

## Effects and costs of a strategic selective treatment for controlling ecto - and hemoparasitosis in Holstein Friesian calves

### Efeito e custos do tratamento estratégico seletivo no controle de ecto e hemoparasitoses em bezerras da raça holandesa

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#### Abstract

This work assessed the effectiveness and net operational costs of a strategic selective treatment for controlling ectoparasites (*Rhipicephalus microplus*, as well as *Dermatobia hominis* and *Cochliomyia hominivorax* larvae) and hemoparasites (*Anaplasma marginale* and *Babesia* spp.) in Holstein Friesian calves. Thirty calves were equally distributed in two groups: ST) strategic selective treatment and CT) conventional treatment. From birth, calves were monitored biweekly until 12 months of age. The CT group was subjected to parasite control according to directions provided by the veterinarian at the Experimental Farm of the Federal University of Lavras (FE/UFLA), whereas parasite control of the ST group followed criteria we had previously established. The ST group experienced fewer changes in physical traits on average and lower globular volume (GV) than the CT group ( $p < 0.05$ ). Approximately 23% of ST calves exhibited external omphalitis, compared with 48% CT calves ( $p < 0.001$ ). Additionally, ST calves were less affected than CT calves ( $p < 0.05$ ) by *R. microplus* females and *D. hominis* larvae. Seropositivity rates from the indirect immunofluorescence reaction for *A. marginale* and *B. bovis* were similar for both groups (ST: 85.6% and 87.8%, respectively; CT: 83.2% and 83.2%). Additionally, subclinical (asymptomatic infection) anaplasmosis predominated in both groups during the dry period of the year. The daily average weight gains (DWG) were identical among ST (580 g) and CT (570 g), but the effective operational cost (EOC) was 3.7 times greater in ST (R\$ 406.58/animal) compared with CT (R\$ 110.90/animal). Laboratory exams to monitor the animals represented the largest share of the cost (82.7%) in ST, whereas drug expenses represented the highest costs (49.8%) in CT. Despite having a greater EOC, ST was more efficient at reducing parasite burdens than CT. Calves in the former group presented fewer *R. microplus* or *D. hominis*, and experienced a lower percentage of hemolytic anemia ( $VG < 24\%$ ). However, ST no affected the dynamic of *A. marginale*- and *B. bovis*-related infections, indicating that the FE/UFLA may be characterized as enzootically stable for these species.

**Key words:** Cattle. *Rhipicephalus (Boophilus) microplus*. *Cochliomyia hominivorax*. *Dermatobia hominis*. *Anaplasma marginale*. Treatment costs.

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## Resumo

Objetivou-se avaliar a eficácia e os custos operacionais efetivos de um tratamento estratégico seletivo no controle de ecto (*Rhipicephalus microplus* e, larvas de *Dermatobia hominis* e *Cochliomyia hominivorax*) e hemoparasitos (*Anaplasma marginale* e *Babesia* spp.), em bezerras da raça holandesa, na Fazenda Experimental da Universidade Federal de Lavras (FE-UFLA), MG. Trinta bezerras foram distribuídas igualmente em dois grupos: TE) Tratamento estratégico seletivo e TC) Tratamento convencional. As bezerras foram monitoradas a cada 14 dias desde o nascimento até os 12 meses de idade. O controle de ecto e hemoparasitos, no TC, foi realizado de acordo com a orientação do médico veterinário responsável pela FE-UFLA. No TE, esse controle de parasitos seguiu critérios, previamente, estabelecidos pelos pesquisadores deste estudo. Nas análises estatísticas utilizou-se o pacote estatístico PASW 18.0. As frequências médias de alterações nos parâmetros físicos e no volume globular (VG) foram menores ( $p < 0,05$ ) no TE, em relação ao TC. As bezerras do TE (23%) apresentaram menos ( $p < 0,001$ ) casos de onfalite externa em relação aos animais do TC (48%). Bezerras do TE estavam menos ( $p < 0,05$ ) parasitadas por fêmeas de *R. microplus* e larvas de *D. hominis* (“berne”) que os animais do TC. Na reação de imunofluorescência indireta (RIFI), as soropositividades para *A. marginale* e *B. bovis*, foram, respectivamente, 85,6% e 87,8% no TE e, 83,2% e 83,2% no TC. Em ambos os tratamentos, predominaram os casos de anaplasmose subclínica (infecção assintomática) no período seco do ano. Os ganhos de peso médios diários (GPD) foram iguais ( $p > 0,05$ ) entre TE (580 g) e TC (570 g). O custo operacional efetivo (COE) foi 3,7 vezes maior no TE (R\$ 406,58/animal) em relação ao TC (R\$ 110,90/animal). No TE, exames laboratoriais para monitorar os animais foi o item com maior representatividade no COE (82,7%), enquanto que, no TC, o maior gasto foi com os medicamentos (49,8%). Apesar de ter um COE maior, o TE foi mais eficiente na redução da carga parasitária em relação ao TC. Bezerras do TE tiveram menor quantidade de carrapatos e “bernes”, e de casos de anemia hemolítica (VG <24%), em relação aos animais do TC. O TE não interferiu na dinâmica das infecções por *A. marginale* e *B. bovis*, e a FE-UFLA pode ser caracterizada, epidemiologicamente, como estável enzooticamente para estas espécies.

**Palavras-chave:** Bovinos. *Rhipicephalus (Boophilus) microplus*. *Cochliomyia hominivorax*. *Dermatobia hominis*. *Anaplasma marginale*. Custos de tratamento.

## Introduction

Effective management practices is critical for controlling the spread of parasitic diseases in dairy farms, the negative consequences of which reflects in Brazil economy through direct and indirect losses in cattle production (BIANCHIN et al., 2007; CHARLIER et al., 2009; VERCRUYSSSE; CLAEREBOU, 2001).

Parasitism from *Dermatobia hominis* (botfly, Brazilian “berne”) and *Cochliomyia hominivorax* (myiasis fly, Brazilian “mosca-da-bicheira”) causes myiasis, a condition that results in scalp damage and lesions predisposing dairy and beef cattle to secondary bacterial infections (BORJA, 2003; HONER; GOMES, 1992). These parasites are ubiquitous in Brazil, and their economic toll on the cattle industry is of utmost importance. For

example, myiasis-fly infections result in an annual loss of 340 million dollars, and botfly infections, around 380 million dollars/year; these outcomes are a combination of decreased milk and beef production, poor weight gain, leather devaluation, as well as labor and drug-use costs in parasitosis control (GRISI et al., 2014).

Even more costly than the botfly or the myiasis-fly is the tick *Rhipicephalus (Boophilus) microplus*. As the primary bovine ectoparasite in Brazil, ticks are responsible for annual burdens of 3.24 billion dollars (GRISI et al., 2014) from stress to cattle, damage to leather, acaricide-related costs, and losses due to tick-borne diseases (“bovine parasitic sadness” in Brazil). For cattle, the latter specifically refer to anaplasmosis and babesiosis, accounting for losses of around 500 million dollars/year (GRISI et al., 2002).

Bovine babesiosis is a hemoparasitosis caused by protozoa of the genus *Babesia*, and in Brazil, *B. bovis* and *B. bigemina*-with *R. (B.) microplus* as vector-are the known etiologic agents. These parasites generally cause fever and apathy anemia, while the most severe forms (from the viscerotropic *B. bovis*) additionally result in neurological damage (KUTTLER, 1988). Bovine anaplasmosis is caused by the transmission of obligate intraerythrocytic bacteria *Anaplasma marginale* by *R. (B.) microplus*, or by hematophagous flies and mosquitoes (GUGLIELMONE, 1995). The disease is characterized by fever, anemia, jaundice, severe weight loss, decreased milk production, abortions, and eventually death (KOCAN et al., 2004).

In dairy farms, calves and heifers are potentially at higher risk of disease because they undergo stressful management practices such as weaning, dehorning, or environmental changes. Thus, it is critical to develop treatment plans successful throughout the bovine life cycle, particularly at vulnerable developmental stages. This work assessed the effectiveness and costs of a strategic selective treatment in controlling the main ecto- and hemoparasites of Holstein Friesian cattle during the early stage of the bovine production system. Our data should contribute to efforts aimed at minimizing the negative economic impact of these parasitic agents.

## Materials and Methods

### *Location and management*

The study was performed from April 2013 to November 2014 at the Experimental Farm of the Federal University of Lavras (FE/UFLA), located in the Ijaci municipality (21° 10' 12" S, 44° 55' 30" W), MG. The climate of the region is characterized by well-defined dry (April to September) and rainy (October to March) seasons. The total average annual precipitation is 1,530 mm and average annual temperature is 19.4°C (BRASIL, 1992; DANTAS et al., 2007). This experiment was approved by

the Ethical Committee of the Use of Animals in Research at the UFLA and registered under the number 106/12.

The herd at the FE/UFLA comprised 51 lactating (averaging 1000 L of milk daily) Holstein Friesian bovines (PO), housed in a *free stall* system. Their diet consisted of corn silage (*Zea mays*) and balanced commercial rations. Experimental calves from these cattle received 4 L of colostrum through a feeding bottle until six hours after birth at maximum. Navel antiseptics was performed for three days using 10% iodine solution, twice a day. In the suckler phase, each animal was housed in individual, roofed stalls (1 x 1.2 m) with wooden dividers arranged in parallel. The stalls were placed in a sunny area and the floors were covered with dry grass. Their solid diet comprised pelleted concentrated food *ad libitum*, in addition to 200 g of milk replacer/day. The liquid diet initially comprised 6 L/day of milk, divided in two equal portions for the morning and afternoon; from the third month of age onwards, 3 L were given in the morning only. *Ad libitum* water was also offered.

Dehorning and excision of supernumerary teats were performed at 20 days of age. Weaning occurred abruptly at 90 days of age, when calves reached approximately 100 kg. The calves were kept in the same stalls for a week more and subsequently moved to the paddocks containing Tifton (*Cynodon* spp.) pasture for the growth phase; they were re-managed periodically to other paddocks according to their age. Calves also had available corn silage and received three kilograms of concentrate food per animal/day. All growth paddocks contained a shaded area of cemented trough and cemented reservoirs for water flow supply. The animals received compulsory vaccinations (foot-and-mouth disease and brucellosis) at the appropriate periods.

### *Experimental animals and treatments*

Thirty Holstein Friesian calves (PO) were selected according to the parturition provisions for

the experiment period and distributed randomly into two groups of 15 animals each: CT = conventional treatment and ST = strategic selective treatment. They were monitored biweekly from birth until 12 months.

In the CT group, generic drugs and antiparasitics were administered according to recommendations and criteria from the veterinarian responsible for the dairy cattle sector at the FE/UFLA. As prevention anaplasmosis and bovine babesiosis, two doses of imidocarb (1 mL 40 kg<sup>-1</sup>) were applied, one during the weaning day and the other after 20 days. Treatment of the anaplasmosis clinical cases comprised applying oxytetracycline LA (20 mg kg<sup>-1</sup>) in two doses, separated by a 24-h interval. Treatment of babesiosis involved applying a single dose of diminazene diacetate (3 mg kg<sup>-1</sup>). Cypermethrin *pour-on* (1 ml 10 kg<sup>-1</sup>) was used for *R. microplus* control, with administration frequency based on visual assessment of parasite burden. The same procedure was used for *D. hominis* larvae control.

Under ST, calves received antiparasitic drugs at schedules selected by the authors: (1) *R. microplus* control - from birth to weaning (90 days), cypermethrin *pour-on* (1ml 10 kg<sup>-1</sup>) if results of the biweekly female *R. microplus* count was  $\geq 20$  ticks/animal, and from post-weaning to 12 months, *pour-on* every 28 days during the dry season (April to September), regardless of age group; (2) *D. hominis* larvae/botfly - from birth to 12 months, ivermectin 1% (0.2 mg kg<sup>-1</sup>) if results of the biweekly nodule count was  $\geq 5$  botfly/animal; (3) prevention of umbilical myiasis (“bicheira”) from *C. hominivorax* larval infection - single dose of doramectin 1% (0.2 mg kg<sup>-1</sup>) applied at birth; (4) anaplasmosis treatment - from birth to weaning, a single dose of enrofloxacin (10 mg kg<sup>-1</sup>) if the biweekly monitor indicated GV <24% and a blood smear positive for *A. marginale*, whereas from weaning to 12 months, three treatments of enrofloxacin every 28 days during the rainy season (October to March), regardless of age group; (5) babesiosis treatment - diminazene diacetate (4 mg kg<sup>-1</sup>) application only in calves with clinical

signs consistent with the disease, a positive blood smear for *Babesia* spp., and GV <24%.

#### *Tick and botfly count*

Counts were performed biweekly during the morning. The calves' right side was used for counting engorged (4.5-8.0 mm diameter) *R. microplus* females, and the total parasite burden was obtained through multiplying the result from each animal by two (WHARTON; UTECH, 1970). For botflies, we inspected and palpated the animals to determine living *D. hominis* larvae and counted all the nodules located on the body.

#### *Blood sample collection*

From birth to 13 months, blood samples ( $\pm 3$  mL) were collected every 28 days through external jugular vein puncture and stored in *vacutainer* tubes containing 1% EDTA. Globular volume (GV) was determined with microhematocrit, and the blood samples were then centrifuged at 500 g for 10 min. An aliquot of plasma ( $\pm 1$  mL) was stored in cryotubes and frozen at -20°C until needed for serology.

Blood smears were prepared with a blood drop obtained from small vessels along the border of the ear region. Smears were dried at room temperature and stained using the quick panoptic method (Laborclin). Subsequently, parasitological diagnosis of the smears was conducted with an optical microscope, under immersion objective (100x). Rickettsia was determined through the percentage of erythrocytes infected by *A. marginale*, observed in 40 homogenous microscopic fields with approximately 200-250 erythrocytes per field.

#### *Clinical score for Anaplasma marginale*

Following a study by Carvalho et al. (2012), the formula proposed by Schetters et al. (2009) was adapted to determine the clinical scores of the *A.*



*marginale*-infected animals. Percentage values from Schettters et al. (2009) were used with adjustments of nomenclature to fit with the present study. Calves were divided into five categories based on clinical score (CS): CS1 = subclinical infection; CS2 = mild clinical infection; CS3 = moderated clinical infection; CS4 = severe clinical infection; and CS5 = very severe clinical infection. Thus, calves with CS1 were considered asymptomatic, whereas bovines with  $CS \geq 2$  were considered to be symptomatic.

#### *Indirect fluorescence antibody test (IFAT)*

Following the technique described by the Instituto Interamericano de Cooperacion para la Agricultura (IICA, 1987), IFAT was performed using slides with crude antigen of *A. marginale* and *B. bovis*, produced at the Laboratory of Parasitic Diseases, UFPA. In the IFAT, a 1:64 dilution of conjugated bovine anti-IgG (SIGMA, Saint Louis, MO, USA) was used. Positive and negative controls of each parasite were also placed on each slide. The *cut off* for a positive plasma sample was one that presented fluorescent antibodies at the dilution 1:320 (SANTOS et al., 2002).

#### *Physical score and body condition score (PBCS)*

Biweekly physical exams of calves were performed in the morning, following previously described methods (DIRKSEN et al., 1993). The sequence of evaluations was as follows: behavior, position, posture, and physical condition; respiratory activity and presence of nasal secretion; color of mucosa; skin turgor; rectal temperature (RT, in °C); navel evaluation; and weight (kg). The PBCS scores were on a scale of one to five, following Hoffman (1996). Monthly indices were calculated as the average of the two measurements within a month, for a total of three monthly scores during the calf period (zero to 90 days of age) and nine during the growth period (91 to 365 days of age).

#### *Economic analysis*

In the economic analysis, all effective operational costs (EOC) were computed and divided into the following groups: labor (farm worker and veterinarian), exams, applied materials, and drugs (LOPES et al., 2014a). In each treatment, labor included the time spent holding the animal, collecting fecal samples, and applying drugs. The duration of each activity (in minutes) was timed by the researchers. Labor value was estimated using treatment duration and a minimum daily salary (veterinarian), as well as a minimum monthly salary (farm worker). These values were transformed into R\$/minute and multiplied by the time spent in each activity (LOPES et al., 2013).

#### *Statistical Analyses*

The statistical analyses were performed in PASW18.0 (STATISTICAL PACKAGE FOR THE SOCIAL SCIENCES - SPSS, 2009). Descriptive analyses of all the variables in both groups were performed. Behavior was considered altered if the animal presented depression or apathy. Respiration was considered normal if the animal only presented thoracic or abdominal breathing. Physical condition was considered altered if pale mucosa, jaundice, or cyanosis was present; and non-normal if any dehydration occurred. Calves were divided in three age groups (0 to 90 days, 91 to 180 days, 181 to 365 days of age), to assess weaning- and moving-related stress (the latter referring to the change from individual housing of the calf period to the paddocks for the growth phase).

Variables were subjected to univariate analysis with the chi-square ( $\chi^2$ ) or the Fisher exact test (less than five observations in at least one cell of the contingency table). Under the  $\chi^2$  test, associated variables with  $p \leq 0.2$  were selected for the construction of multiple models. Logistic regression was used to estimate association strength (or risk) through adjusted *odds ratio* (OR), with a

95% confidence interval (LOPES et al., 2014b). Finally, quantitative analyses of the variables were performed with linear regressions and t tests.

## Results

### *Physical parameters and globular volume (GV)*

Table 1 shows that calves of the CT group exhibited statistically more pale mucosa and

incidences of low GV (<24%). However, the two groups did not differ in the frequency of hyperthermia or in any of the remaining variables. Infection of the intra-abdominal segment did not occur in the umbilical structures (omphalophlebitis, omphaloarteritis, and/or omphalourachitis) of either group, but the CT group had a significantly higher number of omphalitis cases and infections in the extra-abdominal segments of the umbilical structures (Table 1).

**Table 1.** Average frequencies (%) of physical alterations and the globular volume (GV) in Holstein Friesian calves subjected to strategic selective treatment (ST) or conventional treatment (CT).

Parameters	ST	CT	p Value	Odds ratio (OR)	Confidence Interval (IC 95%)
Thoracic or abdominal breathing	1.9	2.2	0.497	-	-
Nasal discharge	5.2	3.5	0.183	-	-
Dehydration	2.2	3.5	0.216	-	-
Hyperthermia**	14.6	15.6	0.160	-	-
Pale Mucosa	14.5	21.8	0.009*	1.644	1.108 - 2.441
GV (<24%)	12.9	24.4	0.000*	2.180	1.462 - 3.251

\*Significant with  $p < 0.05$ , using a chi-squared test; \*\*Rectal temperature  $>39.5^{\circ}\text{C}$ .

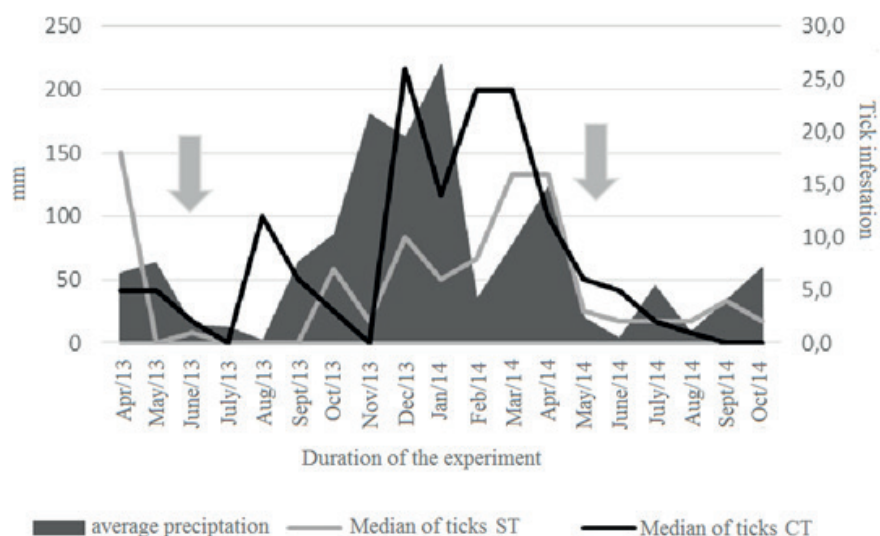
No calves died in the ST group, but mortality rate under CT was 20%, with three deaths counted from March to April 2014. The cause of death in all three cases was septicemia resulting from anaplasmosis, symptomatic carbuncle, and internal omphalitis, with the formation of umbilical abscess.

### *Ectoparasites*

Neither group presented cases of umbilical myiasis. However, average botfly count differed significantly ( $p < 0.05$ , t-test) between the ST ( $0.5 \pm 1.7$ ) and CT ( $1.36 \pm 3.0$ ) groups, with the highest infestation levels occurring during the rainy season for both treatments. In the ST group, the

total number of 1% doramectin and 1% ivermectin treatments for umbilical myiasis and botfly control, respectively, was similar (15 treatments averaging 1 treatment per animal).

Figure 1 presents *R. microplus* count medians in relation to precipitation. Significantly more ticks ( $p < 0.05$ , t-test) were found in the CT ( $15.59 \pm 23.1$ ) group than in the ST ( $10.1 \pm 15.7$ ), with peaks in the rainy season in both treatments. Moreover, tick and botfly counts were both significantly predicted by bovine age, treatment type, and season (Table 2). Finally, in the ST group, the total number of cypermethrin *pour-on* treatments for *R. microplus* control was 98 (average  $6.5 \pm 2.5$  treatments/animal).

**Figure 1.** Medians of the *Rhipicephalus microplus* adult-female counts in Holstein Friesian calves subjected to strategic selective treatment (ST) or conventional treatment (CT). Arrows refer to ST values.**Table 2.** Linear regression of variables affecting *Rhipicephalus microplus* adult female and *Dermatobia hominis* larval counts in Holstein Friesian calves.

Ectoparasites	Variables	b*	p value	Confidence interval (IC 95%)
<i>R. microplus</i>	(Constant)	-13.494	0.000	-19.973 - -7.015
	Season (dry or rainy)	9.617	0.000	6.789 - 12.445
	Treatment (ST or CT)	5.326	0.000	2.499 - 8.154
	Age (months)	0.024	0.000	0.011 - 0.038
<i>D. hominis</i>	(Constant)	0.184	0.645	-0.599 - 0.967
	Season (dry or rainy)	0.606	0.001	0.264 - 0.947
	Treatment (ST or CT)	0.796	0.000	0.454 - 1.138
	Age (months)	-0.008	0.000	-0.009 - -0.006

\*Regression coefficient; ST = strategic selective treatment; CT = conventional treatment.

### Hemoparasites

The seropositivity rates for *A. marginale* were 85% and 78% in ST and CT groups, respectively, and 0.80% and 1.30% for *Babesia* spp., with no significant difference in the overall average of parasitemia among treatments. Rickettsial infection from *A. marginale* in the ST and CT groups averaged 0.09% and 0.08%, respectively. For *Babesia* spp., the parasitemia levels of ST and CT were 0.014% and 0.088%, respectively.

Results of the logistic regression revealed that *A. marginale* seropositivity is statistically affected by season, GV, and rectal temperature ( $p=0.01$ ), but not by treatment or calf age.

Analysis of CS for bovine anaplasmosis indicated that subclinical infection predominated during the calf phase (0 to 90 days of age), but symptomatic cases also occurred in both treatments (Table 3).

**Table 3.** Average frequency of clinical scores (EC) for bovine anaplasmosis in Holstein Friesian calves subjected to strategic selective treatment (ST) or conventional treatment (CT), grouped by age.

Treatments	Age groups (days)	Clinical scores (%)			
		1	2	3	4
ST	0 - 90	54.5	27.3	18.2	-
	91 - 180	26.0	38.0	30.0	6.0
	181 - 365	23.1	50.8	21.5	4.6
CT	0 - 90	65.8	21.1	13.2	-
	91 - 180	31.0	31.0	28.5	9.5
	181 - 365	12.5	42.9	37.5	7.1

CS 1 = subclinical infection; CS 2 = clinical infection (IC) mild; CS 3 = IC moderate; CS 4 = IC severe; CS 5 = IC very severe.

In the IFAT of *A. marginale* and *B. bovis*, the overall seropositivity-rate average was 85.6% and 87.8% in the ST group, respectively, and 83.2% and 83.2% in the CT group. Table 4 depicts the seropositivity for *A. marginale* grouped by age. Treatment type and age groups significantly interacted to affect *A. marginale* positivity index, lowering values in 0-90-day-old calves from the ST group. This group had been subjected to a total of 89 enrofloxacin treatments for *A. marginale* control (average  $5.9 \pm 2.2$  treatments/animal).

**Table 4.** Average (%) and standard deviation of seropositivity for *Anaplasma marginale* in Holstein Friesian calves subjected to strategic selective treatment (ST) or conventional treatment (CT), grouped by age.

Treatments	Age groups (days)		
	0 - 90	91 - 180	181 - 365
ST	71 (0.32) <sup>bA</sup>	92 (0.53) <sup>aA</sup>	90 (0.41) <sup>aA</sup>
CT	83 (0.40) <sup>aA</sup>	75 (0.36) <sup>aB</sup>	88 (0.40) <sup>aA</sup>

Values followed by different lowercase letters in the rows, and uppercase in the columns are statistically different ( $p < 0.05$ ).

#### Body condition score (PBCS) and treatment costs

The daily average weight gains (g) was the same ( $p = 0.768$ , t-test) between the ST ( $580 \text{ g} \pm 0.080$ ) and CT ( $570 \text{ g} \pm 0.070$ ) groups. The average weight (kg) of 12-month-old calves was  $246.7 \text{ kg} \pm 27.5$  in the ST group and  $237.3 \text{ kg} \pm 26.5$  in the CT group ( $p > 0.05$ ). Most calves had PBCS scores between 2.5 and 3.0. Compared with the ST group, the CT group had a higher average number of slim calves (PBCS between 2.0 and 2.5) at 90 days of age (Table 5). The EOC of the ST group (R\$ 406.58) was 3.7 times higher than that of the CT group (R\$ 110.90) (Table 6).

**Table 5.** Frequency (%) of body condition score (BCS) in Holstein Friesian calves subjected to strategic selective treatment (ST) or conventional treatment (CT), grouped by age.

BCS	Treatments	Age groups (days)		
		0 - 90	91 - 180	181 - 365
2 to 2.5	ST	8.5	10.4	2.3
	CT	7.0	19.4	7.7
> 2.5 ≤ 3	ST	85.1	83.4	85.0
	CT	81.4	75.9	83.2
> 3 ≤ 4	ST	6.4	6.2	12.7
	CT	11.7	4.6	9.1



**Table 6.** Effective operational cost (EOC) of the strategic selective treatment (ST) and the conventional treatment (CT) for the control of ectoparasites and hemoparasites in Holstein Friesian calves.

Item	ST		CT	
	R\$	% of EOC	R\$	% of EOC
Work force (farm worker)	11.83	2.90	6.07	5.47
Work force (veterinarian)	2.05	0.50	-	-
Laboratory tests	336.43	82.75	-	-
Materials used in drug application	47.08	11.58	49.55	44.68
Drugs	9.19	2.26	55.28	49.85
Total EOC	406.58	100.00	110.90	100.00

## Discussion

### *Physical parameters and globular volume*

The CT group experienced more physical and GV changes than the ST group, probably as a consequence of diseases such as pneumonia and diarrhea, in addition to anaplasmosis and bovine babesiosis. Specifically, the CT group experienced higher frequencies of pale mucosa and more anemia, two traits that are closely linked and indicate hemoparasitosis, as a result of *A. marginale* or *Babesia* spp. infection. In both anaplasmosis and bovine babesiosis, young animals exhibit weaker clinical signs than adults, which also present higher mortality rates (RADOSTITS, 2002). However, rectal temperature did not differ between treatments. Although this might appear to indicate that the treatments were equally effective, the lack of a significant difference might be attributed to an increase in temperature fluctuation from the CT group. Such a condition is a frequent clinical sign of anaplasmosis, mainly during the rise of rickettsial infection (RICHEY, 1993).

We found that no deaths occurred in the ST group. As lower mortality rates significantly reduce the amount of years required to stabilize bovine herds (LOPES et al., 2009), our proposed treatment plan should contribute to an increase in profitability for the farmer.

### *Ectoparasites*

Botfly infestations were the most severe during the rainy season in both treatments. This pattern accords with findings showing that these parasites are more common between March and April (end of rainy season), after an increase during the warmest months (MAGALHÃES; LESSKIU, 1982). Despite the higher botfly count, we were able to fully present umbilical myiasis with a 1% ivermectin treatment of ST calves at birth, and significantly reduced omphalitis cases compared with TC.

Tick counts also experienced a peak during the rainy season, which favors *R. microplus* reproduction and thus, their population increase (MARTINS et al., 2002). In the southeast region, the average temperature and humidity conditions allow the development and survival of ticks during all months, resulting in four generations per year (DOMINGUES et al., 2008). Likely because of these seasonal patterns, we found that ST reduced tick count as compared to that with CT, but only during the dry season. Thus, the dry period is ideal for strategic control, which aims to eliminate more ticks from one generation, consequently reducing individuals of the next generation (FURLONG; PRATA, 2006). In addition, our selective treatment strategy allows the preservation of a parasite population in *refugia*; these pests are not exposed to drugs and remain susceptible. As a result,

implementation of ST should delay the emergence of ticks resistant to the used drugs (MARTINS et al., 2005; VAN WYK, 2001).

### *Hemoparasites*

The two treatments did not result in significant differences in blood-smear positivity for *A. marginale*. However, calves between 180 and 365 days of age tended to exhibit lower positivity. This is in contrast to findings from Gonçalves et al. (2011), indicating that calves between 60 and 180 days presented more tick-borne diseases, likely caused by a decrease in passive immunity and an insufficient establishment of active immunity.

Both treatments experienced an increase in rickettsial infection and lowered GV during January to April. This period corresponds to an increase in *R. microplus* count. Intense infestations by Arthropoda vectors and consequent inoculation of hemoparasites may lead to severe manifestations of disease across cattle of all age groups, causing high rates of morbidity and mortality (SACCO et al., 2001). Here, we demonstrated that enrofloxacin was effective in controlling acute anaplasmosis among calves of the ST group, even during the rainy season. Our preference for this drug is supported by previous data indicating its greater efficiency (compared with oxytetracycline) in treating acute anaplasmosis among calves, leading to an improved control of rickettsia and fast clinical recovery of animals (FACURY-FILHO et al., 2012).

Cattle between 0 to 90 days were predominantly asymptomatic for anaplasmosis across both treatments. These results are in accord with data showing that colostrum antibodies protect animals in endemic areas until approximately three months after birth (RADOSTITS, 2002). Subsequently, antibody levels rise again from five to six months, due to active infection (ANDRADE et al., 2001). Additionally, our results are also in line with In a study showing the predominance

of subclinical anaplasmosis in a group of young dairy calves from Campos das Vertentes, MG, which subsequently began to exhibit clinical anaplasmosis (CARVALHO et al., 2012). The occurrence of clinical cases in both our study and previous work is probably associated with the “immunological frame,” which results from the loss of passive immunity (colostrum antibodies) in conjunction with an insufficient development of active immunity (natural infection), on top of immunosuppressive factors such as stress and concomitant infections (CARVALHO et al., 2012).

We found that seropositivity rates for *A. marginale* and *B. bovis* ranged around 80% in both groups. Given that areas with seropositivity rates below 10% and above 80% are considered enzootically stable (MARTINS et al., 2005), our data suggest that the FE-UFLA and the surrounding land is such a region. Previous work also indicates that FE-UFLA is located in a region considered enzootically stable for anaplasmosis and bovine babesiosis (CARVALHO et al., 2012).

In the ST group, calves between 0 and 90 days of age had lower seropositivity indices for *A. marginale* than other age groups. This result may be associated with the fact that farm workers face lower challenges with the parasitic vectors when the calves are individually housed than when they are living in the Tifton paddocks during the growth phase. Moreover, preventive treatment in seasons with greater proliferation of ticks and flies generated lower antibody rates in the 0-to-90-day calves (ANDRADE et al., 2001). Alternatively, the lower frequency of positive calves in this age group may be a result of higher passive immunity (colostrum antibodies). Among CT calves, the situation was reversed, with animals from 91 to 180 days of age presenting a lower seropositivity index (75%) compared with other age groups. This pattern may be partially caused by a decrease in passive immunity, which occurs around three months of age (GONÇALVES et al., 2011).

*Body condition score (PBCS) and treatment costs*

Average daily weight gain did not differ significantly between ST and CT groups, remaining below the expected values for well-fed Holstein Friesian calves (800 g day<sup>-1</sup>) (CAMPOS; LIZIEIRE, 2005). In both treatments, the predominant PBCS was between 2.5 and 3. However, the ST group presented a lower frequency of slim calves after 90 days of age. Compensatory growth results in an increase of calf PBCS starting from three months of age (HOFFMAN, 1996). However, the PBCS of heifers should not be >3, and a variation between 2.5 and 3 is acceptable, from the 10 to the 17 months of age, so that excessive fat deposition may be avoided (SANTOS et al., 2002).

The overall average cost per bovine was higher in ST (R\$ 406.58) than in CT (R\$ 110.90). The difference between the two treatments was primarily due to a higher drug cost (ST: 49.85% of the EOC versus CT: 2.26% of the COE). In addition, ST also had significant costs associated with laboratory exams (82.75% of the EOC), required to monitor the animals and determine those requiring treatment. Both of these expenses are the result of a strategic plan that aims to only give drugs depending on animal condition. We agree with Lopes and Lopes (1999) that a clearer understanding of the exact expenditure allows costs to be easily divided into groups, enabling more organized financial monitoring and thus helping technicians or farmers in the treatment decision-making process.

Although there is no strategic program for parasite control implemented at the FE-UFLA, it should be emphasized that the farm contains high-quality sanitary and zootechnic management, as well as constant veterinary support. The calves in the CT group were monitored almost daily by trained staff, who are supervised by professors from the Departments of Zootechny and Veterinary Medicine at the UFLA. The frequent evaluation

of these animals enabled the early diagnosis of diseases and promptness of treatments. We suggest that if this experiment had been performed in a farm with substandard management practices, such as many of the facilities in dairy systems from the Lavras region, the results obtained with ST would differ even more dramatically from those of CT.

**Conclusion**

Although ST was more expensive, the treatment plan was also more efficient in reducing parasite burden when compared with CT. Calves in the ST group exhibited fewer counts of *R. microplus* females and *D. hominis* larvae, and possessed weaker hemolytic anemia conditions compared with CT calves. However, ST did not affect the dynamic of *A. marginale* and *B. bovis*-related infections, indicating that the FE/UFLA may be characterized as enzootically stable for anaplasmosis and bovine babesiosis.

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**References**

- ANDRADE, G. M.; VIDOTTO, O.; VIDOTTO, M. C.; YOSHIHARA, E.; KANO, F. S.; AMARAL, C. H. S. Seroprevalence of *Anaplasma marginale* in dairy cattle and, studies on the dynamics of natural infection of Holstein calves in Southern Brazil. *Semina: Ciências Agrárias*, Londrina, v. 22, n. 2, p. 155-159, 2001.
- BIANCHIN, I.; CATTO, B. J.; KICHEL, N. A.; TORRES, A. A.; HONER, M. R. The effect of the control of endo- and ectoparasites on weight gains in crossbred cattle (*Bos Taurus taurus* × *Bos Taurus indicus*) in the central region of Brazil. *Tropical Animal Health and Production*, Edinburgh, v. 39, n. 4, p. 287-296, 2007.

- BORJA, G. E. M. Erradicação ou manejo integrado das miíases neotropicais das Américas? *Pesquisa Veterinária Brasileira*, Rio de Janeiro, v. 23, n. 32, p. 131-138, 2003.
- BRASIL. Ministério da Agricultura e Reforma Agrária. Secretaria Nacional de Irrigação. *Normais climatológicas: 1961-1990*. Brasília: Departamento Nacional de Meteorologia, 1992. 84 p.
- CAMPOS, O. F.; LIZIEIRE, R. S. *Criação de bezerras em rebanhos leiteiros*. Juiz de Fora: Embrapa Gado de Leite, 2005. 142 p.
- CARVALHO, A. H. O.; SILVA JUNIOR, F. A.; DAHER, D. O.; ROCHA, C. M. B. M.; GUIMARÃES, A. M. Efeito do sistema de produção de leite sobre a estabilidade enzoótica para *Anaplasma marginale* e *Babesia bovis* em bezerras na região do Campo das Vertentes de Minas Gerais, Brasil. *Semina: Ciências Agrárias*, Londrina, v. 33, n. 1, p. 323-332, 2012.
- CHARLIER, J.; HÖGLUND, J.; SAMSON-HIMMELSTJERNA, G.; DORNY, P.; VERCRUYSSSE, J. Gastrointestinal nematode infections in adult dairy cattle: impact on production, diagnosis and control. *Veterinary Parasitology*, Amsterdam, v. 164, n. 16, p. 70-79, sept. 2009.
- DANTAS, A. A. A.; CARVALHO, L. G. de; FERREIRA, E. Classificação e tendência climática em Lavras, MG. *Ciência e Agrotecnologia*, Lavras, v. 31, n. 6, p. 1862-1866, nov./dez. 2007.
- DIRKSEN, G.; GRÜNDER, H. D.; STÖBER, M. *Rosemberg: exame clínico dos bovinos*. 3. ed. Rio de Janeiro: Guanabara Koogan S.A., 1993. 419 p.
- DOMINGUES, L. N.; CUNHA, A. P.; BELLO, A. C. P. P.; BASTIANETTO, E.; LEITE, R. C. Epidemiologia das principais parasitoses dos animais de produção: parte II, controle estratégico de parasitos. *Revista Veterinária e Zootecnia em Minas*, Belo Horizonte, v. 28, n. 97, p. 27-37, 2008.
- FACURY-FILHO, E. J.; CARVALHO, A. U. de; FERREIRA, P. M.; MOURA, M. F.; APOLINÁRIO, B. C.; SANTOS, L. P. H.; RIBEIRO, M. F. B. Effectiveness of enrofloxacin for the treatment of experimentally-induced bovine anaplasmosis. *Revista Brasileira de Parasitologia Veterinária*, Jaboticabal, v. 21, n. 1, p. 32-36, jan./mar. 2012.
- FURLONG, J.; PRATA, M. Controle estratégico do carrapato dos bovinos de leite: instrução técnica para produtores de leite. 2. ed. Juiz de Fora: EMBRAPA Gado de Leite, 2006. 2 p.
- GONÇALVES, R. C.; SILVA, A. A. da; FERREIRA, D. O. L.; CHIACCHIO, S. B.; LOPES, R. S.; BORGES, A. S.; AMORIM, R. M. Tristeza parasitária em bovinos na região de Botucatu, SP: estudo retrospectivo de 1986-2007. *Semina: Ciências Agrárias*, Londrina, v. 32, n. 1, p. 307-312, jan./mar. 2011.
- GRISI, L.; LEITE, R. C.; MARTINS, J. R.; BARROS, A. T.; ANDREOTTI, R.; CANÇADO, P. H.; LEÓN, A. A.; PEREIRA, J. B.; VILLELA, H. S. Reassessment of the potential economic impact of cattle parasites in Brazil. *Brazilian Journal of Veterinary Parasitology*, Jaboticabal, v. 23, n. 2, p. 150-156, abr./jun. 2014.
- GRISI, L.; MASSARD, C. L.; MOYA BORJA, G. E.; PEREIRA, J. B. Impacto econômico das principais ectoparasitoses em bovinos no Brasil. *Hora Veterinária*, Porto Alegre, v. 21, n. 125, p. 8-10, 2002.
- GUGLIELMONE, A. A. Epidemiology of babesiosis and anaplasmosis in South and Central America. *Veterinary Parasitology*, Amsterdam, v. 57, n. 1-3, p. 109-119, 1995.
- HOFFMAN, P. C. Optimum body size of holstein replacement heifers. *Journal of Animal Science (In Press)*, Philadelphia, v. 75, p. 836-845, 1996.
- HONER, M. R.; GOMES, A. O manejo integrado de mosca dos chifres, berne e carrapato em gado de corte. Campo Grande: EMBRAPA-CNPGC, 1992. 60 p. (Circular técnica, 22).
- INSTITUTO INTERAMERICANO DE COOPERATION PARA LA AGRICULTURA - IICA. Técnicas para el diagnóstico de babesiosis y anaplasmosis. San José: IICA, 1987.
- KOCAN, K. M.; DE LA FUENTE, J.; BLOUIN, E. F.; GARCIA-GARCIA, J. C. *Anaplasma marginale* (Rickettsiales: Anaplasmataceae): recent advances in defining host-pathogen adaptations of a tick-borne rickettsia. *Parasitology*, Cambridge University Press, v. 129, p. 285-300, 2004. Supplement S1.
- KUTTLER, K. L. World-wide impact of babesiosis. In: RISTIC, M. *Babesiosis of domestic animals and man*. Florida: CRC Press, 1988. p. 1-22.
- LOPES, M. A.; DIAS, A. S.; CARVALHO, F. M.; LIMA, A. L. R.; CARDOSO, M. G.; CARMO, E. A. Resultados econômicos de sistemas de produção de leite com diferentes níveis tecnológicos na região de Lavras, MG, nos anos 2004 e 2005. *Ciência e Agrotecnologia*, Lavras, v. 33, n. 1, p. 252-260, jan./fev. 2009.
- LOPES, M. A.; LOPES, D. de C. F. Desenvolvimento de um sistema computacional para cálculo do custo de produção do leite. *Revista Brasileira de Agroinformática*, São Paulo, Brasil, v. 2, n. 1, p. 1-12, 1999.



- LOPES, M. A.; NOGUEIRA, T. M.; BARBOSA, G. L. Economic viability of the third milking in systems of production using closed-circuit mechanical milking. *Revista Ceres*, Viçosa, MG, v. 61, n. 4, p. 544-551, jul./ago. 2014a.
- LOPES, M. A.; PACHECO, E. O.; BRUHN, F. R. P.; VICENTE, F. H.; FARIA, P. B.; ROCHA, C. M. B. M. Fatores associados à percepção e atitudes de consumidores de carne bovina com certificação de origem. *Revista Brasileira de Ciência Veterinária*, Niterói, v. 21, n. 2, p. 131-136, 2014b.
- LOPES, M. A.; SILVA, M. das D.; DEMEU, A. A.; GOMIDE, D. R.; BRUHN, F. R. P. Custo da implantação e utilização de dois métodos de identificação de bovinos leiteiros. *Revista Ceres*, Viçosa, MG, v. 60, n. 6, p. 757-764, 2013.
- MAGALHÃES, F. E. P.; LESSKIU, C. Efeito do controle do berne sobre o ganho de peso e qualidade dos couros em novilhos de corte. *Pesquisa Agropecuária Brasileira*, Brasília, v. 17, n. 2, p. 329-326, fev. 1982.
- MARTINS, J. R. S.; FURLONG, J.; PRATA, M. C. A. Carrapatos: problemas e soluções. Juiz de Fora: EMBRAPA Gado de Leite, 2005. 65 p.
- MARTINS, J. R.; EVANS, D. E.; CERESÉR, V. H.; CORRÊA, B. L. Partial strategic tick control within a herd of European breed cattle in the state of Rio Grande do Sul, southern Brazil. *Experimental and Applied Acarology*, Amsterdam, v. 27, n. 3, p. 241-251, 2002.
- RADOSTITS, O. M. Doenças causadas por protozoários. In: RADOSTITS, O. M.; GAY, C. C.; BLOOD, D. C.; HINCHCLIFF, K. W. (Ed.). *Clínica veterinária: um tratado de doenças de bovinos, ovinos, suínos, caprinos e equinos*. 9. ed. Rio de Janeiro: Guanabara Koogan, 2002. p. 1156-1202.
- RICHEY, E. J. *Bovine Anaplasmosis*. Howard current veterinary therapy: food animal practice. 3<sup>th</sup> ed. Philadelphia: W. B. Saunders, 1993. 1002 p.
- SACCO, A. M. S.; KESSLER, R. H.; MADRUGA, C. R. Cepas atenuadas de *Babesia bovis* e *Babesia bigemina* e de *Anaplasma centrale* como imunógenos no controle da tristeza parasitária bovina. *Ciência Rural*, Santa Maria, v. 31, n. 5, p. 849-855, set./out. 2001.
- SANTOS, G. T.; DEMASCENO, J. C.; MASSUDA, E. M.; CAVALIERI, F. L. B. Importância do manejo e considerações econômicas na criação de bezerras e novilhas. In: SUL-LEITE: SIMPÓSIO SOBRE SUSTENTABILIDADE DA PECUÁRIA LEITEIRA NA REGIÃO SUL DO BRASIL, 2. 2002, Maringá. *Anais...* Maringá: UEM/CCA/DZO - NUPEL, 2002. p. 239-267.
- SCHETTERS, T. H. P. M.; ARTS, G.; NIESSEN, R.; SCHAAP, D. Development of a new score to estimate clinical east coast fever in experimentally infected cattle. *Veterinary Parasitology*, Amsterdam, v. 167, n. 2-4, p. 255-259, 2009.
- STATISTICAL PACKAGE FOR THE SOCIAL SCIENCES - SPSS Statistics for Windows. Software. Version 18.0. Chicago: SPSS Inc., 2009.
- VAN WYK, J. A. Refugia - overlooked as perhaps the most potent factor concerning the development of anthelmintic resistance. *Onderstepoort Journal of Veterinary Research*, Pretoria, v. 68, n. 1, p. 55-67, 2001.
- VERCRUYSSSE, J.; CLAEREBOUT, E. Treatment vs non-treatment of helminth infections in cattle: defining the threshold. *Veterinary Parasitology*, Amsterdam, v. 98, n. 12, p. 195-214, July 2001.
- WHARTON, R. H.; UTECH, K. B. W. The relation between engorgement and dropping of *Boophilus microplus* (Canestrini) (Ixodidae) to the assessment of the tick numbers on cattle. *Journal of the Australian Entomological Society*, Melbourne, v. 9, n. 3, p. 171-182, 1970.



