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SHORT COMMUNICATION

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Breed and non-genetic risk factors associated with the prevalence of subclinical mastitis in livestock systems of Arauca, Colombian orinoquia

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ABSTRACT

Subclinical Mastitis (SCM) is caused by several factors associated with the interaction of the individual cow, cow management, and the environment. The aim of this study was to assess the breed and non-genetic risk factors on the prevalence of SCM in cows from the dual-purpose livestock system on the floodplain of Arauca, Colombian Orinoquia. Milk samples were taken from the individual mammary quarters of 481 cows representing 28 different farms where the electrical conductivity (EC) test applied. To determine the factors associated with SCM, a multiple logistic regression analysis was used. The response variable was the SCM presence (1), or absence (0) obtained with the EC test. Breed was included as a genetic risk factor, and as non- genetic risk factors: number of cows in production, daily milk production, lactation month, cow age, climatic period, body condition, and calving number. The factors that were significantly associated with the SCM presence were body condition, climatic period, and breed ($p <$ 0.05). Odds ratio (OR) analysis of significant effects indicates that for each unit increase in body condition, the OR of having animals with SCM is reduced by 71%. In the dry period the OR of animals with SCM increases by 150% compared to the rainy period. Composite breeds reduce the OR of SCM animals by 73%, compared with *Indicus*-predominance animals. In this study, the occurrence of SCM in extensive management systems in Arauca, Colombian Orinoquia, is determined by the risk factors of breed, climatic period, and body condition.

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1. Introduction

Bovine mastitis can be defined as the inflammation of the mammary gland that occurs in response to physical injury or infection by pathogenic microorganisms and affects the dairy industry worldwide [1–3]. The presence of subclinical mastitis (SCM) is difficult to eradicate since it is caused by multiple factors of diverse nature: environmental, physical, mechanical, management, production system, production level, as well as nutritional and/or infectious processes, genetic traits, associated both in the cow and its environment [4]. The SCM generates economic losses by causing a reduction in milk production and quality [5,6]. Due to its asymptomatic nature, SCM it is difficult to diagnose [7], and milk is apparently normal [5].

Regarding the quality of milk, the SCM mainly influences the composition of the milk, including reduction of calcium, phosphorus, protein, fat, and an increase in sodium chloride [8]. Other studies have identified that size of the farm, the climatic period, and the number of cows in the farm influence the presence of SCM [9–11]. Among the factors associated with the cow are genetic traits [12–14], the calving number, days of lactation, the level of production, and reproductive intervals [12,15]. Hygiene and health during milking are key points in mastitis management [6]. Poor milking practices are a common cause of the spread and prevalence of mastitis in herds. Rainy seasons are a predisposing factor for the proliferation of pathogenic microbial transmission, increasing SCM cases [16]. At milking level, the risk factors associated with SCM include not sealing the mammary quarter at the end milking, inadequate washing of the udder and mammary quarters [10,17,18], cowshed hygiene [15] and hand washing [19]. On the other hand, the disease causes public health problems, especially through the consumption of raw milk [20].

At the farm level, a widely used and highly efficient method for SCM diagnosis is the measurement of Electric Conductivity (EC), used since 1942 with different types of conductivity metres

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[21,22]. The EC test has been used in recent decades to monitor and diagnose SCM at the farm level [23–25]. The EC test detects milk changes due to the increase in sodium chloride ions that occur in inflammatory processes, leading to an increase in the electrolytic milk component [25–27]. Test has practical importance since it allows a quick on-farm diagnosis without many technical demands [2,28].

In Arauca, Colombia, milk production mainly comes from the dual-purpose livestock system developed under tropical conditions, with low inputs and lower technological levels. The dual-purpose livestock system involves multiracial groups resulting from crossbreeding zebu cattle with European breeds. In the study area, the microbiological examinations for the detection of SCM are difficult due to public order problems, distance from urban centres and difficulty of access to farms due to lack of passable access roads. The udder health status of milking animals is unknown, due to the scarce reports available in the area. Therefore, the practical applicability of EC test on-farm allows rapid diagnosis of SCM without the need for expensive and time-consuming microbiological culture [29]. The risk factors study for the SCM prevalence allows us to measure the presence of an animal health problem in an area with a livestock attitude [8]. The aim of this cross-sectional study was to assess the breed and non-genetic risk factors the prevalence of SCM in cows from the dual-purpose livestock system in the floodplain of Arauca, Colombian Orinoquia.

2. Material and methods

2.1. Study site

The study was carried out in Arauca department, eastern Colombia (Latitude: 7º 5'5" N; Longitude: 70º 45' 32.7" W) (Figure 1). The climatic regime corresponds to a dry period (November–April) with a relative humidity of 65% and an average ambient temperature of 32.6°C; and a rainy period (May–October) with a relative humidity of 85% and an average ambient temperature of 30.1°C. The annual rainfall is less than 1500 mm [30,31].

The farms where the animals were sampled belong to small producers with a low technological level. The farms were chosen for convenience after a meeting with the Livestock Association Producers of the region. The average farm size was 65 hectares (ranging from 30 to 100 hectares). In most farms, animal feeding is based on extensive grazing systems with grasses, mainly *Brachiaria decumbes*, *Brachiaria arrecta*. In some farms Sodium Chloride (white salt) is provided, and to a lesser extent mineralized salts. An average of 18 dairy cows per farm were found (range 7 t0 57 cows). Milking is done by hand (once a day), with the calf's presence and is done under covered cowshed or in open environments. The calf suck before milking to stimulate milk let-down and then suck after milking; this is a normal handling in tropical dairy production [29]. At the time of milking, the udder washing of the cows is not observed. There is no health plan. The destination of the milk is for self-consumption, homemade or artisan production, reception in collection centres, or direct sale to consumers.

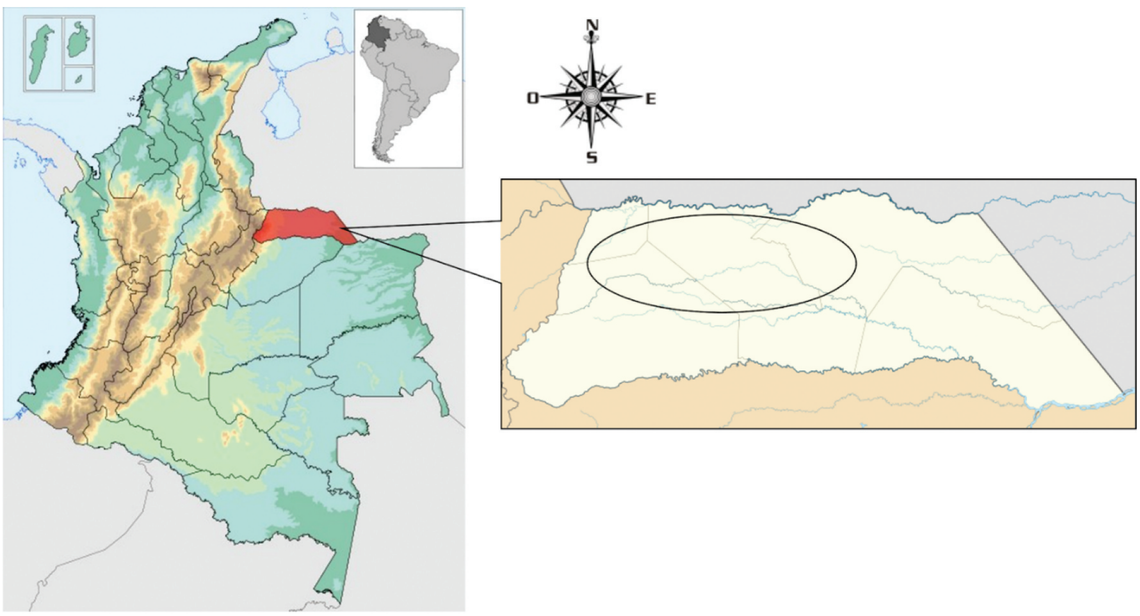

Figure 1. Red color: location of the department of Arauca, Colombian orinoquia. Circle: area where the farms were sampled.

2.2. Evaluated animals

Individual milk samples from 1924 mammary quarters belonging to 481 cows from the dual-purpose livestock system (Bos indicus × Bos taurus multiracial crosses) (Figure 2), aged 3 to 10 years, from 1 to 8 calving's, and 1 to 7 lactation months, from 28 farms in 19 Territorial Division Center, were evaluated.

Due to the lack of genealogical records the animals, the cows breed classification was based on their external appearance and the information provided by the producers as recommended by other authors [32,33]:

- *Taurus-Indicus* (F_1 cows Zebu × Holstein, Zebu × Brown Swiss, Zebu × Norman, Zebu × Simental),
- *Indicus* predominance (cows with external appearance less than 50% European with phenotype of the Gyr, Guzerat, Brahman breeds)
- Composite breed (Girolando breed cows).

Milk samples were taken from the cows in the morning (4:00 am to 6:00 am). Only cows with functional mammary quarters and without antibiotic treatments during the last 3 months were included in the study. The samples were taken during the rainy period (May to October 2021) and during dry period (November to April 2022), following the recommendations available in the literature [2,23,24,34]. Mammary quarters were identified, and samples were taken in that order (Figure 3).

2.3. Electrical Conductivity (EC) test

The EC test measures the conduction capacity of electrical currents between two electrodes, which is expressed in mS/cm (milliSiemens/centimetres) and the result is expressed in units [2,35,36]. EC test for the detection of SCM is based on the differences in the salt concentration (sodium chloride) between infected and non-infected mammary quarters in the same cow

Figure 2. Cows from the dual-purpose livestock system of Arauca department, eastern Colombia.

Figure 3. Mammary quarters position for milk sampling. RP= right Posterior; LP= left Posterior; RA= right Anterior; LA= left anterior.

due to a bacterial infection in the milk [21,22,37]; lactose potassium ions decrease, while the pH increases [28]

For the CE test, the Draminski Mastitis Detector MD4 × 4Q equipment (Draminski S.A., Owocowa, PL) was used [35]. The measurement equipment has a platform with four measuring containers (cups) to deposit the "milk jets" and a measuring and reading block with a special liquid crystal screen, where the results are displayed. The measuring equipment was placed below the mammary quarters and each cup was filled with an amount of milk that did not exceed 1 cm from the upper edge. Then the switch button was pressed and after 3 seconds the results of each mammary quarter were displayed (Figure 3)

The results were interpreted according to the manufacturer's instructions available in the equipment's user manual ISO 9001 [35] as follows: 1) readings less than 250 units indicate "subclinical inflammation" of the mammary quarter or high risk of convert to acute mastitis, 2) a difference greater than 40–50 units between the highest and lowest mammary quarters scores of the cow examined indicates SCM. An example can be seen in Table 1.Interpretation:

Mammary quarter with highest score (RA):	380u
Mammary quarter with lowest score (LP):	310u
Difference:	70u

So that the mammary quarter affected with SCM is the left posterior (LP) since there is a difference of 70u between the highest and lowest score. The other mammary quarters remain normal.

2.4. Prevalence determination

Prevalence is an indicator of existence or "stock", as it considers all present cases, whether new or old, and refers to the number of cases that a disease or infectious event occurs in a given place and time [38]. The cow-level prevalence (with at least one affected mammary quarter), total mammary quarters prevalence, and mammary quarters prevalence by position were processed according to the following mathematical formulas [29,39,40]:

- Cow-level prevalence = (Number positive cows/ Total number sampled cows) x 100
- Prevalence at the level of total mammary quarters = (Total number positive mammary quarters/ Total number sampled mammary quarters) x 100

Table 1. Example of EC results interpretation among mammary quarters.

Right Posterior (RP):	370u
Left Posterior (LP):	310u
Right Anterior (RA):	380u
Left Anterior (LA):	370u

- Prevalence at the level of the mammary quarter by position = (Total number positive mammary quarters per position/Total number mammary quarters per position) x 100

Test data at the farm level were compiled and organized in Excel format for further analysis. The collected information included data related to risk factors: cow age, daily milk production, lactation month, body condition, climatic period, number of cows in production, and breed. The body condition was subjectively evaluated at the time of taking the milk sample, on a scale from 1 to 5, where 1 is a very thin cow and 5 represents a very obese cow [41].

2.5. Statistical analysis

Frequency tables were estimated for the variables related to the positive cows numbers (animals with any degree positivity to SCM in any mammary quarter declared by the test), the number of positive mammary quarters number per cow (1 to 4) and positivity of mammary quarters by position (RP, LP, RA, LA).

To determine the factors associated with SCM, a multiple logistic regression analysis was used, widely employed for this type of study [42]. The response variable was the SCM presence (1), or absence (0) obtained with the EC test under the following model:

$$N_i = log(\pi/(1-\pi)) = M + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \beta_5 X_5 + \beta_6 X_6 + \beta_7 X_7 + \beta_8 X_8 + \varepsilon i$$

Where:

N_i =	i-th modeled probability of having SCM-positive animals.
π =	odds ratio: (1-π) probability of not having the presence of SCM
M =	Pending
X_1 =	Effect the age variable (3 to 10 years)
X_2 =	Effect the milk production variable (2 to 12 L)
X_3 =	Effect the lactation month variable (1 to 7)
X_4 =	Effect the body condition variable (1 to 5)
X_5 =	Effect the climatic period variable (rainy and dry)
X_6 =	Effect the cows number variable (1=7 to 16 cows; 2=17 to 26 cows; 3= > 26 cows)
X_7 =	Effect the breed variable (1=*Taurus-Indicus*; 2=*Indicus* predominance; 3=composite breed)
X_8 =	Effect the calving number variable (1 to 8)
εi =	Accumulated error
β_i =	Regression coefficients associated with each independent variable

To compare the variables of the number of cows and breed, dummy variables were created and the category with more than 26 cows (group 3) and animals with an *Indicus* predominance (group 2) were taken as a reference point, respectively. Breed was included as a genetic risk factor; and as non-genetic risk factors, number of cows in production, daily milk production, lactation month, cow age, climatic period, body condition, and calving number were included. The analyzes were carried out using the statistical software Infostat [43].

3. Results

A total of 1924 mammary quarters corresponding to 481 cows from the dual-purpose livestock system were sampled. The number of cows with mammary quarters affected by SCM, within the evaluated sample, is shown in Table 2.

Results showed that 143 cows were positive for SCM according to the EC test, indicating a cow-level prevalence of 29.7% (143/481). The cow-level prevalence indicates whether a cow presented the disease or at least one its mammary quarters were positive.

The SCM prevalence level for total mammary quarters and for mammary quarters by position are presented in Table 3. The EC test showed total mammary quarters prevalence level 11.3% (218/1924); while the highest mammary quarters prevalence level by position was for LA (12.1%) and RP (11.9%).

Table 4 shows the SCM prevalence according to the number of cows in production, lactation month, calving number, cow age, breed, daily milk production, body condition and climatic period. The lower prevalence of SCM was observed with 17 to 26 cows in production (24.2%), in cows with more of 3 lactation months (25.3%), in cows older than 5 years (24.8%), in cows with 3 calving (27.2%), in cows with a body condition of 3.6–4.5 (29.4%), and during the rainy period (25.3%), in cows with 2 to 5 L daily milk production (29.1%). Regarding breed, the lowest prevalence of SCM was observed in the composite breed (22.7%).

Table 2. Cow-level prevalence and number of cows with affected mammary quarters in the sample evaluated in the dual-purpose livestock system of Arauca, Colombian orinoquia.

Total sampled cows		481	%
Positive cows		143	
Cow-level prevalence			29.7
Affected mammary quarters[1]			
	1	87	18.1
	2	40	8.3
	3	13	2.7
	4	3	0.6

[a]= Cows with 1,2,3 and 4 affected mammary quarters.

Table 3. Prevalence of SCM (%) at level of total mammary quarters and at level of mammary quarters by position (RP, LP, RA, LA) by EC test in a dual-purpose livestock system of Arauca, Colombian orinoquia.

Total mammary quarters sampled		1924	%
Positives		218	
Prevalence at the level of total mammary quarters			11.3
Prevalence at the level of mammary quarters by position			
Mammary quarters	n	Positives	
RP	481	57	11.9
LP	481	54	11.2
RA	481	49	10.2
LA	481	58	12.1

n= number mammary quarters by position; RP= Right Posterior; LP= Left Posterior; RA= Right Anterior; LA= Left Anterior.

Table 5 presents the factors that were significantly associated with the presence of SCM in the evaluated systems. The logistic regression analysis showed that most of the factors did not influence the SCM positivity found with the EC test, with the exception of body condition, climatic period, and breed ($p < 0.05$). The analysis of the odds ratio (OR) of the significant effects indicates that for each unit of increase in body condition, the OR of having sick animals with SCM is reduced by 71% (1–0.29). In the case of the climatic period, it was found that, in drought, the OR of sick animals with SCM increases by 150% (2.5–1) compared to the rainy period. Finally, it was found that animals of the composite breed reduce the OR of sick animals with SCM by 73% (1–0.27), compared to animals with an *Indicus* predominance. Based on these results, statistically significant risk factors associated with the SCM prevalence will be discussed.

The body condition was a statistically significant factor associated with the prevalence of SCM ($p = 0.008$; OR = 0.29), with higher prevalence (36.1%) observed in the animals with the lowest body condition score (2.5–3.5). The climatic period is a statistically significant factor associated with the prevalence of SCM ($p = 0.011$; OR = 2.5), with a higher prevalence during the dry period (55.7%), compared with rainy period (25.3%). The composite breed was a statistically significant risk factor associated with the presence of SCM ($p = 0.029$; OR = 0.27), with a lower prevalence value (22.7%), compared to *Taurus-Indicus* (28.4%) and with *Indicus* predominance (32.6%) (Tables 4 and 5).

4. Discussion

In the current study, the EC test detected a cow-level prevalence of SCM similar to that reported in another study [34] and is considered low compared to other studies in tropical regions [8,17,40,44]. It is stated that milk from a cow with udder inflammation is a better conductor of electrical waves than milk from a healthy cow [45]. Milking with the support of the calf is important since the calf not only stimulates milk letdown, but at the end of the milking process it consumes the residual milk, limiting the bacteria development in the mammary gland that induce of SCM presence [46].

Several studies have reported that genetic traits, lactation period [47,48], the calving's number, lactation days, the production level [15,49], the size of the farm, the climatic period, the numbers of cows in the farm, and cow age [9,18], are risk factors that predispose to the SCM presence. The results obtained in this study showed the significant factors on the prevalence of SCM were body condition, climatic period, and breed ($p < 0.05$). Furthermore,

Table 4. Prevalence of SCM (%) according to the number of cows in production, daily milk production, lactation month, cow age, climatic period, body condition, calving number, and breed of Arauca, Colombian orinoquia.

Variable	RP	LP	RA	LA	Positive mammary quarters	Positive cows	Sampled cows	%
Number of cows in production								
7–16 cows	29	26	22	23	100	63	153	41.2
17–26 cows	11	18	20	21	70	47	194	24.2
>26 cows	17	10	7	14	48	33	134	24.6
Daily milk production/cow								
2 to 5 L	29	25	25	28	107	69	237	29.1
6 to 12 L	28	29	24	30	111	74	244	30.3
Lactation month								
1–3 months	44	37	34	44	159	104	327	31.8
More than 3 months	13	17	15	14	59	39	154	25.3
Cow age								
3 and 4 years	21	15	19	19	74	46	153	30.1
5 years	16	18	11	16	61	38	153	24.8
6 or more years	20	21	19	23	83	59	175	33.7
Climatic period								
Rainy	43	40	36	47	166	104	411	25.3
Dry	14	14	13	11	52	39	70	55.7
Body condition								
2.5–3.5	31	28	21	34	114	75	208	36.1
3.6–4.5	26	26	28	24	104	68	273	24.9
Calving number								
1	16	11	15	16	58	38	116	32.8
2	18	20	16	13	67	46	165	27.9
3	10	10	10	12	42	25	92	27.2
>4	13	13	8	17	51	34	108	31.5
Breed								
Taurus-Indicus	34	31	25	30	120	78	275	28.4
Indicus predominance	21	20	24	26	91	60	184	32.6
Composite breed	2	3	0	2	7	5	22	22.7

RP= Right Posterior; LP= Left Posterior; RA= Right Anterior; LA= Left Anterior.

Table 5. Odds ratios of the variables included in the logistic regression for the associated factors with the SCM prevalence in dual-purpose livestock systems of Arauca, Colombian orinoquia.

Regressor variables	OR	Wald LI-LS (95%)	*p*-value
Non-genetic factors			
Age	1.1	0.84–1.44	NS
Calving's	0.96	0.7–1.32	NS
Lactation	0.85	0.72–1.01	NS
Body condition	0.29	0.11–0.72	0.008
Period	2.5	1.23–5.06	0.011
Number of cows[a]			
7 a 16	1.56	0.86–2.85	NS
17 a 26	0.95	0.56–1.62	NS
Milk production	0.93	0.82–1.07	NS
Genetic factors			
Breed[b]			
Taurus-Indicus	0.72	0.46–1.12	NS
Composite breed	0.27	0.09–0.88	0.029

OR: odds ratio; Wald LI-LS (95%): Wald confidence limits to 95%; [a] = Reference point group 3 (> 26 cows).
b = Reference point group 2 (Indicus predominance); NS = Not-significant.

it is important to clarify that most of the research has been carried out in specialized dairy livestock systems. Therefore, our purpose was to evaluate the risk factors in a dual-purpose livestock system where milking is done by hand and presence of the calf.

"Body condition is defines as the ratio of body fat to nonfat components in the body of a live animal". "The body condition score is an assessment of a cow's body fat reserves, where low values indicate emaciation and high values indicated obesity" [50]. The results of this study showed that body condition is a statistically significant factor associated with the prevalence of SCM in cows from the dual-purpose system in Arauca (p = 0.008; OR = 0.29). The highest value of SCM prevalence was observed in the animals with the lowest score in body condition (2.5–3.5). These results are lower than those reported in other studies in which cows with poor body condition presented a prevalence of mastitis of 60.4%, compared to 30% in cows with good body condition, associated with decreased immune status that predisposes the udder to opportunistic infection [51]. Body condition can decrease 0.25 or more score in the peripartum, presenting a greater increase in polymorphonuclear apoptosis (important in immune defence), which affects a high prevalence of intramammary infection [52]. The nutrition, breed and management are factors that possibly interact with body condition and may determine the risk of health disorders [50]. Therefore, a body condition score of 2.5 or lower is considered a potential risk factor for the presence of SCM [49].

Other studies conclude that cows must gain a certain body condition during the dry period (near calving) to maintain the physiological processes that preserve energy balance and performance, and thus decrease the probability of presenting SCM [53]. In contrast to low body condition, it has been reported in cows from organic ecological herds that arrived 6 weeks prior to calving with body condition ≥3.75, had a 3.12 higher probability of intramammary infection ($p < 0.01$) [54].

The present study showed that the climatic period is a statistically significant factor associated with the SCM prevalence in cows from the dual-purpose system in Arauca ($p = 0.011$; OR = 2.5). A higher prevalence of SCM was observed in the dry period, compared to the rainy period. These results are possibly associated with the high temperatures which for Arauca averages 32.6°C in the dry period, with a relative air humidity of 65% [30], which can cause a decrease in food consumption, causing a negative energy balance affecting their immunity and facilitating the colonization of pathogens in the mammary gland. The results are similar to those reported in Mexico for milking cows where the prevalence was higher in summer (drought, temperature >42°C) [55], possibly due to heat stress caused by high temperatures in the region in the dry season. In Arauca, the production system is extensive, where the supply and quality of pastures depends on weather conditions, rainfall, temperature, and soil quality.

In other studies, it has been reported that an increase in the temperature index can negatively affect the supply of nutrients and vitamins that directly affect the immunity of the animal [56]. Studies have reported that heat stress occurs when the temperature and humidity index is ≥71, causing cattle to reduce feed intake, immunity, in milk presence of somatic cells, and mastitis in dairy cattle [57]. Researchers in India reported an increase in mastitis related to increased temperature and humidity index in dairy cows during hot weather [58]. Likewise, it is indicated that temperature and humidity index values ≥79 are associated with a high incidence of mastitis in first calving cows [56].

In a study carried out in Colombia [59] no significant differences were found in the prevalence rates in relation to the months of the year; however, the highest levels occurred in April/May and September/October/November, when the rainfall is at its heaviest. These results differ from ours, which indicates the need to carry out studies of risk factors in different regions with respect to the climatic period.

In this study, the composite breed was a statistically significant risk factor associated with the SCM prevalence ($p = 0.029$; OR = 0.27), with a lower prevalence value, compared to *Taurus-Indicus* and those with *Indicus* predominance. Various studies have reported that the breed effect is related to the presence of mastitis [3,60]. In a prevalence study in adapted zebu breeds, Holstein Friesian crosses with local zebu breeds and Jersey breeds, a higher infection of the mammary gland was detected in Jersey cows (78.6%), and crossbreeds (51.9%) compared with the adapted zebu breeds (16.7%) [51], which implies that the presence of SCM is associated with high-yielding cows [61]. The higher prevalence of SCM reflected in *Taurus-Indicus* cows and *Indicus* predominance, suggests that milk production with hand milking and with the calf presence in the dual-purpose cattle system in Arauca with composite breed may be a less associated risk factor.

In another study, they found that breed and/or crossbreeding was not a risk factor in the prevalence of SCM, while the calving number (primiparous and multiparous), geographic region, and milk production (<10 L, 10–20 L or >20 L) and the region significantly contributed ($p = 0.036$) to the risk of SCM. In multiparous cows, the odds of SCM were 2.51 times higher than the odds in first calving cows [34]. This information differs from the present study, where breed was a risk factor for the prevalence of SCM, with a lower risk factor in the composite breed compared to the taurus-indicus and indicus predominance studied. Cows of breeds with high milk production are more susceptible to the presence of SCM [62] due to the size of the teats that can easily become loose and allow the entry of pathogens, ultimately causing an infection [63].

5. Conclusions

The cow level prevalence of SCM detected in the present study is considered low compared to other studies in tropical regions. The SCM occurrence in extensive management systems of Arauca, Colombian Orinoquia is determined by the risk factors of breed, climatic period, and body condition. The low body condition of the cow in the period of drought is a factor that predispose to the presence of SCM, probably due to the nutrient scarcity and high temperatures, which can generate stress in the animal. Under extensive management conditions and manual milking, the composite breeds, according to the results of this study, are less susceptible to the presence of SCM. The number of cows in production, the daily milk production, the cow age, the calving number, and the lactation period were not risk factors for the presence of SCM. According to the current results, it is suggested to carry out studies of risk factors associated with SCM in different regions and in dual-purpose livestock production systems.

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Writing, Preparation of the Original Manuscript, A. S-C.; M. V-T; Methodology and Formal Analysis, M.V-T.; R.T.; A. S-C.; Statistic Analysis: M.V-T; R.T; Review, Edition and Conceptualization, R. J-J., P.M.p-C., D.P.B-P. All Authors have read and accepted the published version of the manuscript.

Data availability statement

Data is available upon reasonable request to the second author.

Ethical approval

Bioethical Concept No. BIO163 issued by the Research Bioethics Subcommittee of the Bucaramanga Sectional Universidad Cooperativa de Colombia (Act No. 4, 4 November 2021)

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