Susceptibility of Boophilus microplus (Canestrini, 1887) (Acari: Ixodidae) to seven ixodicides in Nuevo Leon, Mexico

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Abstract

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Animal husbandry and meat industry in Mexico are one of the principal activities of the agricultural sector of the country, which are threatened by factors that affect the production of meat, skin and milk; among these, the damages caused by *Boophilus microplus* (Canestrini), the common tick of cattle, and the diseases it transmits, such as anaplasmosis and babesiosis (Xianxun and Wenshun, 1997; Yeruhan et al., 1998), are of special interest. The objective of this study was to determine the susceptibility of the larvae of *Boophilus microplus* to seven tick-killing agents commonly used in the state of Nuevo Leon. The methodology used for the diagnosis of the susceptibility of *B. microplus* tick to Organochlorine, Organophosphorate and Pyrethroid compounds was the one applied by Rodriguez-Vivas et al., (2007); in which it was used a discriminant dose (table 1), using the larvae package test technique (Stone & Haydock, 1962). The results of the CL_{50} to the ticks of cattle in the state of Nuevo Leon, Mexico showed a greater susceptibility to Deltamethrine, followed by Chlorfenvinphos; in third place Diazinon, then Flumethrin; after that Cypermethrin, in sixth place Lindane and a less toxicological action for the Coumaphos (Asuntol). The result was already expected due to the fact that it is the most commonly used product in the control of ticks through the larval immersion technique. The

Asuntol was used in the strain of El Bisonte Ranch of Montemorelos, Nuevo Leon, which is a homogeneous population and with certain short-time pressure could become resistant ticks.

key words: Rhipicephalus (Boophilus) microplus tick is susceptible to pesticides in the state of Nuevo Leon, Mexico.

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Introduction

Boophilus microplus ticks are one of the most important hematophagous ectoparasites that exist in cattle (Guglielmone et al., 2004; Guglielmone et al., 2003; Hernandez, 2005). Not only for the direct damages they cause by feeding themselves but for the transmission of pathogens such as anaplasmosis and babesiosis (Guglielmone et al., 2004; Guglielmone et al., 2003), the production of milk, meat and skin is affected indirectly (Bock et al., 2008; FAO, 1984; Kocan et al., 2008; Lopez & Vizcaino, 1992).

B. microplus is distributed in America, causing significant annual economic losses in cattle (Benavides, 2001; Lopez, 1990; Späth et al., 1994; Späth et al., 1994a). In Mexico it was estimated that ticks and illnesses transmitted by them produced losses of approximately 48 million dollars annually. Because of this the use of acaricides was intensified, resulting in the emergence of tick's resistance to synthetic chemical products like pyrethroids and organophosphorates (Fragoso & Soberanes, 2001; SAGARPA, 2006). In the state of Nuevo Leon there are no recent studies that allow us to know the condition of susceptibility to chemical products used for the control of ticks; because of this, the objective of this investigation is to determine the demographic fluctuation of ticks associated to cattle in their different types of use. Besides, to know the condition of susceptibility of *Boophilus microplus* (Acari: Ixodidae) to the tick-killing agents commonly used in the state of Nuevo Leon.

Materials and Methods

The samples of cattle tick *B. microplus* were obtained from five cattle ranches in the state of Nuevo Leon (Terrabel, Bisonte, El Pajaro, Valle Escondido and 14 de Mayo) because of the concern of the cattle ranchers that these ectoparasites presented resistance to the products used in their control. In the collection, the procedure used by Bennett (1975) and Hernandez (1978) was used, in which we passed our hand softly over the different regions of the body of the host where usually these parasites are located: mammary regions, back legs, haunches, flanks, abdomen, ribs, and front legs including armpits, neck and four fronts, jowl and head.

We placed 10 gravid ticks in a Petri dish in the incubation stove in darkness at a temperature of 28 ± 2° C, with a relative humidity of 80 to 90 % for 14 days until obtaining oviposition (Cen et.al., 1998). The obtained eggs were deposited in crystal vials of 30 ml volume, 90 mm height, and 25 mm diameter covered with cotton under the same conditions of temperature and humidity for 26 days in order for the larval hatching to be presented.

The procedure followed for the diagnostic of the susceptibility of *B. microplus* tick to Organochlorine, Organophosphorate and Pyrethroid compounds was the one applied by Rodriguez-Vivas et al., (2007) in which it was used a discriminant dose (table 1), along with the larvae package test technique applied by Stone and Haydock (1962), which was recommended by the United States Department of Agriculture (USDA) (Miller 2007, personal communication).

Family	Compound	Discriminant Dose
ORGANOCHLORINE	Lindane	0.1 %
ORGANOPHOSPHORATE	Coumaphos	0.2 %
	Diazinon	0.08 %
	Chlorfenvinphos	0.2 %
PYRETHROID	Flumethrin	0.01 %
	Deltamethrin	0.09 %
	Cypermethrin	0.3 %

Table1. Discriminant doses for the control of *Boophilus microplus* ticks recommended by the National Center of Services of Verification in Animal Health CENAPA.

In the technique, number one Whatman paper of 7.5 x 8.5 cm was used, which was sprayed with Finney (1971) dose. The impregnation of the envelopes of the larvae package was done with a graduated micropipette of 1000 microliters capacity, being used with 0.67 microliters of the dilution of the filter papers soaked with the tick-killing agents. In all of the bioessays three repetitions were done for every dilution, including the controls, beginning with the highest concentration and finishing with the lowest.

Once the papers were dry, they were folded in the form of a package, closing them with snaps of 55 mm. Between 80 and 100 7 to 14 days old larvae approximately, were introduced with a brush in each package and later sealed with a snap. These larvae packages were incubated at a temperature of 28 ± 2° C and relative humidity of 80 to 90 % for 24 hours. At the end of the incubation or exposition time, alive and dead larvae were counted to determine the mortality percentage.

The mortality data were analyzed with the program “Statistical Package for the Social Science” (SPSS, Ver. 17), which provided measurements of the Medium Lethal Concentration (CL_{50}) and the Lethal Concentration 99 (CL_{99}) and their respective confidence limits to 95 % (table 2).

The obtained results of CL_{50} (figure 1), showed a greatest susceptibility to Deltamethrin, followed by Chlorfenvinphos, in third place Diazinon, then Flumethrin, Cypermethrin, in sixth place Lindane and a minor toxicological action for the Coumaphos (Asuntol). These populations are not homogeneous and with certain short-term stress they could turn into resistant ticks.

Tick-killing Agents	CL_{50}	CL_{99}
Deltamethrin	0.00005 ( )	0.0015 (0.001, 0.002)
Chlorfenvinphos	0.008 (0.002, 0.013)	0.030 (0.021, 0.071)
Diazinon	0.008 (0.004, 0.017)	0.027 (0.018, 0.078)
Flumethrin	0.0027 (-0.003, 0.006)	0.015 (0.009, 0.072)
Cypermethrin	0.017 (0.028, 0.056)	0.085 (0.051, 0.815)
Lindane	0.012 (0.010, 0.013)	0.035 (0.031, 0.041)
Coumaphos (Asuntol)	0.010 (0.009, 0.012)	0.035 (0.031, 0.042

Table 2. Lethal Concentration 50 (Cl_{50}) and 99 (Cl_{99}) with their confidence limits to common use tick-killing agents in *Boophilus microplus*.

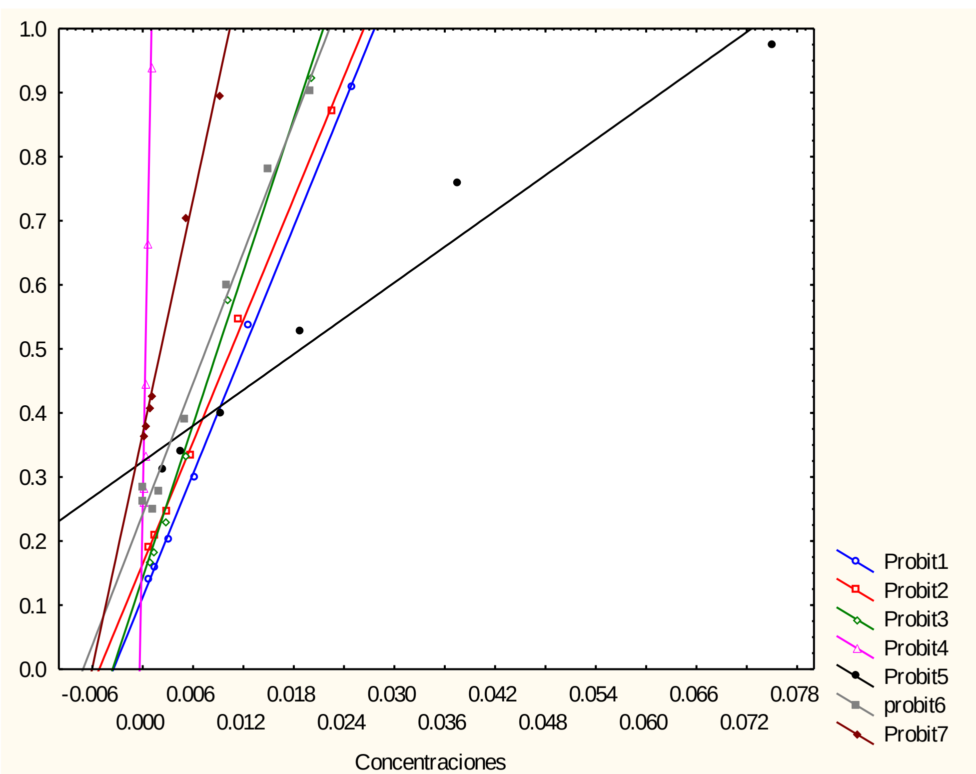

Figure 1. Lines of mortality dose obtained for *Boophilus microplus* for Lindane, Coumaphos, Diazinon, Deltamethrin, Cypermethrin, Chlorfenvinphos and Flumethrin.

Discussion

The results of the present study showed varying degrees of susceptibility of *B. microplus* to the evaluated tick-killing agents, even when a minor response was obtained with Asuntol, expected result as it is the most common product in the control of ticks through the larval immersion technique (Aguirre et al., (1986); Ortiz et al., 1994; Rodríguez et al., 2005; and Marin, 2005), not being detected resistance in this species of ectoparasite.

The obtained results of CL_{50} showed a greater susceptibility to Deltamethrin, followed by Chlorfenvinphos, in third place the Diazinon, then Flumethrin, Cypermethrin, in sixth place the Lindane and in a minor toxicological action for the Coumaphos (Asuntol). These populations are not homogeneous and with certain short-term stress they could turn into resistant ticks. Because of these results it is advisable to use other ixodicides family and appeal to integral ticks control methods (Rodriguez et al., 2005).

On the other hand, Kunz and Kemp (1994) mentioned that the development and the level of susceptibility to ixodicides in ticks populations depends on the frequency of resistant individuals in the population and the intensity of the selection stress exerted by the tick killing agent. These must be used in a rational way in ranches with populations classified as susceptible, considering that the ticks' resistance to ixodicides persists up to 32 years (Davey et al., 2006).

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