. Tween-80 10 mg/mL (Negative control)
T8. Jatropha Oil 2017 0,5 mg/mL + Tween-80
T9. Jatropha Oil 2017 1,75 mg/mL + Tween-80
T10. Jatropha Oil 2017 2,5 mg/mL + Tween-80
T11. Jatropha Oil 2017 5 mg/mL + Tween-80
T12. Jatropha Oil 2017 10 mg/mL + Tween-80
T13. Jatropha Oil 2014 0,5 mg/mL + Tween-80
T14. Jatropha Oil 2014 1,75 mg/mL + Tween-80
T15. Jatropha Oil2014 2,5 mg/mL + Tween-80
T16. Jatropha Oil 2014 5 mg/mL + Tween-80
T17. Jatropha Oil 2014 10 mg/mL + Tween-80

Biological material. Larvae from the Cayo Coco strain of *Rhipicephalus (Boophilus) microplus* were used, obtained by National Laboratory of Veterinary Parasitology, located in the San Antonio de los Baños municipality, Artemisa province, Cuba. The selection of the biological material was based out based on the characteristics reported by Farias *et al.* (2007).

Oil extraction. The oil was obtained from the mature, sun-dried and shelled fruits of the Cabo Verde provenance, collected in the Paraguay Farm, Guantánamo. Afterwards, the seeds were pressed, according to the methodology described by Sotolongo *et al.* (2007). Oils with different storage periods: 1 year (extraction 2017) and 4 years (extraction 2014), were evaluated.

For determining the profile of chemical compounds the gas chromatography–mass spectrometry (GC-MS) was used.

Experimental procedure. The methodology described by Leite (1998), modified by Chagas *et al.* (2012), was applied, with the technique of immersion in the evaluated substances. The essay lasted 72 hours and 21 day-old larvae were used. To calculate mortality the formula: [Mortality (%) = dead larvae /total larvae x 100] was used, and the values were transformed to efficacy in mortality.

Statistical analyses. The data were recorded in Microsoft Excel® spreadsheets to perform the respective statistical analyses. The efficacy of larval mortality was transformed according to the square root arcsin of the value to achieve a distribution close to normal. Variance analysis was used, after testing the variance homogeneity (Levene's test) and normal distribution (Shapiro-Wilk) assumptions. For the mean comparison Duncan's test at applied for a significance level of $p<0,05$. The statistical package IBM SSPS® version 22 for Windows were used.

Results and Discussion

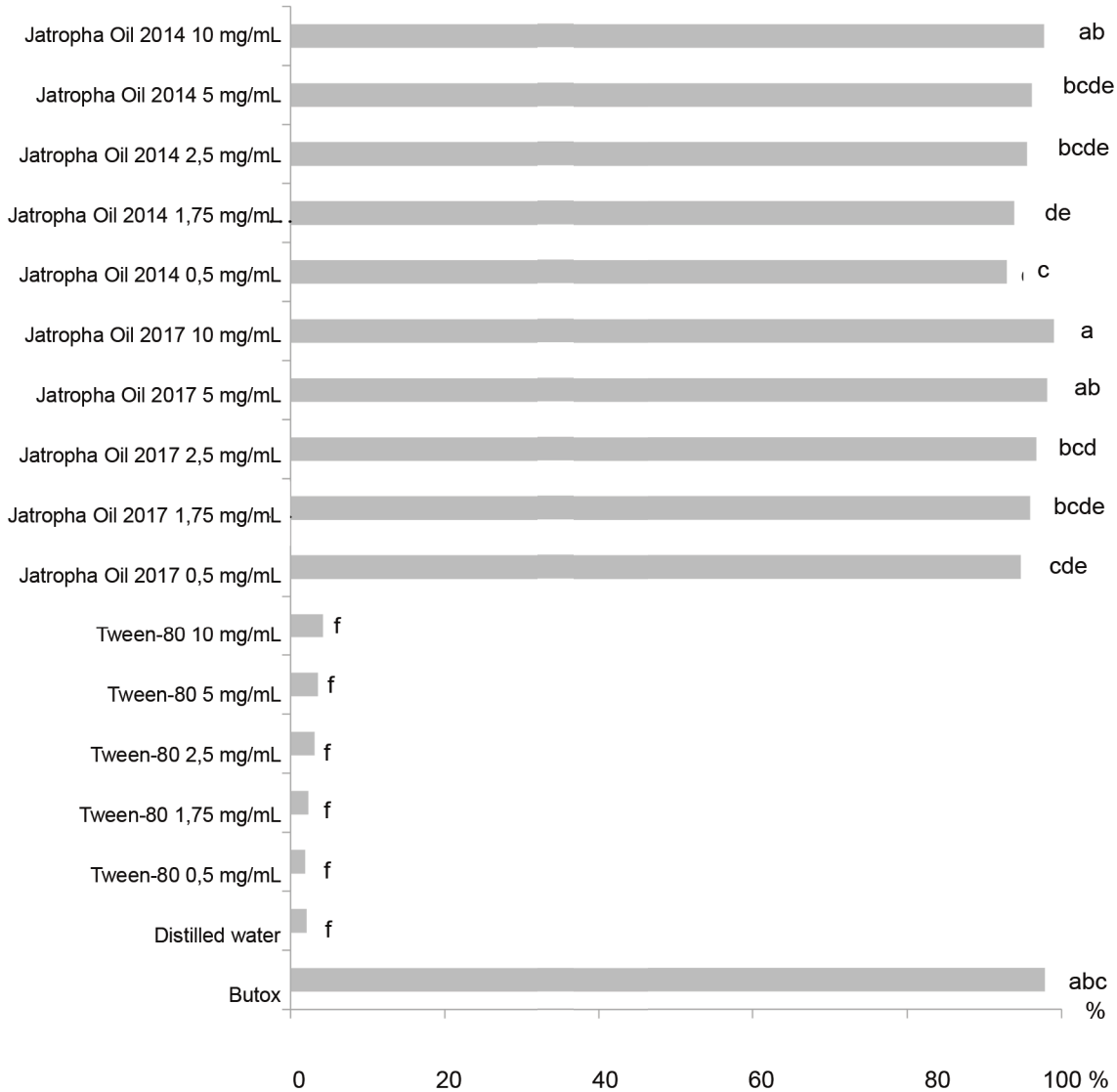
The results of the efficacy of the evaluated substances on *R. (B.) microplus* larvae are shown in fig. 1. The oil from the *J. curcas* fruit showed a marked larvicidal action in the two storage periods, with significant differences ($p<0,05$) among the experimental treatments, the negative control and the solvent.

All the Jatropha oil concentrations showed efficacy values higher than 93 %. However, the best results were obtained for the concentrations of 10 mg/mL in the oil extracted in 2014 (97,72 ± 0,58 %) and 10 and 5 mg/mL in the oil from 2017 (98,99 ± 0,33 and 98,11 ± 0,30 %, respectively), without significant differences with regards to Butox® (97,83 ± 0,45 %).

As expected, the negative control (distilled water) did not show acaricidal activity on the larvae (2,07 ± 0,40 %). A similar performance was observed in the solvent with values of 1,89 ± 0,27; 2,28 ± 0,33; 3,10 ± 0,60; 3,55 ± 0,41 and 4,19 ± 0,57 % for the concentrations of 0,5; 1,75; 2,5; 5 and 10 mg/mL of Tween-80, respectively. This proved that this solvent can be used in the biological tests with *R. (B.) microplus,* because according to Chagas *et al.* (2003) it has low molecular weight and little viscosity, and does not interfere with the efficacy of the evaluated substances.

Martínez-Velázquez *et al.* (2011), when evaluating the essential oils from *Cuminum cyminum* seeds and from *Pimienta dioica* (L.) Merr. fruit, reported acaricidal activity and high toxic effects which caused 100 % of mortality in 10 day-old *R. (B.) microplus* larvae.

On the other hand, Chagas *et al.* (2012), in studies conducted with larvae of this same



Different letters indicate significant differences (p<=0,01**)

Figure 1. Efficacy of the oil from *J. curcas* seed against *R. (B) microplus* larvae with different storage periods.

tick species, reported acaricidal effects of the essential oils and the emulsifiable concentrate of *Eucalyptus* ssp. The authors informed that the essential oil of *Eucalyptus citriodora* Hook and *Eucalyptus staigeriana* F. Muell. ex Bailey showed maximum efficacy (100 %) on larval mortality, in concentrations of 10 and 20 % for each of the species, respectively.

Several studies have been conducted to evaluate the acaricidal activity of essential or non-essential oils, from different plant parts, in order to determine their potentialities as a valuable natural resource in tick control (Campos *et al.,* 2012). Nevertheless, the reports about the acaricidal properties of *J. curcas* are scarce (Juliet *et al.,* 2012).

The results of this study are similar to the ones reported by Fuentes-Zaldívar *et al.* (2017), when evaluating the acaricidal activity of the *J. curcas* oil on ingurgitated *R. (B.) microplus* engorged females and larvae, but in higher concentrations (5, 10, 15 and 25 %), with mortality values which oscillated between 95 and 98 % for the larvae.

According to the authors, larvae are much more sensitive to the effect of Jatropha oil than

the ingurgitated females. This could be given by their size, because they need lower quantity of the product to appreciate their effect; besides, their structure is more sensitive and the layers that form their tegument are thinner, which allows the absorption of the product and its distribution in the organism.

According to Lopera-Vélez *et al.* (2017), *J. curcas* is a promising plant species, with a richness of bioactive compounds that allows it to control efficiently bacteria, fungi, parasites and other organisms which affect animal growth and production. Yet, there are still few studies conducted with this plant.

The profile of chemical compounds showed, besides the presence of fatty acids (oleic, linoleic, lignoceric, elaidic, palmitic, palmitelaidic and stearic), substances such as camphor (2,4-Decadienal) and phytosteroids (Stigmasterol and β-sitosterol), of which effects as nematicidal, larvicidal, insecticidal and repellent, are acknowledged (Caboni *et al.*, 2012). These substances have an important biological activity in animal and human medicine, given their pharmaceutical value.

Although there is still much to study about the biological activity of this species in tick control, especially defining which of its substances have the highest biological activity, the results reached until now indicate that in its chemical composition there is a diversity of principles and metabolites, which could act individually, additively or in synergy between them, favoring the control of these parasites.

Conclusions

The oil from *Jatropha curcas* seeds has acaricidal activity against *Rhipicephalus (B.) microplus* larvae, without significant differences between the storage periods and with values that exceeded 90 % of efficacy. The highest acaricidal efficacy was found for the oil extracted in 2017 with higher values than 98 %, although no significant differences were found between the storage periods and the concentrations of 5 and 10 mg/mL were the ones with higher acaricidal activity.

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