

**Acaricidal Activity of Aqueous Extracts of Leaves and Stems
of *PolygonumHydropiperoides*Michaux**

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¹Financial support: EMBRAPA, FAPEMIG, CNPq, ²Laboratory of Phytochemistry, Institute of Biological Sciences, Federal University of Juiz de Fora, Street José Lourenço Kelmer s/n, Campus, São Pedro, Juiz de Fora/MG, Brazil, ³Embrapa Dairy Cattle, Street Eugênio do Nascimento n° 610, Dom Bosco, Juiz de Fora/MG, Brazil

Phytotherapy has been rescued by ethnoveterinary research, in order to control various diseases that affect livestock. The use of many of the current acaricides affects the environment and leaves residues in milk. Therefore an alternative to commercial synthetic products is necessary. In a previous ethnoveterinary survey was verified that the species *Polygonumhydropiperoides*Michaux, an herbaceous plant that belongs to Polygonaceae family was used by farmers against the cattle tick, *Rhipicephalus (Boophilus) microplus*. This herb, therefore, was selected for this study. Aqueous extracts were obtained by infusion of *P.hydropiperoides* leaves and stalks separately. After lyophilization they were resuspended in distilled water at a concentration of 200 mg/mL. The acaricidal test was made by the immersion of engorged females of *R.(B.) microplus* for five minutes and then evaluated the biological parameters. The parameters were analyzed by ANOVA test and Tukey *post-hoc* considering $p < 0.05$. According to the classification of the efficacy index proposed by the World Association for the Advancement of Veterinary Parasitology, the aqueous extract of *P.hydropiperoides* stalk was classified as "low effectiveness", showing 72,11% of efficacy, while the aqueous extract of leaves was "ineffective", showing 47,96% of efficacy. The extract of stalk showed a significant reduction of the egg mass ($p < 0.01$). These results suggest that both extracts have active compounds against the tick and show the potential acaricide activity of the aqueous extract of stalk.

Key Words: Ethnoveterinary, Medicinal plants, *Rhipicephalus microplus*

INTRODUCTION

Brazil is the country that holds the greatest amount of biodiversity, around 15 to 20% of the whole world, specially the superior plants. Even with this big vegetal diversity, around 60,000 superior vegetal species listed (most of which in the Amazon, then in the Mata Atlantica, followed by the Cerrado), only 8% were studied for the research of bioactive compounds and 1,100 species were evaluated into their medicinal properties (Health Ministry, 2006). The traditional use of medicinal plants has been rescued by ethnoveterinary research, seeking the control of various ills that harm the productive animals, as the control and treatment of mastitis and infestation by endo and ectoparasites (Pimenta & Pires, 2008). *Rhipicephalus*

(Boophilus) microplus is considered the most harmful ectoparasite for the world's cattle raising. This parasite can be found in some hosts, but cattle is the most affected (Spickler, 2007). This specie causes blood spoliation through the hematophagism, damages the peltry, and also is the main transmitter of pathogenic agents for the bovines (Bittencourt et al, 1998). This tick presents a high resistance to the synthetic acaricides used nowadays (Chagas, 2002). The modern synthetic acaricides affect the environment and leave residuals in the milk, causing the necessity of the search for alternatives. Phytotherapy is a low cost option to the use of synthetic products, making the family dairy farming more viable (Arcego, 2005). The plants have been an important source of substances with different chemical structures and with many actions against arthropods (Vivan, 2005). Therefore, it is believed that the use of vegetal extracts in an isolated or associated form can provide a much lower development of the resistance (Broglio-Micheletti, 2009). A preliminaryethnoveterinary study in farms of some districts in Ecological Corridor of Mantiqueira, found the use of the *Polygonumhydropiperoides*Michaux., popularly known as "Erva de bicho", by the farmers of that region to cattle tick control. *P. hydropiperoides* is aherbaceous speciebelonging to Polygonoaceae family, very common in the Minas Gerais state. In the folk medicine it is used as anti-inflammatory, anti-hemorroidal and anti-diarrheal (Gupta, 1995; Correa, 1969). The aim of the present study was to evaluate and compare the acaricidal effect of the infusion of stalks and leaves of *Polygonumhydropiperoides* on the biological parameters of *R. microplus*.

MATERIALS AND METHODS

The herbal material was collected in the Medicinal Garden of Universidade Federal de Juiz de Fora (UFJF) and taken to the Laboratory of Phytochemistry of the Instituto de Ciências Biológicas /UFJF for drying at room temperature and preparation of the extracts. An voucher specimen was deposited in the CESJ/UFJF herbal. The preparation of aqueous extract was made by infusion of the selected parts, since this is the method utilized by the farmers. The aqueous extracts of stalks and leaves of *P. hydropiperoides* were prepared separately by infusion at 2.5 and 2% m/v, respectively, followed by lyophilization. The aqueous solutions for the tests were prepared at 200 mg/mL of leaves extract (EFPH) and stalks extract (ECPH) of *P. hydropiperoides*. The *in vitro* acaricide activity of the extracts were evaluated in the Laboratory of Parasitology of Embrapa Dairy Cattle on engorged females of *R. (B.) microplus* originated from São João Del-Rey-MG, Brazil. According Drummond et al (1973), the females were immersed for five minutes into the vegetal extracts. After immersion the females were placed in Petri's plates for oviposition in climatized cameras at $27 \pm 1^\circ \text{C}$ and relative humidity higher or equal to 80%, for 14 days. The eggs were collected, weighed and transferred to syringes, which were capped with hydrophilic cotton, similarly kept in acclimatized cameras at $27 \pm 1^\circ \text{C}$ and relative humidity higher or equal to 80%. The larval hatching was visually verified after 20 days. The negative control group was treated with distilled water and the positive ones were treated with acaricides available in the Brazilian market and commonly used by the cattle breeders, pertaining to the chemical groups of the amidine, organophosphate and pyrethroids. The egg mass weight and the hatching

percentages of control and treated groups were submitted to the variance analysis (ANOVA) considering $p < 0.05$. The estimated reproduction (ER) and the effectiveness index of the product or of the vegetal extract (EP) are calculated according to Drummond et al. (1973).

$$ER = \frac{\text{egg-mass weight} \times \text{hatching \%} \times 20,000^*}{\text{Weight of the engorged ticks}}$$

* Constant that indicates the number of eggs present in 1g of egg mass

$$EP = \frac{(\text{ER control} - \text{ER treated}) \times 100}{\text{ER control}}$$

RESULTS AND DISCUSSIONS

The stalk extract of *P. hydropiperoides* reduced significantly the egg mass weight ($p < 0.01$) and presented small reduction of the hatching percentage, not being significant. The leave extract did not present significant reduction in none of the parameters, although it has presented small reductions for both (Table 1). The effectiveness of the leave extract of *P. hydropiperoides* was of 50.15%, while that of the stalk extract was of 76.24%. In accordance with the classification of the effectiveness index proposed by the World Association for the Advancement of Veterinary Parasitology (Coles et al., 1992), a commercial chemical product is highly effective if it presents more than 90% of action against the treated parasite, moderately effective when acts in between 80% and 90%, little effective if the action stands between 60 and 80% and non-effective for levels under 60%. Therefore, according to this index, the aqueous extract of the stalk of *P. hydropiperoids* has shown little effectiveness and the aqueous extract of the leave was non-effective. However, it must be considered that the tested population was resistant to pyrethroids and to amitraz, and sensible to some associations between organophosphates and pyrethroids. The results point to a potential acaricide effect, although tests with different preparation methods, different seasons for collection and different culture conditions become necessary.

Table 1 Egg mass weight, hatching percentage and estimated reproduction of *Rhipicephalus microplus* exposed to *Polygonum hydropiperoides* extracts

	Egg mass weight (mg)	Hatching percentage (%)	Estimated Reproduction (%)
Control	103,00	96,00	45,93
EFPH	57,7	86,14	22,89
ECPH	37,1**	62,29*	10,91

EFPH=Aqueous extract of leaves of *P. hydropiperoides*; ECPH=Aqueous extract of stalk of *P. hydropiperoides*. *- p<0,05; ** - p<0,01

CONCLUSION

The results obtained point to a potential acaricidal effect of both tested extracts of *P. hydropiperoides* against *R. (B.) microplus*, being the aqueous extract of the stalk more effective. This work also suggests that new studies must be conducted to isolate and quantify the chemical compounds responsible by the acaricidal effect of *P. hydropiperoides* leaves and stalk

REFERENCES

- Arcego, M. S. C. Plantas Medicinais no Controle de doenças no gado leiteiro. São João da Urtigas/RS: EMATER, 2005. 11 p.
- Broglio-Micheletti, S.M.F. et al. Extratos de plantas no controle de *Rhipicephalus (Boophilus) microplus* (Canestrini, 1887) (Acari: Ixodidae) em laboratório. Rev. Bras. Parasitol. Vet., Jaboticabal, v. 18, n. 4, p. 44-48, out.-dez. 2009.
- Chagas, A.C.S et al. Efeito acaricida de óleos essenciais e concentrados emulsionáveis de *Eucalyptus* spp em *Boophilus microplus*. Braz. J. Vet. Res. Animal. Sci., São Paulo, v.39, n.5, p.247-253, 2002.
- Coles, G. C.; Bauer, C.; Borgsteed et. al. World Association for the Advancement of Veterinary Parasitology (W.A.A.V.P.) methods for the detection of anthelmintic resistance in nematodes of veterinary importance. Veterinary Parasitology, 1992. 44: 35-44.
- Correa, M.P. Dicionário das plantas úteis do Brasil e das exóticas cultivadas. Rio de Janeiro: Ministério da Agricultura. 1969.
- Drummond, R. O.; Ernest, S. E.; Trevino, J. L.; et. al. *Boophilus annulatus* and *B. microplus*: laboratory tests of insecticides. Journal of Economic Entomology, 1973. v. 66, n. 1, p. 130-133.
- Gupta, M.P. 270 Plantas medicinales iberoamericanas. Santafé de Bogotá: Presencia. 1995.

- MINISTÉRIO DA SAÚDE MS. [2006] Política Nacional de Plantas Medicinais e Fitoterápicos. Brasília. Disponível em <<<http://dtr2004.saude.gov.br/dab/docs/publicacoes/geral/pnpmf.pdf>>> data de acesso 26/03/2009.
- Pimenta, D. S.; Pires, M. F. A. A fitoterapia na pecuária de leite. In: Diniz, F. H.; Ferreira, J. R.; Souza, A. D.; Albuquerque, L. C.; Fagundes, R. B. S.. (Org.). Sustentabilidade da produção de leite na agricultura familiar. Juiz de Fora, MG. Embrapa Gado de Leite., 2008, CD.
- Spicler, A. R. *Rhipicephalus (Boophilus) microplus*. College of Veterinary Medicine Iowa State University. jul. 2007. Disponível em: <http://www.cfsph.iastate.edu/Factsheets/pdfs/boophilus_microplus.pdf>. Acesso em: 10 jan. 2012.
- Vivian, M. P. Uso do cinamomo (*Meliaazedarach*) como alternativo aos agroquímicos no controle do carrapato bovino (*Boophilusmicroplus*). Florianópolis, 2005. 72 p. Dissertação (Mestrado) – Universidade Federal de Santa Catarina.