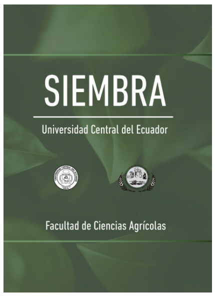
Level of tick infestation associated with individual factors of cattle in subtropical livestock farms in Ecuador



Grado de infestación de garrapatas asociado con factores individuales del ganado bovino en ganaderías subtropicales del Ecuador

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Abstract

In Ecuador, *Rhipicephalus microplus* is the primary tick species affecting cattle, and its control requires an understanding of its behavior. The objective of this study was to assess the degree of infestation by R. *microplus*, examining the anatomical location of ticks and their potential association with intrinsic factors of animals, such as overall health, body condition, and coat color. Four samplings were conducted every three months on a total of 15 farms in the Northwest of Pichincha (location1) and 15 farms in the Valle de los Quijos (location2). Ticks larger than 0.5 cm were counted on 5 randomly selected animals. The results from four samplings in both locations revealed an average of 99.39 ± SD156.28 (0–1426) ticks per animal, with averages of 108.8 ± SD179.43 (0–1426) and 88.2 ± SD122.80 (0–824) for locations 1 and 2, respectively. Only the variable "apparent skin and coat health" showed a statistically significant positive association, with a P-value <0.05 and odds ratios of 5.13 (95% CI: 2.04-13.65) and 25.83 (95% CI: 7.63-109.39) in the "fair" and "poor" categories compared to the considered good condition. Additionally, the category "old" in the Age variable had a P-value of 0.05 and an odds ratio of 2.88 (95% CI: 1.04-8.80). Anatomically, more ticks were found in the anatomical areas corresponding to thighs, rear udder, legs, and hock, with an average of 38.52±SD70.17 (0–448) ticks. In conclusion, tick infestation poses a serious problem affecting cattle in tropical regions of the country. This study contributes to better directing methods and control practices for cattle.

Keywords: parasitism, vectors, cattle, control

Resumen

En Ecuador, *Rhipicephalus microplus* es la principal garrapata que afecta al ganado bovino, cuyo control exige conocer su comportamiento. El objetivo de este estudio fue evaluar el grado de infestación de *R. microplus*, examinando la ubicación anatómica de las garrapatas y su posible asociación con factores intrínsecos de los animales como son el estado general de salud, condición corporal, color del pelaje. Se llevaron a cabo cuatro muestreos cada tres meses en un total de 15 fincas en el Noroccidente de Pichincha (localidad1) y 15 fincas en el Valle de los Quijos (localidad2).

Las garrapatas mayores a 0,5 cm fueron contadas en 5 animales escogidos al azar. Como resultados, en cuatro muestreos realizados en las dos localidades, se encontró un promedio de 99,39 ± SD156,28 (0–1.426) garrapatas por animal, y un promedio de 108,8±SD179,43 (0–1.426) y 88,2 ± SD122,80 (0–824) para la localidad 1 y 2, respectivamente. Solo la variable “salud aparente de piel y pelaje” mostró una asociación estadística positiva, con un valor $p < 0,05$ y odds ratios de 5,13 (IC 95 %: 2,04-13,65) y 25,83(IC 95%: 7,63-109,39) en las categorías “regular” y “malo” respecto del estado considerado como bueno. Además, la categoría animal denominado “viejo” en la variable Edad tuvo un valor $p = 0.05$ y un odds ratio de 2,88 (IC 95 %: 1,04-8,80). Anatómicamente se encontraron más garrapatas en la zona anatómica correspondiente a muslos, ubre posterior, pierna y corvejón, con un promedio de 38,52±SD70, 17 (0–448) garrapatas. En conclusión, la infestación con garrapatas representa un problema grave que afecta a las ganaderías de regiones tropicales del país, este estudio ayuda a direccionar de mejor manera las formas y prácticas de control sobre los bovinos.

Palabras Claves: parasitismo, vectores, ganado, control

1. Introduction

Ticks that affect tropical and subtropical livestock worldwide are hematophagous ectoparasites (Nava et al., 2022) that cause significant impacts on animal health (Guglielmone et al., 2021). In Ecuador, several studies measure the problem of ticks in livestock (Bustillos & Rodríguez, 2016; Orozco Álvarez, 2018; Paucar et al., 2022; Pérez Otáñez, 2016; Rodríguez-Hidalgo et al., 2017). The main tick species infesting cattle is *Rhipicephalus microplus* (Bustillos & Rodríguez, 2016; Maya-Delgado et al., 2020; Tinoco et al., 2023), although the species *Amblyoma* spp. with a localized distribution has also been described (Orozco Álvarez, 2018; Paucar et al., 2022).

Rhipicephalus microplus causes direct damage to animals by feeding on their blood, causing anemia, reduction in milk and meat production, transmission of diseases (babesiosis and anaplasmosis) and, in severe cases, the death of cattle. The main method for ticks control has been the use of acaricides; however, their prolonged, anti-technical and indiscriminate use has caused significant levels of resistance in field ticks (Pérez-Otáñez et al., 2023; Rodríguez-Hidalgo et al., 2017) and represents considerable economic costs to the (livestock) farmer (Paucar-Quishpe et al., 2023).

The distribution and degree of infestation of *Rhipicephalus microplus* in cattle is influenced by the complex interaction between biotic and environmental factors. In biotic terms, the presence and density of cattle, as well as their genetic resistance, play a fundamental role in the proliferation of ticks (Jonsson et al., 2014; Tabor et al., 2017). On the other hand, environmental temperature and humidity are crucial for the development of their life cycle (Castañeda Arriola et al., 2021). The climatic conditions of tropical regions, as well as the availability of suitable habitats, mainly grasslands, provide a favorable environment for the development and survival of *R. microplus* (Bustillos & Rodriguez, 2016). According to research conducted in other countries factors such as the coat color, breed and age of animals are related to variable levels of tick infestation in cattle (da Silva et al., 2013; da Silva et al., 2014; Ferraz da Costa et al., 2014). Also, certain areas of the animal’s body tend to be more or less infested (González-Cerón et al., 2009a).

Therefore, the objective of this research is to determine quantitatively and qualitatively the degree of infestation by *R. microplus* in cattle in two subtropical regions of Ecuador, and the possible relationship with individual factors of each cattle/animal, including age, sex, breed, coat color, lactating condition, apparent health, body condition, apparent condition of skin and coat, capillary filling and temperature. The results of this study allow us to understand the behavior of these ectoparasites and, based on this knowledge, to develop guided acaricide treatments, and specific control plans adapted to the reality of the country.

2. Materials and Methods

2.1. Study area

This study was part of the project “Socio-eco-epidemiology of ticks, tick-borne parasites, acaricide resistance and residual effects of acaricides in Ecuadorian tropical livestock: impacts on environmental, animal and public health” (TICKs&TBD), which was conducted in two livestock regions of the humid subtropics of northwestern Pichincha province, and the Quijos Valley in Napo province.

The northwestern region of Pichincha includes the cantons of San Miguel de los Bancos and Pedro Vicente Maldonado, as well as several rural parishes. It is located in the western foothills of the Andes, and is characterized by a tropical rainy climate, with an average annual temperature of 20.6 °C, and annual precipitation exceeding 3,000 mm. The rainy season goes from January to June, and the period of lower precipitation between July and December. The average monthly relative humidity is 88.50 % (GAD San Miguel de los Bancos, 2015). In the area known as Los Bancos, livestock breeding began in the 1990s, with Holstein and Brown Swiss cattle (Centro de la Industria Láctea [CIL], 2015). The activity became legally formalized in 2003 through the creation of the Asociación Agrícola Ganadera - *Livestock Farming Association* (Guzmán Saltos & Sánchez Rodríguez, 2015). The Quijos Valley is, instead, located in the Quijos Canton in the province of Napo, in the eastern foothills of the Andes Mountains, and is part of the Ecuadorian Amazon. It has a temperate-cold climate, and is divided into two distinct strips depending on altitude. The high fringe registers an average temperature of around 10.5 ˚C; while, the low fringe has an average temperature of 17 ˚C (Flor, 2015). Rainfall ranges from 1,000 to 1,500 mm in the high strips, and from 3,000 to 3,500 mm annually, reaching up to 4,500 mm in the lower and sheltered strips. The rainy season runs from March to August, and the low rainfall season extends from September to February (GAD Quijos, 2015). The regions under study, with their respective participating farms, are shown in Figure 1.

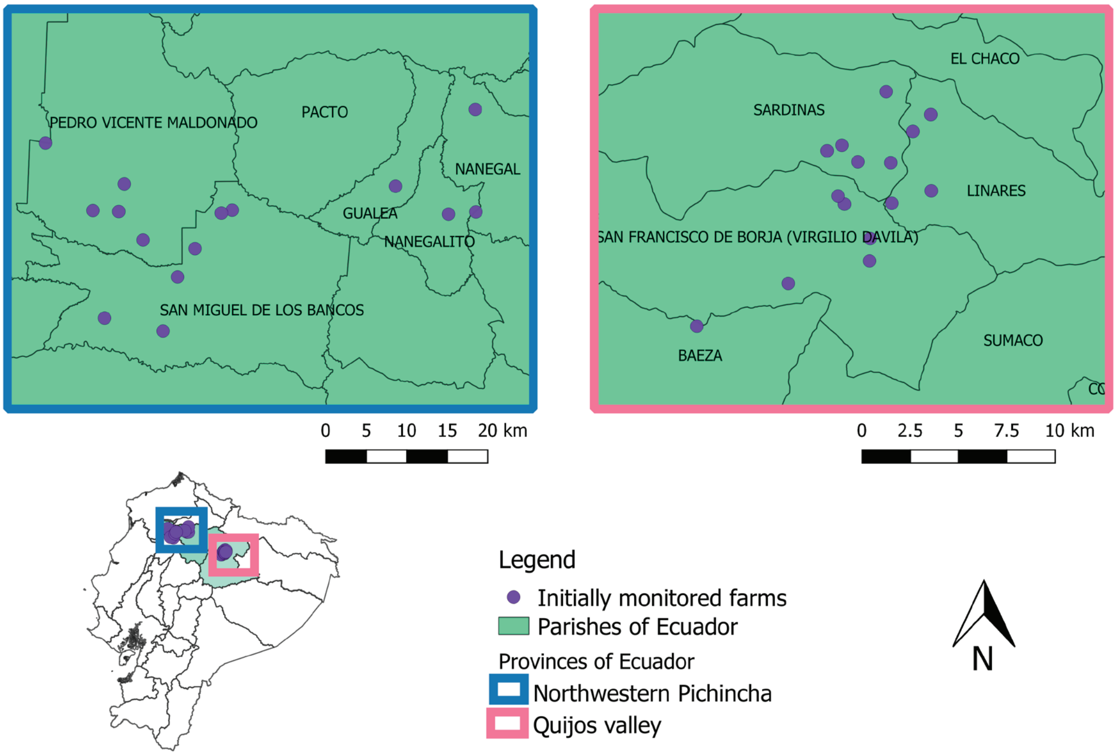

Figure 1. Spatial location of monitored farms in Northwest of Pichincha and Quijos Valley.

Within the universe of farms of the TICKs&TBD project, 15 farms per region were randomly selected; these farms were monitored and sampled on four occasions with an interval of three months from December 2020 to January 2022. However, due to the unavailability of cattle at the time of sampling, the number of farms varied throughout the study (Table 1).

2.2. Tick count

In each farm, five animals were randomly selected; they were visually inspected on their left side, and all ticks larger than 5 mm were counted. The result was duplicated according to the recommendations of Miraballes et al. (2022). After counting, 10 ticks were collected from each animal, and preserved in tubes with absolute alcohol for identification in the laboratory. This was done for each sampling. Prior to the visit, the producers

Table 1. Farms monitored by each of the samplings in the study regions.

Farms effectively participating by sampling				
Region	1er	2do	3er	4to
Northwestern Pichincha	14	12	11	11
Quijos valley	15	8	7	10

were contacted so that they would not carry out tick control 15 days prior, so that the control applied would not interfere with the counts.

2.3. Variables under study

For the first sampling only, breed, coat color, sex, lactation condition, age, nutritional status, capillary fill time, apparent health (good to poor), temperature and apparent skin and coat quality (good to poor) were recorded for each cattle. Each category used can be reviewed in Table 2.

Table 2. Variables used to determine their relationship with the number of ticks in each of the categories by descriptive statistics and as possible risk factors for low or high tick infestation.

Variables	Ítem	Description
Sex	Male	Animals with male reproductive organs
	Female	Animals with female reproductive organs
Age	Young	Less than 23 months
	Adult	Between 24 and 83 months
	Old	More than 84 months
Breed	*Bos taurus* X *Bos indicus*	Crossbreeds
	Bos Taurus	Breeds such as Holstein, Jersey, Brown Swiss.
	Bos indicus	Gyr, nelore.
Coat color	White	White animals
	Black	Black animals
	Brown	Brown and red animals
	Black and White	Black and white mixed color animals
Lactation status	Yes	Lactating animal
	No	Animal not lactating
Capillary filling time	Normal	Between 1 and 2 seconds
	Anormal	More than 3 seconds
Body condition	Thin	Grade 1 and 2
	Ideal	Grade 3
	Obese	Grade 4 and 5
Apparent health	Good	Animal standing, apparently healthy
	Regular	Animal is coughing, limping, or in any discomfort
	Bad	Animal is unable to stand up or has obvious trouble breathing
Temperature	Normal	Animals with rectal temperature between 36 and 39°C (36 and 39°C)
	High	Animals with rectal temperature greater than 39.1° C
Apparent coat and skin health	Good	Shiny coat, no scars, no deformities and no wounds
	Regular	Dull coat, with scars or deformities, but without wounds.
	Bad	Dull coat, with scars or deformities and wounds.

To obtain the dichotomous variable of tick infestation level, we followed the methodology of Paucar et al. (2022). The tick count values per animal were divided into three anatomical zones: a) anatomical zone 1, from the head to the tip of the thorax, b) anatomical zone 2, from the tip of the thorax to the sacrum, and c) anatomical zone 3, sacrum, perineum and hind legs (Figure 2). Each anatomical zone was considered infested when it had 20 or more ticks. The degrees of infestation were classified as 1) "null" if no zone was infested, 2) "low" if one anatomical zone was infested, 3) "medium" if two anatomical zones were infested, and 4) "high" if all three anatomical zones were infested. Finally, for statistical analyses of association, the degrees of infestation were transformed into a dichotomous variable, where the states "null" and "low" represented a LOW level (0) and "medium" and "high" represented a HIGH level (1). In total, data from 147 animals were collected for analysis.

To analyze the bovine anatomical zone with the highest number of ticks, we used the classification into three anatomical zones described above (Figure 2).

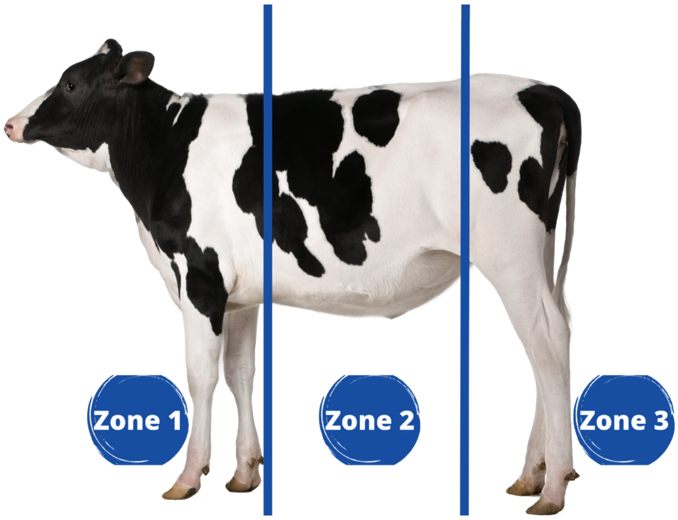

Figure 2. Anatomical zone division of the bovine body for tick counts and determination of the level of infestation. Zone 1" comprised the head to the tip of the thorax, "zone 2" covered from the tip of the thorax to the sacrum, and "zone 3" included the sacrum, perineum, posterior udder, between the legs, and hind legs.

2.4. Morphological identification of ticks

At the Applied Entomology Unit of the Zoonosis Research Institute of the Central University of Ecuador [CIZ], ticks were morphologically identified using a stereomicroscope (NIKON model SMZ745T, Tokyo, Japan) at magnification ×0.67-5 and taxonomic keys of Barros-Battesti et al. (2006) and Guerrero (1996).

2.5. Statistical analysis

The data collected were digitized in Excel spreadsheets for further analysis. Data related to quantitative infestation were organized by animal, locality and sampling for presentation with descriptive statistical analysis.

The data evaluating specific variables of the animals with the number of ticks were analyzed by using exclusively the information of the first sampling, in both localities. Through these data, the average, minimum, maximum and standard deviation of the number of ticks per animal were obtained.

In order to determine if there is a statistical relationship between the variables studied and the intensity of tick infestation, a univariate analysis of association was carried out using Fisher's exact test and a multiple logistic regression. In the univariate analyses, all the variables in Table 2 were evaluated individually, excluding sex and breed due to the lack of variability between categories in the results (at least 10 different cases per category). For multiple models, variables with a p value ≤ 0.20 in the univariate analysis were chosen. In the first step, the Variance Inflation Factor (VIF) was calculated and those with a VIF ≤ 8 were retained to avoid collinearity between variables. A stepwise variable elimination algorithm was implemented to select the simplest model. The StepAIC function of the MASS package in R facilitated model selection, using a threshold based on the Akaike Information Criterion (AIC). The model with the lowest AIC value was retained. The statistical significance level was set at 5 %. Model adequacy was assessed using Nagelkerke's R^2, Area Under the Receiver/Operator Curve (AUC), and sensitivity and specificity using the pROC package in R (Paucar et al., 2022).

3. Results

All (100 %) of the ticks identified on the farms under study correspond to the species *Rhiphicephalus microplus* (Figure 3).

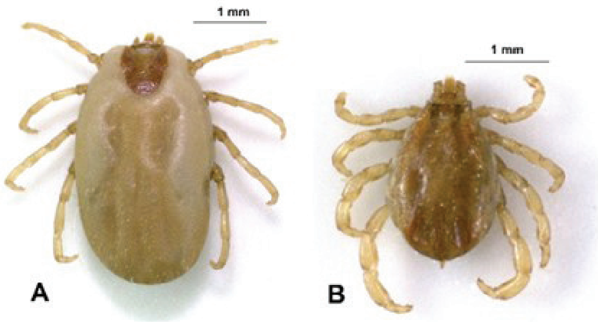

Figure 3. Dorsal view of the species *Riphicephalus microplus*: A) female, B) male.

3.1. Tick count

The average number of ticks per animal in the two study regions was 99.39 ± SD 156.28 (0 - 1,426), being 108.8 ± SD 179.43 (0 - 1,426) for Northwestern Pichincha and 88.2 ± SD 122.80 (0 - 824) for Valle de los Quijos. Table 3 shows the average number of ticks and the standard deviation per animal in each sampling, and by region.

Table 3. Mean and standard deviation of ticks on the animals, for each of the samplings in the study localities.

Total ticks per animal in overall average per sample				
Region	**1st sampling (Nov-Dec)**	**2nd sampling (Jun)**	**3rd sampling (Sept)**	**4th sampling (Dec-Jan)**
Northwestern Pichincha	150,4 ± SD 239,96 (0 – 1.426)	68,53± SD 124,11 (0 – 594)	81,82± SD 133,97 (0 – 594)	129,4± SD 168,93 (0 – 600)
Quijos valley	110,3 ± SD 148,25 (0 - 824)	99,95± SD 113,38 (0 – 406)	24,73± SD 31,89 (0 – 122)	87,59± SD 113,65 (0 – 670)
Both locations	129,6 ± SD 198,21 (0 – 1426)	81,1± SD 120,34 (0 – 594)	60,16± SD 110,80 (0 – 594)	108,5± SD 144,76 (0 – 670)

At the farm level, in all the samplings, higher average infestation values were found in the farms in the northwest of Pichincha compared to the Quijos Valley. In the northwest of Pichincha, the second sampling showed the lowest average number of ticks (medium infestation level), according to the categorization used in this study, while in Valle de los Quijos it corresponded to the third sampling (null infestation level). However, per farm, the presence of ticks was reported in all samples, with variations in the number of ticks in each one (Figure 4).

3.2. Bovine anatomical regions and tick counts

During the four samplings carried out in northwestern Pichincha, anatomical region 3 had the highest number of ticks, with an average of 44.79 ± SD 81.65 (0 - 448), followed by anatomical zone 1 with an average of 33.01 ± SD 69.36 (0 - 554), and anatomical zone 2 with an average of 31± SD 54.49 (0 - 472) ticks. In contrast, in Valle de los Quijos, anatomical zone 1 had the highest average number of ticks, with 33.3 ± SD 50.91 (0 - 304), followed by anatomical zone 3 with 31.08 ± SD 52.67 (0 - 438), and anatomical zone 2 with 23.73 ± SD 37.52 (0 - 210) ticks.

It is relevant to highlight that, in both study locations, anatomical zone 3 of the bovine body is the area with the highest number of ticks, given the higher number of individuals found (Figure 5).

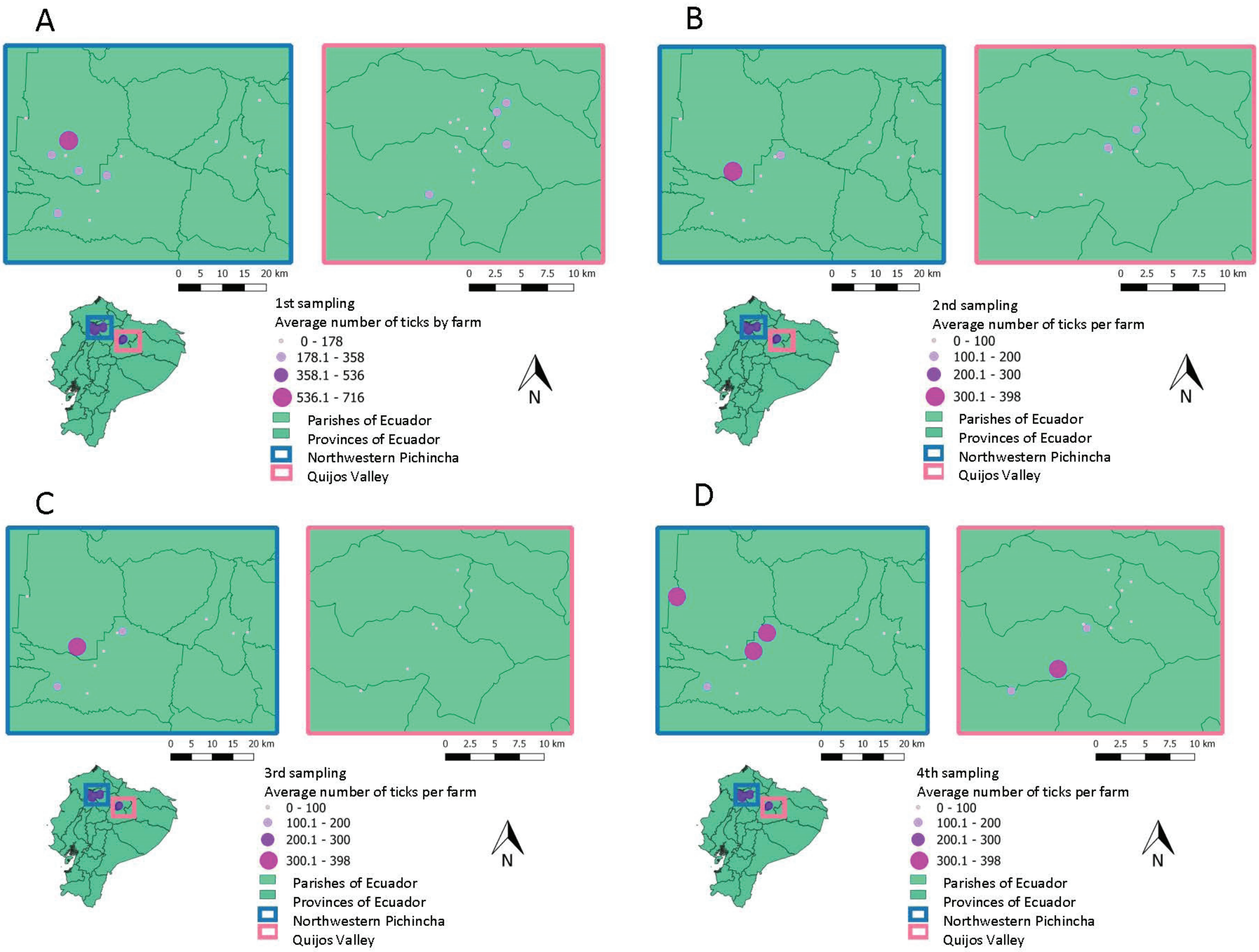

Figure 4. Maps of average number of ticks on cattle at the farm level, in each of the participating farms: A) first sampling, B) second sampling, C) third sampling, D) fourth sampling.

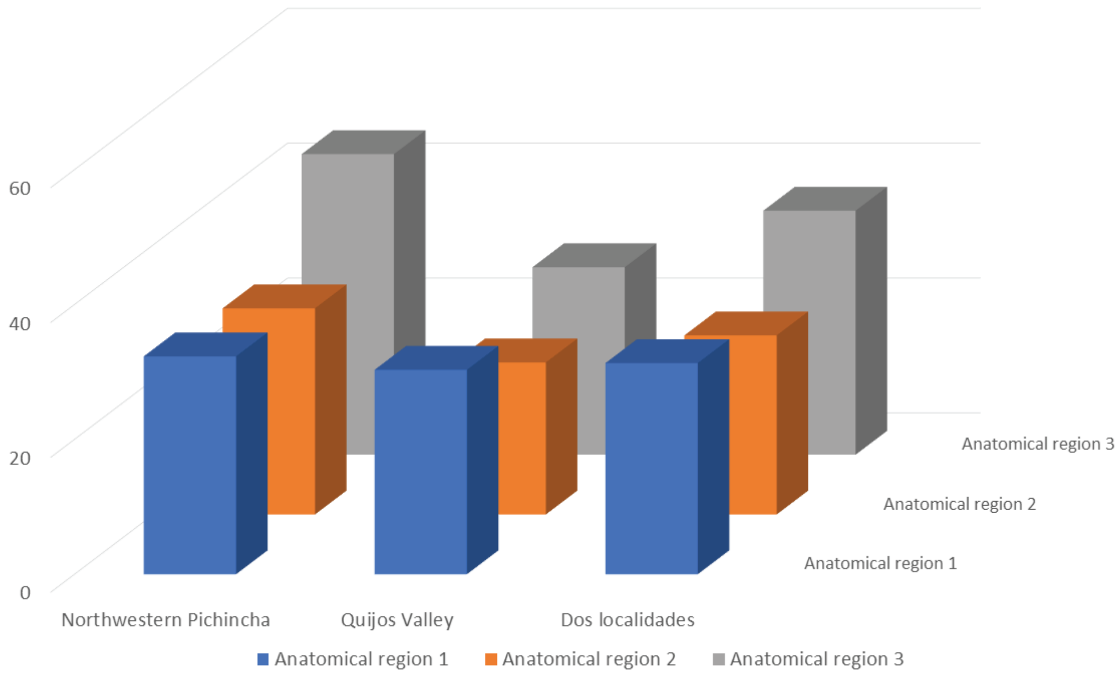

Figure 5. Average number of ticks for each of the anatomical zones of the animals in each study location.

3.3. Individual variables related to the number of ticks per animal

In the 'sex' variable, female cattle presented an average of 131.7 ticks, and males an average of 24.67 ticks. In relation to 'age', old animals presented 160.9 ticks on average, compared to young and adult animals, which presented 22.46 and 133.8 respectively. It can be highlighted that black animals, in lactation state, with low body condition, of regular apparent health, with abnormal capillary filling, and with poor skin and coat health, presented a higher number of ticks on average compared to the other categories within each variable (Table 4).

Table 4. Number and percentage of animals corresponding to each category, with their respective average number of ticks counted on the animal, standard deviation and minimum and maximum, by category.*

Variable	Ítem	# Animals		# Ticks		
		Number/ Total	%	Avg.	SD	min – max
Sex	Male	3/147	2	24,7	21.6	0 - 40
	Female	144/147	98	131,7	198.3	0 - 1.426
Age	Youth	13/147	9	22,5	25.1	0 - 66
	Adult	104/147	71	133,8	213.3	0 - 1.426
	Old	30/147	20	160,9	164.4	0 - 740
Breed	*Bos taurus* X *Bos indicus*	8/147	5	82,0	116.3	0 - 336
	Bos taurus	137/147	93	134,2	201.6	0 - 1.426
	Bos indicus	2/147	1	0,0	0.0	0 - 0
Coat color	White	4/147	3	149,5	181.6	0 - 410
	Black	28/147	19	191,3	229.8	0 - 1.042
	Brown	58/147	39	89,5	104.3	0 - 368
	Black and White	57/147	39	138,6	243.0	0 - 1.426
Lactation status	Yes	110/147	75	149,2	214.0	0 - 1.426
	No	37/147	25	71,0	117.4	0 - 480
Capillary filling time	Normal	126/147	86	124,3	196.7	0 - 1.426
	Abnormal	21/147	14	161	199.7	4 - 828
Body Condition	Thin	39/147	27	174,9	273.1	0 – 554
	Ideal	100/147	68	112,1	160.6	0 - 1.042
	Obese	8/147	5	125,8	151.3	16 - 470
Apparent health	Good	82/147	56	71,1	96.3	0 - 520
	Regular	59/147	40	205,9	266.3	0 - 1.426
	Bad	6/147	4	177,3	175.8	14 - 480
Temperature	Normal	120/147	82	131,3	198.0	0 - 1.426
	High	27/147	18	36,7	195.1	0 - 226
Apparent coat and skin health	Good	52/147	35	58,2	107.4	0 - 470
	Regular	59/147	40	93,7	98.4	2 - 520
	Bad	36/147	24	291,2	303.6	4 - 1.426

* *Num* = number of individuals within a given item; *Total* = total number of animals within the study; *Avg* = average number of ticks per item; *SD* = standard deviation.

Figure 6 presents boxplots of total tick counts per animal in relation to the individual variables in each of the categories used in the study. In support of the information given in Table 4.

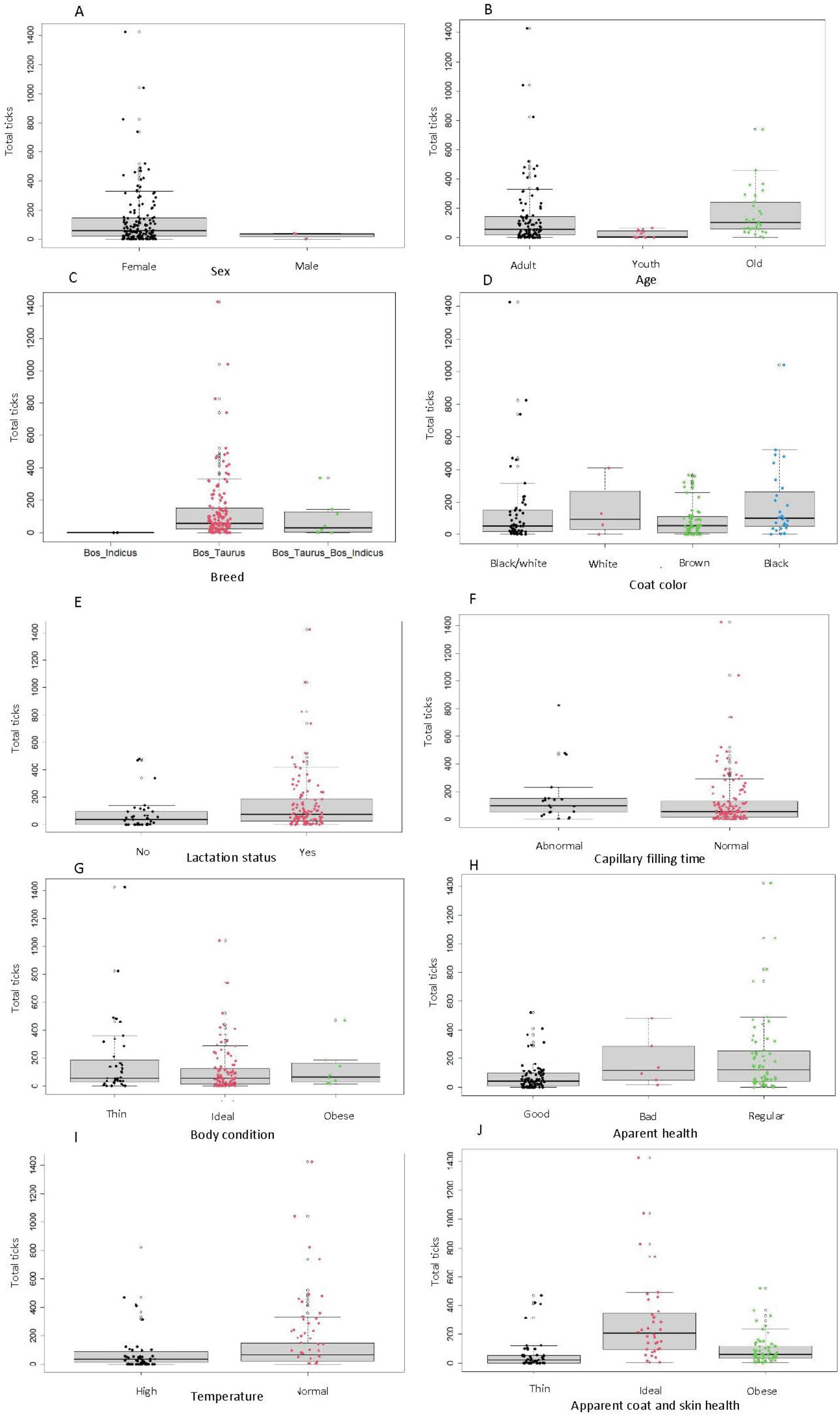

Figure 6. obtained with the values of the total tick count per animal, versus each of the individual variables and their categories, to observe those with the highest number of ticks. A sex, B age, C breed, D coat color, E lactation status, F capillary filling time, G body condition, H apparent health, I skin and hair quality, J temperature.

In the univariate statistical analysis, using the individual variables per animal versus dichotomous infestation degree, was emerged that the variables: 'age', 'apparent health' and 'apparent health of skin and coat' presented *p*-value < 0.05, the complete values can be reviewed in Table 5.

Table 5. Univariate analysis using explanatory variables in relation to the high level of infestation at the 'per animal' level, using Fisher's exact test.

Variable	Item	Total animals	Animals (+)	%	*P* value	OR	CI inf	CI sup
Age	Adult	104	54	51.92	< 0,001	Reference		
	Youth	13	2	15.38		0,18	0,02	0,73
	Old	30	23	76.67		2,98	1,22	8,17
Coat color	Black and White	57	27	47.37	0,05	Reference		
	White	4	3	75.00		3,02	0,33	90,47
	Brown	58	28	48.28		1,04	0,50	2,17
	Black	28	21	75.00		3,25	1,23	9,51
Lactation status	No	37	15	40.54		Reference		
	Yes	110	64	58.18	0,09	2,02	0,95	4,41
Capillary filling time	Anormal	21	15	71.43		Reference		
	Normal	126	64	50.79	0,10	0,42	0,14	1,12
Body condition+	Ideal	100	51	51.00	0,63	Reference		
	Thin	39	23	58.97		0,41	0,45	0,40
	Obese	8	5	62.50		0,56	0,72	0,53
Apparent health	Good	82	33	40.24	< 0,001	Reference		
	Regular	59	41	69.49		3,34	1,66	6,93
	Bad	6	5	83.33		6,58	0,95	179,86
Temperature	High	27	11	40.74		Reference		
	Normal	120	68	56.66	0,14	1,89	0,81	4,55
Apparent coat and skin health	Good	52	12	23.08	< 0,001	Reference		
	Regular	59	35	59.32		4,76	2,11	11,30
	Bad	36	32	88.89		24,68	7,91	98,95

* *NA* = No result was obtained for one of the items due to a low number of positive observations. *Animals (+)* = Animals with high levels of tick infestation; OR = odd ratio; inf CI = confidence interval at 95% confidence lower value of OR; sup CI = confidence interval at 95% confidence upper value of OR.

The final model in Table 6 included three variables, of which only the apparent health of the coat and skin was positively associated with the high level of tick infestation, with a *p*-value < 0.001 in the categories 'Regular' and 'Bad', with Odd Ratios of 5.13 and 25.83, respectively. This model obtained the lowest Akaike Information Criterion (AIC) value, corresponding to 162.9; its sensitivity corresponds to 0.75, and its specificity to 0.80. The AUC value is 0.84 (0.77-0.90). In addition, the R2 Nagelkerke value is 0.42, i.e.que, the model had a good fit.

Table 6 Results of multivariate logistic regression analysis using explanatory variables in relation to the high level of infestation at the per animal level, using general linear models.

Variable	Item	*P* value	OR	CI inf	CI sup
Apparent coat and skin health	Good		Reference		
	Regular	< 0,001	5,13	2,07	13,65
	Bad	< 0,001	25,83	7,63	109,39
Coat color	Black and White		Reference		
	White	0,07	15,13	1,06	480,41
	Brown	0,18	1,91	0,76	5,07
	Black	0,11	2,52	0,82	8,32
Age	Adult		Reference		
	Youth	0,11	0,25	0,03	1,17
	Old	0,05	2,88	1,04	8,80

4. Discussion

Understanding the dynamics of cattle ticks infestation is fundamental, not only to safeguard animal health and food, but also to mitigate the negative economic repercussions of their presence in the cattle industry (Paucar-Quishpe et al., 2023).

All ticks identified in this study were *R. microplus* in both regions analyzed. The results confirmed, as reported by other authors, that this species of tick is the one mainly affecting cattle in tropical and subtropical regions (Bermúdez Bajaña, 2018; Bustillos & Rodríguez, 2016; Orozco Álvarez, 2018; Paucar et al., 2022; Rodríguez-Hidalgo et al., 2017; Villamarín Álvarez, 2022).

On average, 99.39 ± SD 156.28 (0 - 1,426) *R. microplus* ticks per cattle were found. This represents a relevant data within the context of tick infestation in cattle. The value obtained could be considered relatively high when compared with other studies from the American continent, which reported values between 12 to 155 ticks per cattle (Castañeda Arriola et al., 2021; González-Cerón et al., 2009b; Miraballes et al., 2022; Rodríguez-Gallegos & Acosta-Rodríguez, 2011); however, it is important to consider that most animals were of dairy phenotype and breeds, and that these are more susceptible to tick infestation. It is important to note that the magnitude of infestation can vary significantly depending on the bioclimatic conditions of the geographic region and livestock management practices (Paucar et al., 2022). In the Ecuadorian context, studies such as Bustillos and Rodriguez (2016) mention an average of 70 ticks on dairy cattle, with minimum and maximum values of 4 and 119, respectively. Although they show an average similar to the one obtained in this study, the maximum values per animal in the present study result much higher, having few animals with extremely high infestations. This is expected due to individual susceptibility and the different conditions of each animal and farm. The need to take these aspects into consideration when deciding and applying tick control methods on each farm is also stressed.

This study found a greater number of ticks in certain anatomical zones of the animal, being “anatomical zone 3” which included the sacrum, perineum, posterior udder, crotch and posterior legs the most relevant area of infestation. This is similar to the results obtained by Jacho Merino (2015), who evidenced that the udder and between legs were the anatomical zones with more ticks. This is important because it implies that acaricide products are not properly applied. It is important to emphasize that at the moment of applying the acaricide, the whole animal should be taken into account, starting to apply the acaricide from the head of the animal and continuing backwards, by following a systematic pattern to cover the whole body and avoiding sensitive areas such as the eyes, nose and mouth. This is the only way to ensure that the acaricide is well distributed and covers the entire coat of the animal. Additionally, acaricide application should be avoided in strong wind conditions, as there is a risk of dispersing and affecting people and other animals nearby (Araque et al., 2014; Zintz, 2018).

A series of variables play a crucial role in the dynamics of parasitic infestation. In this study, the results of univariate statistical analysis showed a positive statistical association of the degree of infestation with the color of the cattle coat, being black the group that presented 3.25 times more probability of presenting a high level of tick infestation compared to white, brown and black/white cattle, as well as old animals with 2.98 OR, similar to results obtained by Bianchi et al. (2003). This characteristic has been observed in other studies and is related to the mimicry of ticks to avoid being observed by potential predators in animals with lighter coats. Animals with fair and poor apparent health presented OR values of 3.34 and 6.58 respectively, and animals with fair and poor apparent coat and skin health presented OR values of 4.76 and 24.68 respectively. Young animals were observed as a protective factor with an OR of 0.18, which indicates that older animals tend to have higher levels of infestation, this characteristic would in principle be related to the care that the owners give to the calves by not allowing them to exceed certain levels of infestation. In the multivariate model, only a positive association was observed with the apparent health of the coat and regular and bad skin with 5.13 and 25.83 OR, respectively. This differs from the results obtained by Paucar et al. (2022), who found a positive association with age, lactating cows and body condition. This may be due to the small number of animals in this study.

Based on the descriptive analysis, it was observed that certain groups within the individual variables presented a higher number of ticks. In this study it was observed that the taurine breed had a higher number of ticks, consistent with other studies (Wambura et al., 1998) where crosses of B. taurus breeds presented a higher number of ticks at count, which together with skin composition or immune response, were associated with a higher infestation of *R. microplus* (Jonsson et al., 2014).

In relation to age, this study showed higher tick counts in older animals, which is attributed to the immune system that tends to reduce with age, older animals tend to move less which makes it easier for ticks to attach to them in paddocks, or grazing areas. Contrary to, some studies revealing higher susceptibility in calves (Adugna & Tamrat, 2022; Miraballes et al., 2022).

Quantitatively, black cows showed a higher number of ticks, confirming, in line with other authors, that ticks tend to be more active in shaded and humid environments, and that the dark coat provides them with this condition. In addition, during days and periods of higher light intensity, dark coats can prevent them from drying out (Marufu et al., 2011). It is known that certain arachnids such as ticks use mimicry to go unnoticed in their environment, so being in dark fur could avoid being easily detected by predators (Pekár et al., 2011).

Females, especially from the category 'cows in production', seem more vulnerable to attack by a greater number of ticks, given that this category is usually in permanent grazing, and therefore ticks can more easily attach to cows (Bermúdez Bajaña, 2018; Rehman et al., 2017). Additionally, it is noteworthy that lactating cows experience a higher energy demand to sustain milk production, which exposes them to a physiological stress condition (Bianchi et al., 2003). The effectiveness of *Rhipicephalus microplus* is also explained by its own biology, which is characterized by stalking the host and attaching to animals when they approach vegetation (Polanco Echeverry & Ríos Osorio, 2016). On the other hand, males and cattle that are not producing milk, including animals that are in feedlots or grazing pastures with long resting periods, generally have less exposure to the environment and, as a result, have lower tick infestation (Adugna & Tamrat, 2022). This information is consistent with the results of this study, since female cows had higher numbers of ticks compared to males. As well as the cattle (that were) in lactation.

The body condition revealed a higher number of ticks in animals with low body condition, whose results are consistent with other studies that indicate that animals with low body condition have a higher probability of infestation compared to those in good condition (Adugna & Tamrat, 2022). This phenomenon is apparently due to a lower resistance of animals with low body condition. On the other hand, it is important to note that low body condition could rather be a consequence of high tick infestation. This means that ticks cause stress in animals, alter their behavior and reduce feed intake due to itching and irritation caused by these ectoparasites, which can also occur in animals with apparently normal health (Solomon & Tanga, 2020).

A greater number of ticks was observed in animals with slow capillary filling. In this regard, it is known that ticks feed on the blood of their hosts, which could cause anemia in highly infested animals (Anderson & Magnarelli, 2008; Polanco Echeverry & Ríos Osorio, 2016). The presence of ticks in cattle was associated with a higher quantity of these parasites in animals with poor skin and coat quality. This phenomenon negatively impacts the coat quality of cattle, since ticks, being parasites, generate adverse effects on the health and behavior of cattle. The constant feeding activity of ticks causes itching, irritation and discomfort on the skin of infested cattle. In response, affected animals tend to scratch and rub against objects or structures available in their environment to relieve itching, which can result in damage and tearing of the coat (Khan et al., 2022).

As to seasonality, in the Quijos region, two opposing seasons can be clearly distinguished in terms of rainfall patterns. The months of January, February and March are characterized by dry periods, while the rainy season goes from July to September, corresponding to the months where the fewest ticks were reported in this locality. This contrasts with the northwestern region of Pichincha, where the dry months occur between June and September, months in which fewer ticks were reported, and the rainy season extends towards the end of the year and the beginning of the new year. This climatic contrast underscores the high variability in tick presence throughout the year regardless of season, as tick infestation has been reported year-round (Bustillos & Rodriguez, 2016; GAD Quijos, 2015; GAD San Miguel de los Bancos, 2015).

It should be noted that the higher number of ticks found per animal in northwestern Pichincha could be attributed to the effect of the higher average temperature in this region with respect to the Quijos valley, especially in those sheltered sites such as Pedro Vicente Maldonado that records higher temperatures fluctuating between 20 and 25° C, and also to the high rainfall. It is discussed whether the characteristics of the predominant pastures of the genera *Setaria* and *Bracharia* in both regions provide a favorable scenario for the development of the immature stages of ticks (Condo Velastegui, 2023).

This study worked with 30 cattle farms, providing a detailed and valuable insight into the local situation with respect to tick infestation in cattle. However, it is essential to recognize that this limited sample size may present challenges when attempting to generalize results to a macro level. Inherent variations in environmental conditions, livestock management practices, and other factors at the regional level may not be fully represented in this limited sample. Therefore, although the findings provide insight into local realities, caution should be exercised in extrapolating these results to a broader scale. Diversity in agricultural practices and climatic conditions among different regions could influence the results significantly. More extensive and comprehensive studies are recommended. Increasing the number of participating farms and the number of animals per farm in future research would allow capturing a wider range of conditions and management practices. Although this could imply a considerable investment in terms of resources and research time, the potential benefits would outweigh these limitations.

5. Conclusions

Understanding the infestation dynamics of *Rhipicephalus microplus* is critical to safeguarding livestock health, ensuring food safety, and mitigating the negative economic impact on the livestock industry. Ticks identified in both study locations were consistent with *R. microplus*, in line with previous findings in the country. The average infestation of 99.39 ± SD 156.28 ticks per cattle highlights the importance of controlling tick infestation in cattle. Comparisons with other studies indicate a relatively high level of infestation, emphasizing the influence of cattle management practices. Anatomical analysis revealed that "anatomical zone 3," which includes the sacrum, perineum, posterior udder, groin and hind legs, is the most infested, suggesting suboptimal acaricide application. Several individual variables, such as coat color, age and health status, played a crucial role in the dynamics of parasitic infestation. The study also highlights the need to comprehensively understand the factors influencing tick infestation in order to implement effective control strategies.

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Contributor Roles

- Ximena Pérez-Otáñez: data curation - equal, formal analysis, investigation - equal, writing – original draft - lead.
- Valeria Paucar: data curation - equal, investigation - equal.
- José Buitrón: data curation - equal, writing – original draft - supporting.
- Claude Saegerman: conceptualization - equal.
- Jorge Grijalva: writing – review & editing - support.
- Cecilia Pérez-Escalante: writing – review & editing - supporting.
- Darío Cepeda-Bastidas: writing – review & editing - supporting.
- Susana Arciniegas: writing – review & editing - support.
- Sandra Enríquez: methodology - equal, writing – review & editing - supporting.
- Lenin Ron-Garrido: methodology - equal, writing – review & editing - supporting.
- Sophie O. Vanwambeke: conceptualization - equal, funding acquisition - equal.
- Richar Rodríguez-Hidalgo: conceptualization - lead, funding acquisition - equal, writing – review & editing.

Ethical Issues

All animals involved in the research were treated in accordance with animal welfare guidelines and standard procedures for on-farm tick collection. Farmers were informed and provided written consent prior to conducting scheduled visits. Data collected from animals on each farm were coded using specific numbers and letters referring to the farm and locality visited. The data were analyzed respecting the confidentiality and privacy of the participants involved in the study.

Conflict of interest

The authors declare that they have no affiliation with any organization with a direct or indirect financial interest that could have appeared to influence the work reported.

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