

Article

Sensitivity of Senepol and Caracu breeds to parasitism by *Rhipicephalus microplus*

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Abstract

Taurine breeds and animals crossbred for genetic resistance to *Rhipicephalus microplus* have been the focus of several studies aimed at producing better responses of cattle to tick infestations and seeking to reduce treatment costs and improve the quality of bovine byproducts. Another way to increase gains is the introduction of taurine breeds with the goal of obtaining morphological and physiological characteristics more adapted to production. Although several studies have compared taurine and zebu breeds, little is known about the differences found within the taurine group. Therefore, the present study aimed to evaluate the sensitivity of the Senepol and Caracu breeds to artificial *R. microplus* infestations. For this purpose, a stall test was conducted on 16 Caracu and 14 Senepol bulls. The animals were subjected to artificial infestation with a total of 15,000 *R. microplus* larvae per animal. The Senepol group accounted for 71.7% of the retrieved engorged female ticks. The biological parameters analyzed that showed a significant difference included the mean number of engorged female ticks retrieved ($p = 0.000$), weight of engorged female ticks ($p = 0.000$), rate of engorged female ticks retrieved ($p = 0.03$) and egg mass weight ($p = 0.032$). Thus, the present study demonstrated a higher susceptibility of the Senepol breed to parasitism by the Asian blue tick than the Caracu breed, providing support for new studies on the subjects of genetic resistance, animal breeding and cattle production.

Keywords: Tick, susceptibility, genetic resistance, *Bos taurus*, stall test, infestation, cattle

Introduction

The Asian blue tick (*Rhipicephalus microplus*) (Canistrini, 1887) (Acari; Ixodidae) is distributed in tropical and subtropical areas, and this species is monoxenous and preferentially parasitizes cattle, especially taurine breeds (Utech *et al.* 1978; Piper *et al.* 2010; Ibelli *et al.* 2012; Andreotti *et al.* 2016). The economic losses caused by *R. microplus* in Brazil are estimated to equal 3.24 billion dollars per year (Grisi *et al.* 2014). Other factors involved include the bovine babesiosis and anaplasmosis complex, which consists of a complex involving the protozoa *Babesia bovis* and *Babesia bigemina* and the bacteria *Anaplasma marginale* (Bahia *et al.* 2020) and tick resistance to most chemical classes available for its control (Reck *et al.* 2014; Higa *et al.* 2015; Maciel *et al.* 2016).

The crossing of adapted and non-adapted taurine breeds with zebu breeds allows exploration of heterosis, promoting an increase in performance in the pre- and postweaning phases and in meat quality (Neves *et al.* 2017). In the Brazilian Central-West region, the climatic conditions are favorable

for the survival of *R. microplus* populations in the pasture during the year, which leads to a high rate of infestation in cattle, particularly in taurine breeds and their crosses (Furlong & Evans 1991).

The genetic resistance of cattle to *R. microplus* has been described as an alternative control method that can be complemented with existing alternatives, enabling the control of severe tick infestations using breeds and animals with high genetic resistance to ticks (Frisch 1999). Therefore, it is of great importance to study the sensitivity of cattle breeds to Asian blue ticks.

Thus, the objective of this study was to compare the sensitivity of Senepol and Caracu cattle breeds using a stall test, taking into account the recovery of engorged female ticks and the biological parameters of the *R. microplus* tick.

Materials and methods

The study was conducted at Embrapa Beef Cattle in the municipality of Campo Grande in the state of Mato Grosso do Sul, Brazil (S 20° 44' 25" 76' W 54° 72' 20" 66', 530.0 m) from July to October 2018. A total of 16 Caracu bulls with an average weight of 195 kg and age of 8 months and 14 Senepol bulls with an average weight of 209 kg and age of 8 months were used. Castrated and healthy male animals were used in both groups. All animals were individually identified using numbered ear tags. The cattle were fed twice a day with forage silage, sorghum and corn, with the goal of achieving a weight gain of 200 grams per day, and provided water *ad libitum*. The management processes of the experiment were approved by the Ethics Committee on the use of animals (CEUA n° 699/2015).

Engorged females, from Embrapa Beef Cattle farm, were collected from naturally infested cattle, taken to the laboratory, washed, weighed and placed in flasks properly capped with holes and kept in a biochemical oxygen demand (BOD) (Solab®, Piracicaba-SP, Brazil) oven at 27 °C and 80% humidity to obtain the larvae. After the oviposition period of the engorged females, the egg mass was separated and placed in 5 ml tubes for subsequent hatching. Each animal was artificially infested three times with an interval of one day, and 5,000 larvae (0.25 g of eggs) were used in each infestation, totaling 15,000 *R. microplus* larvae. The infestations were performed through tubes containing the larvae, which were applied to the dorsal line of the animals. Starting from the twenty-first day after infestation, the detachment and recovery of engorged female ticks began using the stall test, according to BRASIL (1997). For 11 days, the engorged females were collected daily, identified according to stall and animal, separated and processed in the laboratory. Thirty-five engorged females were selected from the set of ticks retrieved from each animal to monitor the biological parameters.

Two types of analyses were performed. A general analysis was conducted to evaluate the retrieval rate of engorged females (RREF) based on the total number of engorged females retrieved. The RREF was calculated by the following formula: $RREF = (TNEF/NIL) \times 100$, where RREF = retrieval rate of engorged females, TNEF = total number of engorged females retrieved per animal and NIL = number of infested larvae per animal.

The other analysis was performed on 35 engorged females selected from each animal, which were grouped according to the two breeds, and the following parameters were measured: weight of engorged females (WEF) (g); egg mass weight (EMW) (g), total egg mass from the beginning to the end of laying and reproductive efficiency rate (RER) (%), defined by the formula: total egg mass/initial mass of engorged females $\times 100$ and hatchability rate (HR) (%).

The following classifications were used to determine the parasite intensity: $1 < I < 10$: very low parasite intensity, $10 < I < 50$: low parasite intensity, $50 < I < 100$ medium parasite intensity and $I < 100$: high parasite intensity (Bilong-Bilong & Nijimé 1998). In the statistical analysis, nonparametric Kruskal-Wallis and Mann-Whitney U tests were used to compare the differences in data collected

for the two breeds. The analyses were performed using the statistical software R version 3.6.1 (R Core Team, 2019), and p values less than or equal to 0.05 were considered significant.

Results

A total of 1509 engorged females (28.3%) from the Caracu breed and 3825 (71.7%) from the Senepol breed were retrieved. The Senepol group had a significantly higher number of retrieved engorged females ($p = 0.045$) than the Caracu breed (Figure 1) during the experimental period. The curves had a similar shape but were significantly different regarding the number of ticks, and in both groups, two peaks were observed in the curve of the total count of engorged females retrieved. Therefore, the mean number of engorged females retrieved per Caracu animal during the experiment was $125,7 \pm 65,5$ ticks and the mean number of engorged females retrieved per Senepol animal was $273,2 \pm 193,5$ (Figure 2).

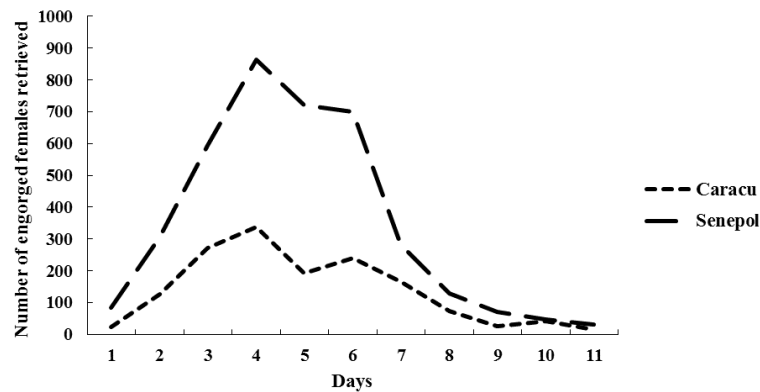


FIGURE 1. Number of engorged *Rhipicephalus microplus* females recovered during the stall test period in both breeds.

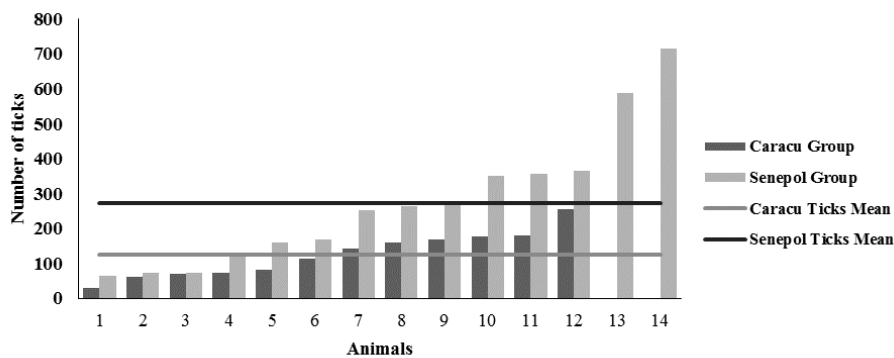


FIGURE 2. Overall mean and number of engorged *Rhipicephalus microplus* females per animal recovered in Caracu and Senepol cattle.

The total number of ticks showed different distributions in the two breeds, where 91,6% of the recovered ticks were in animals with up 200 ticks in the Caracu group, whereas Senepol animals showed 56% of the recovered ticks in animals with more of 251 ticks, exhibiting a higher sensitivity of the Senepol breed. According to the frequency table, this breed had a greater number of animals with a high parasite load and consequently a greater number of engorged females retrieved (Figure 3).

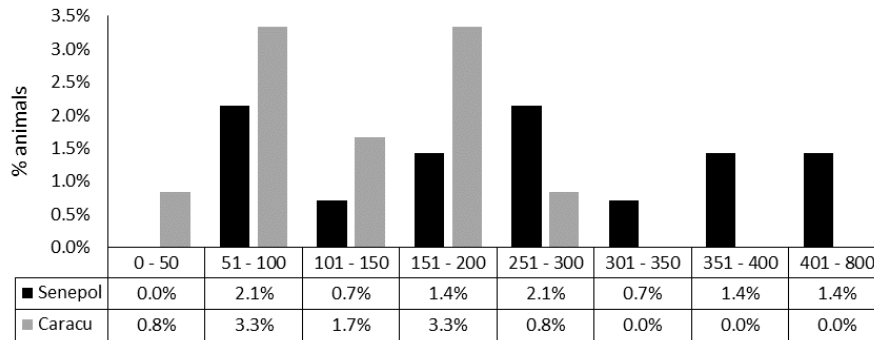


FIGURE 3. Frequency distribution of the number of engorged *Rhipicephalus microplus* females retrieved in Caracu and Senepol cattle.

Senepol animals had a RREF 2.18 times greater than the Caracu group. The Senepol animals did not have low infestation rates similar to Caracu animals, but rather had a higher proportion of animals with high parasitism; 79% of Senepol cattle exhibited high parasitic infestation. In both breeds, the four animals that were most parasitized were responsible for 50% of the total number of engorged females retrieved. Regarding parasite intensity, Caracu animals did not present high parasite intensity, unlike the Senepol group, but animals with a high parasite intensity were observed in both groups, with a higher proportion in the Senepol cattle.

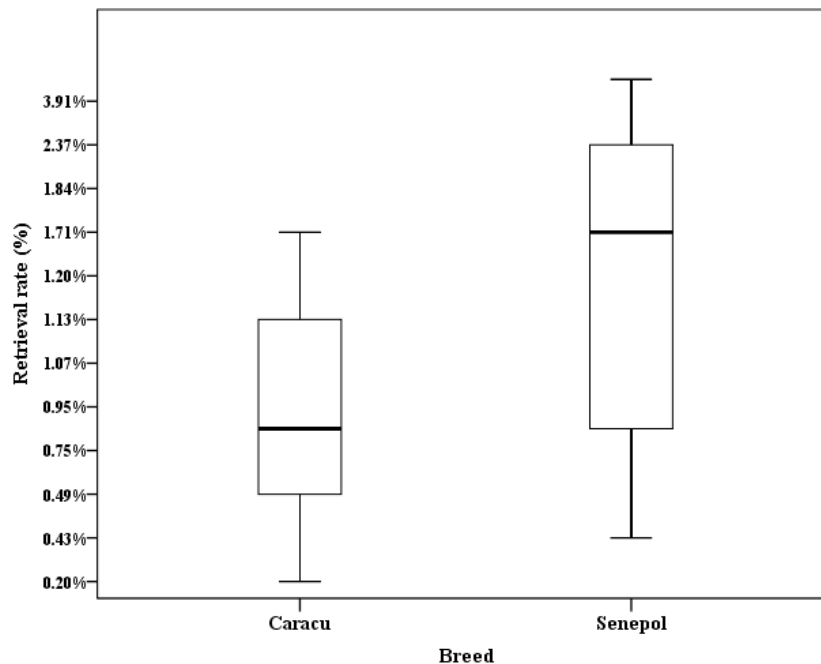


FIGURE 4. The retrieval rate of engorged *Rhipicephalus microplus* females of the Senepol and Caracu breed.

The biological parameters analyzed that showed a significant difference were the mean number of engorged females retrieved ($p = 0.000$), WEF ($p = 0.000$), RREF ($p = 0.03$) (Figure 4) and EMW ($p = 0.032$). No significant difference was observed in the RER and HR (Table 1). The Caracu cattle showed a lower RREF, lower WEF and, consequently, lower EMW than the engorged female ticks retrieved from the Senepol group.

TABLE 1. Biological parameters of engorged *Rhipicephalus microplus* female ticks collected according to breed.

| Breed | Parameters | Mean and standard deviation | CI (95%) |
|---------|------------|-----------------------------|---------------|
| Caracu | WEF * | 0.237 g ± 0.04 g | 0.228–0.245 |
| | RREF * | 0.83% ± 0.43% | 0.560–1.116 |
| | RER | 39.24% ± 20.93% | 35.754–42.856 |
| | EMW * | 0.117 g ± 0.03 g | 0.110–0.124 |
| | HR | 80.9% ± 21.27% | 75.897–85.970 |
| Senepol | WEF * | 0.262 g ± 0.03 g | 0.255–0.269 |
| | RREF * | 1.81% ± 1.28% | 1.074–2.563 |
| | RER | 40.75% ± 19.02% | 37.787–43.610 |
| | EMW * | 0.127 g ± 0.02 g | 0.122–0.132 |
| | HR | 87.75% ± 8.65 | 85.768–89.748 |

WEF = weight of engorged females; RREF = retrieval rate of engorged females; RER = reproductive efficiency rate; EMW = egg mass weight and HR = hatchability rate.

CI = Confidence interval.

* There was a significant difference between the two breeds ($p \leq 0.05$).

Discussion

Rhipicephalus microplus tick infestation has been known to cause adverse effects on cattle production. The resistance of a breed to ticks can lead to a decrease in these losses, and such resistance should be quantified by artificial infestation, thus providing information about the different levels of resistance of the host according to the parasite load (Jonsson 2006).

Taking into account the breed resistance to ticks, the Caracu breed had a lower sensitivity to *R. microplus* than the Senepol breed, both based on the number of engorged females retrieved and their distribution in the individuals. The results indicate a greater number of Senepol animals with high parasite load, WEF and respective EMW. Marques (2003) described that the Caracu breed has advantages in terms of hardiness, resisting the tropical climate, pasture conditions and parasites. Senepol is a breed selected for meat production and is described as a breed that has high ectoparasite infestations in pastures in tropical climates (Marques 2003).

Because of a lower sensitivity to the *R. microplus* tick, *Bos indicus* breeds are of interest for improved tick control through the use of crosses to achieve greater resistance to the Asian blue tick (Sutherst & Utech 1981). Examination of infestations in different genetic groups of *Bos taurus* crossed with Nellore indicated greater resistance to *R. microplus* in a tropical environment (Ibelli *et al.* 2012).

The animals used in this experiment were in the postweaning period; in more sensitive breeds, this period is critical due to the stress caused in the animals, which may favor tick infestation, especially when weaning occurs in the dry season. Junior *et al.* (2019) observed a high infestation rate by counting ticks in Brangus animals in the postweaning period, which occurred in the dry period in the winter. They described the stress and possible decrease in immune resistance, which favors parasitism. Thus, they suggested the implementation of a strategic control program in the Central-West region of Brazil for animals in the rearing phase because high *R. microplus* infestation rates have been observed in Brangus animals. These infestations cause considerable damage because they exceed the economic threshold of 40 ticks described by Gonzales (2003).

In this context, in which the number of ticks in crossbred breeds arouses great interest, Andreotti *et al.* (2018) conducted a stall test with Nellore (n = 10), Brangus (n = 10) and Angus (n = 9) animals,

which were infested with 10,000 *R. microplus* larvae. Mean values of 83 (0.83%), 201 (2.01%) and 615 (6.15%) engorged females and RREF values of 0.83%, 2.01% and 6.15%, respectively, were obtained. In the Caracu and Senepol animals used in this study, with infestation of 15,000 larvae, we obtained an average of 125.75 and 273.2 engorged females and an RREF of 0.83% and 1.81%, respectively; thus, the most sensitive breeds have a higher RREF, demonstrating that the Senepol breed is 3.3 times more sensitive than the Nellore breed and 2.1 times more sensitive than the Caracu breed.

Under natural infestation conditions, Rodrigues *et al.* (2018) obtained daily averages of 121.5 and 14.7 ticks in the first phase of an experiment and 98.7 and 20.7 ticks in the second phase for the Angus and Nellore groups, respectively, demonstrating a greater sensitivity of the Brangus breed. Fraga *et al.* (2003) observed an average of 97.4 engorged females in Caracu females with natural infestation during the experimental period. In the present experiment, a daily average of 137.1 engorged females were retrieved from the Caracu breed and 347.7 were retrieved from the Senepol breed.

According to the daily means for RREF, production of larvae per gram of eggs (1 g = 20,000 larvae) (Pereira 2008), EMW and HR under laboratory conditions, we estimated a larval production 2.9 times higher for Senepol (774,971.10) than for Caracu cattle (259,538.50) in this study. Under pasture conditions, production of 675,000 larvae/day and 4,590,000 larvae/day was described in Nellore and Brangus animals, respectively, according to the count of engorged females in these animals (Andreotti *et al.* 2018).

Despite the production of eggs and larvae, there are some factors that limit their survival, such as egg maturation and incubation, larval survival, temperature, relative humidity and predators (Brovini *et al.* 2003; Pereira 2008). In the southeastern region of Brazil, *R. microplus* has been reported to be capable of completing four annual generations (Pereira 2008). However, a more recent study by Cruz *et al.* (2020) described an increase to five generations per year, indicating a decrease in the nonparasitic phase of the tick, suggesting a better adaptation to climatic and environmental conditions.

Regarding the biological parameters, the data of the present study were similar to those of Santos *et al.* (2002), who divided the engorged female ticks into three groups according to weight: small (0 g–199 g), medium (200 g–299 g) and large (over 299 g). The mean weight of the engorged females in the present study was within the medium weight range, and the EMW was close to that obtained by those authors.

Conclusion

Based on the data analyzed, we can conclude that the Caracu breed showed lower sensitivity to the *R. microplus* tick than the Senepol breed, both according to the RREF and their biological parameters.

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Conflict of interests

The authors declare to have no conflict of interest.

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