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In vitro efficacy of oil from the seed of Carapa guianensis (andiroba) in the control of Felicola subrostratus

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Abstract: This study was perfored to assess the *in vitro* effect of oil from the seed of andiroba (Carapa guianensis Aubl.) on Felicola subrostratus (Burmeister, 1838) (Mallophaga: Trichodectidae). Six hundred specimens of F. subrostratus from neighborhood of Jordão, Recife-PE, Brazil, were collected by hand directly from the fur of cats infested naturally. The lice were transported in plastic recipients to the Laboratory of Parasitic Diseases of Domesticated Animals of the Department of Veterinary Medicine of the Universidade Federal Rural de Pernambuco (Brazil) for the immersion test. Four dilutions of andiroba oil (100, 50, 25 and 10%) in distilled water were tested, using Tween 80 as dispersant; two control groups (one with distilled water and the other with monosulfiram) were also formed; four replicates were performed, with 25 specimens for each dilution, totaling 100 lice per treatment. After the test, the insects were kept at room temperature and observed for mortality rates for 72 h. The biological activity of the product achieved 100% mortality of the insects in the first hour at concentrations of 100 and 50% and in the third hour at concentrations of 25 and 10%. The results demonstrate the possibility of controlling F. subrostratus throught the use of oil extracted from andiroba seeds.

Introduction

Lice infestation is denominated pediculosis. In cats, the etiological agent is *Felicola subrostratus*, which belongs to the order Mallophaga, with its characteristic pointed head This species of lice is cosmopolitan and feeds on epidermal debris and fur. Infested cats exhibit a variable degree of pruritus. This species of lice is cosmopolitan and feeds on epidermal debris and fur. Infested cats exhibit a variable degree of pruritus (Noli, 2002). Parasitism by this species of lice has been described in Brazil and other parts of the world (Pollmeier et al., 2004; Jittapalapong et al., 2008; Ferreira et al., 2009).

Transmission occurs by direct contact or contaminated brushes and combs. The fur of infected cats is often unclean, matted and has an ill appearance, as the disease is associated to the overpopulation of animals and sanitary conditions of the environment. Lice copulation occurs on the animal and females deposit their eggs, called nits, on the fur of the host. A nymph hatches from each nit and transforms into an adult. The complete cycle lasts two to three weeks. Far from the host, lice survive only a few days (Noli, 2002).

Lice infestation is generally treated with synthetic pyrethroid insecticides, carbamates, fipronil

or selamectin. These drugs come in powder, liquid, shampoo, spray or spot-on form (Noli, 2002). However, plants are potential alternative to currently employed insecticides, as they are a rich source of bioactive chemical products and have both aromatic characteristics and essential oils, which are considered responsible for insecticidal activity (Carvalho, 2004). Plants constitute a viable, less aggressive treatment for cats, with less harm to ecosystems (Marinho et al., 2007).

Andiroba (or crabwood) (*Carapa guianensis* Aubl.) is a species of tree belonging to the family Meliaceae that is distributed throughout the northern portion of South America, including the Amazon Basin, as well as Central America, the Antilles and tropical Africa. The oil derived from andiroba seeds is used in folk medicine in northern Brazil for fever, rheumatism and as an anti-inflammatory agent, antibacterial agent and insect repellant (Neves et al., 2004).

Considering the high occurrence of parasitic dermatitis affecting domestic cats and the possibility of obtaining alternative products for combating this condition, the aim of the present study was to assess the *in vitro* efficacy of andiroba oil (*Carapa guianensis* Aubl.) on *Felicola subrostratus*.

Material and Methods

The experiment was carried out at the Laboratory of Parasitic Diseases of Domestic Animals, Preventive Medicine Sector, Veterinary Medicine Department of the Universidade Federal Rural de Pernambuco (Brazil). Six hundred specimens of *Felicola subrostratus* were manually collected directly from the fur of naturally infected male and female domesticated cats of different ages from the neighborhood of Jordão in the city of Recife (northeastern Brazil).

The lice were transported to the laboratory in plastic recipients and divided into six treatment groups. Four repetitions were performed with 25 specimens, totaling 100 lice per treatment. An andiroba seed oil formula was obtained from Beraca Sabará SP in plastic package with 5 L, batch 05083140ST, rain forest RE3140. Four concentrations of the oil were tested (100, 50, 25 and 10%), using Tween 80 as the dispersant. Two other groups served as the controls, one using distilled water and the other using a conventional ectoparasiticide containing monossulfiram (Sarnasol® Schering Plough) at the concentration recommended by the manufacturer. The insects were submitted to an immersion bath in 50 mL disposable cups containing 3 mL of the respective solutions. The solution was maintained under constant agitation for three minutes, after which the excess liquid was removed and the specimens were transferred to paper-filter envelops and kept at room temperature. Six observations were made: 1, 3, 6, 24, 48 and 72 h after beginning the test. The assessment of efficacy was performed by counts of living and dead specimens, with mortality based on the physical aspect of the insect.

For the statistical analysis, absolute and relative distributions were determined. Pearson's chi-square test was used for comparisons between treatments, with the margin of error set at 5.0%. The Statistical Package for

the Social Sciences (SPSS, v. 15) was employed for all analyses (Zar, 1999).

Results and Discussion

Table 1 displays the percentage values of the in vitro efficacy of the oil derived from seeds of Carapa guianensis on F. subrostratus. One hundred percent mortality of the insects occurred within the first hour of the tests at concentrations of 100 and 50%. These results were similar to those achieved with monosulfiram. which is also known as tetraethyl thiuram monosulfide. This ectotoparasiticide is a sulfur compound used by a number of researchers and has demonstrated efficacy, especially for ectoparasites of mice (Santos et al., 1988; Alexandre & Bressnan, 1994). On cats, monosulfiram has demonstrated 100% efficacy against lynxacariosis in a study carried out in the city of Belém (northern Brazil) (Serra-Freire et al., 2002). At concentrations of 25 and 10%, 100% mortality of the lice occurred after three hours.

The specimens in the control group with distilled water remained viable throughout the experiment, with an increasing mortality rate beginning at 48 h (Table 1). This finding stems from the biology of the insect, which only survives two to three days without a host (Noli, 2002).

Although studies on andiroba oil as a phytotherapeutic agent are rare, the action of this oil has been demonstrated on the larvae of *Musca domestica* (Farias, 2007), *Aedes* sp. (Miot et al., 2004) and acaricidic activity has been demonstrated on ingurgitated females of *Boophilus microplus* (Farias et al., 2007), *Anocentor nitens* and *Rhipicephalus sanguineus* (Farias et al., 2009). In the experiment carried out by Farias (2007), larval mortality rates were 20, 7.5, 2.5 and 7.5% for concentrations of 100, 30, 25 and 10% and the inhibition of the emergence of adults was demonstrated. Miot et al. (2004) concluded that

Table 1. Absolute (n) and relative (%) frequency of specimens of *Felicola subrostrat*us killed following immersion in solutions, according to evaluation time.

Treatment	Time (h)						Tetal		
	Up	Up to 1		>1 to 3		> 3		Total	
	n	%	n	%	n	%	n	%	
100% andiroba oil	100	100.0	-	-	-	-	100	100.0	
50% andiroba oil	100	100.0	-	-	-	-	100	100.0	
25% andiroba oil	80	80.0	20	20.0	-	-	100	100.0	
10% andiroba oil	20	20.0	80	80.0	-	-	100	100.0	
2,5% monosulfiram	100	100.0	-	-	-	-	100	100.0	
distilled water	-	-	-	-	100	100.0	100	100.0	
Total group	400	66.7	100	16.7	100	16.7	600	100.0	

^{*}Significant difference at 5.0%; Pearson's chi-square test between all treatments (p<0.001*); Pearson's chi-square test between 25 and 50% andiroba treatments (p<0.001*); Pearson's chi-square test between 25 and 10% andiroba treatments (p<0.001*); Pearson's chi-square test between 10% andiroba treatment and monossulfiram (p<0.001*); Pearson's chi-square test between 25% andiroba treatment and monossulfiram (p<0.001*).

pure andiroba oil offers a discreet repellant effect for *Aedes* sp., but significantly less than that offered by 50% DEET. The *in vitro* tests carried out by Farias et al. (2007; 2009) achieved similar results to those of the present study, with 100% efficacy for all concentrations tested (10, 25, 30, 50 and 100%).

Studies assessing the pediculicide action of different vegetal extracts have reported lower efficacy rates than those of the present study. Neto et al. (2004) performed in vivo tests of extracts from Aspidosperma pvricollum and Anandenanthera *macrocarpa* for combating Damalinia caprae on goats raised in the semiarid region of the state of Paraíba (Brazil) and achieved mortality rates of 72.82-77.46% and 60.76%, respectively. Heath et al. (1995) tested extracts of neem (Azadirachta indica) and pyrethum (Chrysanthemum sp.) on Bovicola ovis on sheep in comparison to conventional insecticides and obtained a reduction of 85 to 100% of the lice over a period of at least 40 to 50 days. On the other hand, Ramos et al. (2009) carried out an in vitro study on the efficacy of a solution containing extracts of Saccharum officinarum, Azadirachta indica and Eucalyptus sp. for the control of Pediculus capitis and report a 60.28% mortality rate between 3 and 6 h following an immersion bath, with the remaining specimens dying within 24 h from the beginning of the treatment, thereby demonstrating faster action in comparison to the group treated with a pyrethroid.

Substances found in vegetal extracts, such as terpenoids and alkaloids, contribute toward their activity against insects (Isman, 2000). The oil from andiroba seeds is composed nearly exclusively of saponificable material, with a high concentration of unsaturated fatty acids, such as oleic acid and palmitic acid, as well as limonoids in the non-saponificable portion, among them: andirobina, epoxiazadiradiona, 6α-acetoxigedunina, 6α-acetoxiepoxiazadiradiona, 6β-cetoxigedunina, 11βacetoxigedunina, 6α,11β-diacetoxigedunina, diacetoxigrdunina, 6α-hidroxigedunina and 7-desacetoxi-7-oxogedunina, which are likely the greatest representatives of the terpene class and responsible for the insecticidal activity (Dantas et al., 2000). But despite the evidence of the bioactivity of andiroba against insects and mites, other studies must be implemented in order to assess its spectrum of action "in vivo" for treatment of ectoparasitic infestations that attack pets.

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