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Short communication

Development of amitraz resistance in field populations of *Boophilus microplus* (Acari: Ixodidae) undergoing typical amitraz exposure in the Mexican tropics

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Abstract

The purpose of this study was to determine the effect of amitraz selection pressure on the development of resistance in field populations of *Boophilus microplus* in the Mexican tropics. Three farms (FA_1, FA_2 and FA_3) in Yucatan, Mexico, were selected in this study. Amitraz was applied as a whole body sprays to all cattle on each farm for tick control once monthly for 15 months. From each farm, 20–30 *B. microplus* engorged females were collected every 3 months. The modified larval immersion test was used to test susceptibility of *B. microplus* to amitraz. Larvae were exposed to serial dilutions of amitraz. Probit analysis was used to determine lethal dose at 50% mortality and associated 95% confidence limits. The resistance factors found in the three farms during the 15 months of amitraz selection pressure were FA_1 (1, 2, 4, 4 and 13), FA_2 (1, 6, 23, 21 and 22) and FA_3 (2, 13, 2, 6 and 6). It is concluded that amitraz selection pressure on field populations of *B. microplus* increased the resistance level in all populations studied in the Mexican tropics.

Keywords: Resistance; Acaricides; Amidine; Amitraz; Resistance ratio; *Boophilus microplus*; Mexican tropics

1. Introduction

Boophilus microplus (Canestrini) is an endemic cattle pest in tropical and subtropical regions of the world, causing major economic losses to cattle producers directly through feeding on parasitized cattle and indirectly by transmitting several disease-causing pathogens to the host (*Babesia bovis, Babesia bigemina* and *Anaplasma marginale*) (Solorio et al., 1999; Rodriguez-Vivas et al., 2004).

Acaricides have played a pivotal role in the control of *B. microplus*. However, as a consequence of extensive use of chemicals this tick specie has developed resistance to most of the major classes of acaricides in several countries. In Mexico, the first case of organophosphate (OP) resistance was detected in *B. microplus* ticks from a ranch near Tuxpan in the state of Veracruz in 1983 (Aguirre and Santamaría, 1986). Soon

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after, resistance to OPs became widespread in central and eastern Mexico. In 1986, synthetic pyrethroid (SP) acaricides were introduced into Mexico in order to alleviate OP resistance problems. Resistance to SPs was first detected in 1993 and soon became extensive (Fragoso et al., 1995). As a result resistance to both acaricide classes has developed in this cattle pest in at least 15 states of Mexico (Santamaría et al., 1999).

The formamidine, amitraz was introduced to control OP-resistant ticks at the same time the SPs were introduced in 1986, but its use was initially limited due to higher cost. The use of amitraz became more frequent after 1993 when SP-resistance started to reduce tick control efforts in Mexico. The first case of amitraz resistance was confirmed in 2001 from a farm near to Emiliano Zapata, Tabasco and is now widespread in the Mexican tropics (Soberanes et al., 2002). In southern Mexico, amitraz is the principal acaricide used to control ticks on cattle (Rodriguez-Vivas et al., 2006a).

The development of acaricide resistance in a tick population is dependent on the frequency of resistant individuals in the population and the intensity of chemical selection pressure (Kunz and Kemp, 1994). Although rapid onset and development of *B. microplus* resistance to SP, OP (Davey and George, 1998, Davey et al., 2003) and amitraz (Li et al., 2004) have been observed *in vitro* and *in vivo* (controlled laboratory trials); the frequency of acaricide resistance development on field populations under typical operational conditions has not been reported to our knowledge. The purpose of the present study was to determine the effect of amitraz selection pressure on the development of amitraz resistance in field populations of *B. microplus* in the Mexican tropics under typical tick control regimes and cattle production conditions.

2. Materials and methods

2.1. Study background

The study was conducted in Yucatan, Mexico, located between 19°30′ and 21°35′ North latitude and 90°24′ West longitude of the Greenwich meridian. Climate is generally sub-humid tropical with two seasons: rainy (June–October) and dry (November–May). The monthly maximum temperature varies from 35 °C to 40 °C (mean 26.6 °C). The relative humidity (RH) varies from 65% to 100% (mean 80%) and the annual rainfall varies from 415 mm to 1290 mm depending on the area (INEGI, 1996).

2.2. Production system and use of acaricides in Yucatan

The predominant livestock-production system is semi-intensive (beef farms), based mainly on year-round grazing on improved pastures, e.g., Guinea grass (*Panicum maximum*) and Star grass (*Cynodon plectostachyus*), with supplementary feeding during the dry season. The use of acaricides to control ticks is a common practice in Yucatan, Mexico (Solorio et al., 1999). Forty-one percent of the farms in the Yucatan state use amitraz to control ticks (Rodriguez-Vivas et al., 2006b), with 42% of those farms applying acaricides >12 times/year.

2.3. Study population

Based on the results of a survey of acaricide susceptibility in Yucatan, Mexico, by Rodriguez-Vivas et al. (2006a), three cattle farms were selected for this study (FA_1, FA_2 and FA_3). Each farm had a population of 80–200 *Bos indicus* and *Bos taurus* cross-bred cattle (approximately 60% cows, 4% bulls, 20% calves and 16% yearly stockers) utilizing a semi-intensive livestock-production system with stocking density of 0.60–0.62 animal unit/ha.

2.4. Acaricide management and sampling

All animals on each farm were treated with amitraz (Taktic® E.C. 12.5% [A.I.], Intervet, Mexico), as a whole body spray at the recommended dose using at least 4 l of total finished spray volume per animal. Treatments at the three farms were carried out monthly for a period of 15 months. During this period, five *B. microplus* generations were expected to develop on farms (Rodriguez-Vivas and Dominguez, 1998).

From each farm, 20–30 *B. microplus* engorged females were collected from at least 10 bovines at months 0, 6, 9, 12 and 15 of the experiment.

2.5. Dose–response bioassays

Engorged adult females were placed into small plastic boxes with air holes and transported to the parasitology laboratory at the College of Veterinary Sciences (FMVZ-UADY). Upon arrival, engorged adult females were placed on Petri dishes and incubated at laboratory conditions, at 27 ± 1.5 °C and a RH of 85–86% (Cen et al., 1998). After oviposition, eggs were transferred into 10-ml glass vials with a cotton cap. Eclosion of larvae occurred approximately 30 days after