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GENETIC AND ENVIRONMENTAL FACTORS INFLUENCING THE RESISTANCE OF TERMINAL CROSS CALVES TO TICK *Rhipicephalus (Boophilus) microplus* AND HORN FLY *Haematobia irritans*

[FACTORES GENÉTICOS Y AMBIENTALES QUE INFLUYEN EN LA RESISTENCIA DE BECERROS DE CRUZA TERMINAL A LA GARRAPATA *Rhipicephalus (Boophilus) microplus* Y LA MOSCA DE LOS CUERNOS *Haematobia irritans*]

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SUMMARY

One of the main problems affecting cattle production in tropical regions is infestation by ectoparasites, particularly the tick *Rhipicephalus (Boophilus) microplus* and the horn fly *Haematobia irritans*. The objective of this study was to evaluate the genetic and environmental factors that influence on the resistance of grazing calves against the tick *R. microplus* and the fly *H. irritans*. The study was carried out in Veracruz, Mexico. The ticks and flies counts were made from September 2007 to March 2008, in 27 eight month-old terminal cross calves. Data were analyzed through ANOVA, with the amount of ticks and flies as the dependent variables. Tick infestation was influenced by genotype ($P < 0.05$), period (months; $P < 0.001$), temperature ($P < 0.001$) and rainfall ($P < 0.05$), but not by sex and relative humidity ($P > 0.05$). Fly infestation was influenced ($P < 0.001$) by sex, genotype, period (months), temperature and rainfall, but not by relative humidity. In conclusion, the genetic and environmental factors did influence infestation by ticks and flies in growing calves.

Keywords: *Rhipicephalus microplus; Haematobia irritans*; resistance.

RESUMEN

Un problema que afecta la producción bovina en regiones tropicales es la infestación por ectoparásitos, particularmente la garrapata *Rhipicephalus (Boophilus) microplus* y la mosca de los cuernos *Haematobia irritans*. El objetivo del estudio fue evaluar los factores genéticos y ambientales que influyen en la resistencia de becerros en pastoreo contra la garrapata *R. microplus* y la mosca *H. irritans.* El estudio se realizó en Veracruz, México. Los conteos de garrapatas y moscas se hicieron de septiembre 2007 a marzo 2008, en 27 becerros de cruza terminal de ocho meses de edad. Se analizó la información mediante ANOVA, y las cantidades de garrapatas y moscas fueron las variables dependientes. La infestación por garrapata fue influida por genotipo ($P < 0.05$), periodo (mes; $P < 0.001$), temperatura ($P < 0.001$) y precipitación pluvial ($P < 0.05$), pero no por sexo ($P > 0.23$) ni humedad relativa ($P > 0.79$). La infestación por mosca fue influida ($P < 0.001$) por sexo, genotipo, periodo (meses), temperatura y precipitación pluvial, pero no por humedad relativa ($P > 0.54$). En conclusión, los factores genéticos y ambientales sí influyeron en la infestación por garrapatas y moscas en becerros en crecimiento.

Palabras clave: *Rhipicephalus microplus; Haematobia irritans*; resistencia.

INTRODUCTION

In tropical regions where ambient temperature and solar radiation are high, and where forage production fluctuates seasonally and parasitic diseases are present, cattle production depends, among other factors, on the productive potential of the animals and their ability to adapt to the surrounding environment. Some of the main problems affecting cattle production are parasite infestations, particularly those caused by ectoparasites,

which are responsible for important losses in the productive indices (Gallardo and Morales, 1999).

There are several genera of ticks that affect cattle productivity, such as *Amblyomma* spp., *Rhipicephalus* spp. and *Dermacentor* spp., the two first being the most frequent throughout the Mexican coast. On the other hand, the most known cattle flies are the stable fly or *Stomoxys calcitrans* and the horn fly or *Haematobia irritans* (Quiroz, 1990).

The tick *Rhipicephalus* (*Boophilus) microplus* and the fly *H. irritans* are the ectoparasites with the greatest economic importance in cattle production in tropical regions (Alonso *et al*., 2007a; Suárez *et al*., 2007). Their negative economic impact has a repercussion on meat and milk production, as well as on the hide quality, besides the possibility for transmission of hemoparasites (Wambura *et al*., 1998; Suárez *et al*., 2007).

The tick *R. microplus* exerts a traumatic, toxic, infectious and spoliation effect in the animals, whereas the fly *H. irritans*, besides of the traumatic effect, causes indirect damages such as stress generated by the infestation, which causes a decrease in meat or milk production.

The life cycle of the tick consists of two stages: 1) the parasitic stage: larva, nymph and adult; and 2) the non-parasitic stage: egg-laying, incubation and hatching (Passos and Furlong, 2002). The optimal conditions for tick development are temperature of 27 to 39 °C and relative humidity of 60 to 80 % (Rodríguez and Domínguez, 1998). With respect to the life cycle of the fly, it parasites the host day and night, and the cycle has an average duration of 10 to 14 days. The fly development is favored at temperatures from 10 to 38 °C, with an optimal development at around 25 °C and relative humidity of 60 to 80 % (Rodríguez and Domínguez, 1998; Maldonado *et al*., 2006). The breed, sex and age of cattle, as well as the season of the year, weather and management, are factors that influence on the infestation caused by both parasites (Cruz *et al*., 2000; Bossi *et al*., 2003; Alonso *et al*., 2007a).

MATERIALS AND METHODS

The study was conducted at the Centro de Enseñanza, Investigación y Extensión en Ganadería Tropical (CEIEGT), owned by the Universidad Nacional Autónoma de México, located in Tlapacoyan, in the State of Veracruz, Mexico, at 105 m altitude. The climate in the region is warm humid Af (m) w (e), with mean annual temperature of 23.6 °C and mean annual rainfall of 1991 mm (García, 1981). Data corresponding to temperature, rainfall and relative humidity were recorded daily from the CEIEGT weather station. With these data, the climatic variables for the sampling period, for both ticks and flies, were established. Cattle were maintained under intensive rotational grazing of African Star grass (*Cynodon nlemfuensis*) and native grasses (*Axonopus* spp. and *Paspalum* spp.), and followed the appropriate sanitary schedule.

For the present study, the months were grouped into three periods: 1) September-October, 2) November-January, and 3) February-March. A total of 27 male and female calves at 8 months of age were used; the calves were terminal crosses of Holstein x Zebu dams with Black Angus (11), Red Angus (8) and Braunvieh (8) sires.

The count of ticks was carried out by visual inspection according to the technique by Wharton *et al.* (1970); all the ticks ≥ 4 mm long were counted and detached from the animal. The count of flies was made by direct observation (Foil and Hogsette, 1994) on only one side of the animal, duplicating the number at the end for the analysis (Lima *et al*., 2002). In both cases the counts were made every seven days at 7:00 a.m.

Statistical model

An analysis of variance GLM of the statistical package SAS (1990) was used according to the following model:

$$y_{ijklmn} = \mu + G_i + S_j + T_k + RH_l + RF_m + Mes_n + e_{ijklmno}.$$

Where:
y_{ijklmn} = number or flies and number of ticks
μ = overall mean
G_i = effect of the i-th effect of genotype (½ Black Angus, ½ Red Angus and ½ Braunvieh)
S_j = effect of the j-th effect of sex (male - female)
Tk = effect of the k-th temperature
RH_l = effect of the l-th relative humidity
RF_m = effect of the m-th rainfall
Mes_n = effect of the n-th sampling period (September 2007 to March 2008)
$e_{ijklmno}$ = random error of each observation

RESULTS

The probability of association of the variables with the tick and fly infestation is shown in Table 1.

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Table 1. Association of the genetic and environmental variables with tick and fly infestation in terminal cross male and female calves, in Veracruz, Mexico.

Variable	DF	MS tick	P>F tick	MS fly	P>F fly
Sex	1	2732	0.22	81991	0.001
Genotype	2	5797	0.05	136501	<.0001
Period (months)	2	104255	<.0001	150099	<.0001
Temperature	1	32167	<.0001	58025	0.01
Rainfall	1	7397	0.05	231032	<.0001
Relative humidity	1	138	0.79	3121	0.54

DF: degrees of freedom. MS: mean squares

***Rhipicephalus (Boophilus) microplus* tick**

Regarding tick infestation, sex and relative humidity were not significant ($P > 0.05$), whereas genotype and rainfall ($P < 0.05$), as well as period and temperature ($P < 0.001$) were significant. To this respect, tick infestation was not different between the Black Angus or Red Angus crosses (78±5 and 79±6 ticks, respectively; $P > 0.05$), whereas the Braunvieh cross showed fewer ticks (61±6 ticks; $P < 0.05$).

Likewise, the period with the greatest tick infestation was February-March (155±10 ticks), different to September-October (26±15 ticks) and November-January (37±8 ticks; $P < 0.001$). The lowest infestation was observed during the period with the most abundant rainfall, whereas the greatest infestation was observed during the driest period.

When temperature increased over 26 °C the number of ticks decreased to 26±15, whereas in February-March, when the temperature was 22 °C, the number of ticks was 155±10. On the contrary, the most abundant rainfall was observed during November-January (110±56 mm), and the number of ticks in this period was 37±8. The results are shown in Table 2.

Table 2. Effect of the period (months) on the population of *Rhipicephalus (Boophilus) microplus* ticks in terminal cross calves in Veracruz, Mexico.

Period	Observations	Number of ticks	Temperature (° C)	Rainfall (mm)	Relative humidity (%)
Sept-Oct	54	26 ±15[a]	26.4 ± 0.9	271 ± 3.0	77.8 ± 2.8
Nov-Jan	81	37 ± 8[a]	22 ±1.3	110 ± 56	84 ± 2.6
Feb- Mar	54	155 ±10[b]	22 ± 0.2	49 ± 18	78 ± 2.3

[a,b]Different superscript indicates statistical difference ($P < 0.001$).

***Haematobia irritans* fly**

With respect to fly infestation, all the effects were significant ($P < 0.001$), except the relative humidity.

The difference between male (358±9.5 flies) and female (316±10.7 flies) calves was significant ($P < 0.001$); apparently, the males attracted more flies than females, which was more evident in September-October.

The crosses with Black Angus (386±10 flies), Red Angus (326±13 flies) and Braunvieh (299±12 flies) were different among them ($P < 0.001$), contrary to what was observed in the tick infestation. The lighter hair color of the Braunvieh cross attracted fewer flies than the black and red-black striped colors of the Angus crosses.

Fly infestation during September-October (399±32 flies; $P < 0.001$) was greater than the infestation observed during November-January (244±17 flies) and February-March (369±20 flies).

When the count of flies was the lowest (November to January), the rainfall was intermediate in comparison

with the other two periods (110±56 mm), and the relative humidity was higher (89.9±2.6 %) than in the other two periods. Likewise, when the temperature (26.4±0.9 °C) and the rainfall (271±3 mm) increased, the number of flies also increased (399±32 flies). The results are shown in Table 3.

Table 3. Effect of the period (months) on the population of *Haematobia irritans* flies in terminal cross calves in Veracruz, Mexico.

Period	Observations	Number of flies	Temperature (° C)	Rainfall (mm)	Relative humidity (%)
Sept-Oct	54	399 ± 32[a]	26.4 ± 0.9	271.0 ± 3.0	77.8 ± 2.8
Nov-Ene	81	244 ± 17[b]	22 ± 1.3	110 ± 56	84.9 ± 2.6
Feb- Mar	54	369 ± 20[c]	22.4 ± 0.6	49.5 ± 18	78 ± 2.3

[a,b,c]Different superscript indicates statistical difference (P < 0.001).

DISCUSSION

Rhipicephalus (Boophilus) microplus tick

During the period of September 2007 to March 2008, *R. microplus* was present in the animals, showing variability in the infestation levels. These results were similar to those obtained by Alonso *et al.* (2007a) and González-Cerón *et al.* (2009) in the humid tropic of Veracruz, Mexico, to the observation by Labruna and Veríssimo (2001) in Sao Paulo, Brazil, and to the reports by Quijada *et al.* (1997) in Venezuela, where the changes in tick populations were dependent on the diversity of climatic conditions in each region.

In the present study, the greatest tick populations were observed during the period with the lowest rainfall and vice versa. These results were similar to those reported by Quijada *et al.* (1997), who observed a greater tick infestation during the dry season (January to March) and a decrease in the number of ticks during the rainy season; it was also similar to the observation by Gallardo and Morales (1999), of a greater infestation by *Boophilus microplus* in May and October, when the rainfall was scarce, and it also agreed with the results obtained by Alonso *et al.* (2007a), who found the lowest tick infestation in January and November, and the greatest infestation in May, and they also observed that the abundant rainfall caused a decrease in tick population, since in the months when rainfall was > 200 mm the amount of ticks decreased, and when rainfall was 100 to 150 mm it favored the infestation of the animals. However, these results were contrary to those obtained by González-Cerón *et al.* (2009), of a high correlation between rainfall and number of ticks. This differs to the observation by Álvarez *et al.* (2003) in Costa Rica, where the average count of ticks was not different during the rainy and dry seasons.

The cattle genotype is another factor that influences the incidence of *R. microplus* (Álvarez *et al.*, 2003; Alonso *et al.*, 2007a). In the present study, when the different genotypes of the calves were evaluated, it was observed that the ½ Braunvieh was the least susceptible to tick infestation, followed by the ½ Red Angus. The most susceptible genotype to infestation by *R. microplus* was the ½ Black Angus. Utech *et al.* (1978) provoked an artificial tick infestation in cattle, and they observed that the zebu breeds were more resistant to the infestation than the European breeds. Likewise, Paiva (2004) during a natural tick infestation, indicated that the Nelore (*Bos indicus*) cattle showed greater resistance to *R. microplus*, whereas the Holstein (*Bos taurus*) cattle showed the highest susceptibility to the infestation.

Álvarez *et al.* (2003) and Alonso *et al.* (2007a) stated that the terminal cross cattle (¾ European x ¼ Zebu) are more susceptible to tick infestation than the F1 (½ European x ½ Zebu) cattle. This suggests that the higher the percentage of Zebu genotype in the cattle, the less susceptibility towards this parasite, which consequently may contribute to diminish the losses caused by high infestations in the herd. In Costa Rica, a greater susceptibility to the tick was observed in cattle herds with predominance of *Bos taurus* genotype, different to the cattle herds which were based on Zebu breeds (Álvarez *et al.*, 2003).

Several authors (Maldini, 1992; Tawah, 1992; Jonsson, 1997; Wambura *et al.*, 1998) have mentioned the high resistance (93 %) of purebred Zebu cattle (African and

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Asian) to ticks (*Boophilus spp.*), compared with *Bos taurus* cattle. The inheritance of resistance to the tick is high, up to 80 % in cattle with more than 50 % of *Bos indicus* genotype, and 40 % in animals with a lower percentage of this genotype. This resistance has been determined by exposure of the animals to areas that are endemic to ticks.

***Haematobia irritans* fly**

Haematobia irritans was continuously present from September 2007 to March 2008 with certain changes in the range of infestation, similar to the observations by other authors (Almazán *et al*., 2001; Barros, 2001; Quijada *et al*., 2002; Pruett *et al.*, 2003; Bianchin *et al*., 2006; Alonso *et al.*, 2007b). The greatest amount of flies was observed from September to October, when the population peak occurs, and decreased in December, when temperature was 22.5 °C and rainfall dropped off to 40 mm. Similar observations were reported in Tamaulipas, Mexico in grazing calves, where *H. irritans* was continuously present throughout the year, with the greatest infestation in September, and the lowest in August, when temperature was higher than 20 °C and rainfall drastically decreased (Almazán *et al*., 2001). Likewise, Alonso *et al.* (2007b) observed the greatest amount of flies during August to November (rainy season), and the lowest populations were coincident with the dry season (March to June). On the contrary, Bianchin and Alves (2002) mentioned that when it rains over 100 mm in a short period of time the fly infestation diminishes, perhaps because when it rains a lot within few days the feces spread in the ground, interrupting the life cycle of the fly as it lays its eggs in the feces, and these are easily fragmented by the action of the rain, which destroys the favorable environment for the growth and development of the insect.

In contrast, in the dairy belt of Aguascalientes, Mexico, the fly distribution is seasonal, with the greatest infestation during summer and autumn. During winter, the fly disappears (diapause state) for around 60 days and starts its biological activity at the beginning of spring (end of March) (Cruz *et al*., 2000). This diapause behavior was present in a warm region of the Mexican high plateau that Maldonado *et al*. (2006) studied, where fly infestation was almost continuous, except during two weeks at the beginning of winter (middle January), and the changes observed in the fly population were mainly related with the fact that outside temperature during winter was 0 °C.

The results of the present study demonstrated that the diapause in this region does not affect all the fly populations, as insects were observed parasiting the animals during the months of the evaluation. This could be because temperature was not very low during winter.

In the present study, the color characterized the genotypes of the calves used, and the animals with dark skin were more infested by *H. irritans* than the animals with lighter skin color. Apparently, the fly prefers the heat of the animal body and the black skin retains more heat than skin of lighter colors, and it appears to be more attractive to the insect. This result is coincident with those of Quijada *et al*. (2002) and Pruett *et al.* (2003), who observed that animals with dark skin suffered more by the attack of *H. irritans* compared with those with skin of lighter color, but it differs from the report by Guglielmone *et al*. (2002), who found no significant differences in the number of adult flies in Holstein (black and white coat) and Holstein x Jersey (black coat) steers, even in the months of greatest infestation.

Although in this study the sex had no influence on tick infestation, in the case of the fly it showed some affinity towards males, which resulted similar to the observations by Bianchin and Alves (2002), that the amount of flies was greater in males, in comparison with the cows and their calves, indicating the preference of the fly for the adult males. In the study of Hughes and Randolph (2001) with rodents, the males resulted more susceptible than the females to tick infestation, because of the presence of testosterone in the adult males, and these authors suggested that this could also be the case with other ectoparasites such as the fly.

The distribution of ticks and flies in the present study clearly demonstrated that during the periods of more abundant rainfall *R. microplus* is absent and *H. irritans* is present, and when rainfall drops off *R.. microplus* is present and *H. irritans* is also present in its second peak of infestation (February). These distributions would allow to strategically plan their control throughout the year, although it would be convenient to know the behavior of both insects during the whole year.

The genetic and environmental factors studied had a significant influence on the infestation of the calves. The breeds used for the terminal crosses were important for the infestation by both parasites, as was also the coat color of the calves. The environmental variables have always influenced the behavior of parasites, both of which greatly depend on temperature, rainfall and humidity for their

development, according to the region where they are found.

For ticks and flies, the results of the different studies differ with respect to the seasons of infestation and the climatic variables, and the life cycles vary according to the regions where the insects are located.

CONCLUSION

The breed used in the terminal crosses of the calves influenced the infestation by both parasites, mainly the fly, which showed preference for the coat color, and as a result the Red Angus and Black Angus were the most highly infested. The combination of temperature with rainfall played an important role in the infestation by both parasites. Rainfall higher than 200 mm acts as an important factor that regulates tick populations. In the season of high infestation, the previous rainfall favored the development of populations of *H. irritans*.

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