Risk factors associated with leptospirosis in dual-purpose cattle based on the One Health approach in the department of Huila, Colombia

C.A. Murcia[1], *N.V. Acevedo*[1], *S. Falla-Tapias*[1*], *W.O. Burgos-Paz*[2].

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ABSTRACT

Leptospirosis significantly impacts beef and dairy cattle production, particularly in tropical regions, although it remains a disease of global relevance. This zoonotic disease primarily causes abortions during the final third of gestation. The present study aimed to analyze the risk factors associated with leptospirosis in the southern Andean region of Colombia. To this end, 360 cattle were sampled across 24 municipalities within the department of Huila. A seroprevalence of 49.2% was determined using enzyme-linked immunosorbent assays (ELISA). These results were subsequently correlated with various variables obtained through epidemiological surveys.

The findings revealed that the primary risk factors for leptospirosis were linked to inadequate livestock management practices. These included improper methods of food storage, failure to implement quarantine protocols for infected animals, the introduction of new fattening cattle into the herd without proper precautions, infrequent weighing of animals, poor storage management of veterinary medications, lack of segregation of sick animals, insufficient estrus detection practices, inadequate hand hygiene when interacting with the herd, and the use of unclean instruments during routine procedures. Conversely, the presence of well-defined internal pathways and the separation of poultry production from cattle operations were identified as protective factors. Understanding these risk factors is critical from a One Health perspective, as it facilitates the implementation of preventive measures aimed at safeguarding both animal and human health.

Keywords: *Leptospira* spp., abortions, ELISA, livestock, human health.

Factores de riesgo asociados con la leptospirosis en bovinos doble propósito basado en el enfoque Una Salud en el departamento del Huila, Colombia

RESUMEN

La leptospirosis afecta la producción de ganado vacuno de carne y leche principalmente en regiones tropicales, aunque es una enfermedad de importancia mundial. Esta enfermedad ocasiona abortos en el último tercio de la gestación. El propósito del presente estudio es desarrollar planes de prevención de la enfermedad que se ajusten a la realidad

[1] Corporación Universitaria del Huila CORHUILA. Faculty of Veterinary Medicine and Related Sciences, Program of Veterinary Medicine and Zootechnics, KYRON Research Group, Calle 21 # 6-01, Neiva, Huila, Colombia.

[*] Correo electrónico: sergio.falla@corhuila.edu.co

[2] Corporación Colombiana de Investigación Agropecuaria - AGROSAVIA. Turipaná Research Center, Km 13 vía Montería-Cereté, Córdoba, Colombia.

del entorno, considerando el enfoque Una Salud. En este contexto, se seleccionaron 360 bovinos de 24 municipios del departamento del Huila. Se determinó una seroprevalencia de 49,2% para leptospirosis mediante ensayos inmunoabsorbentes ligados a enzimas (ELISA). Los resultados se correlacionaron con una serie de variables obtenidas a través de encuestas epidemiológicas. Los principales factores de riesgo están asociados a las malas prácticas de manejo del ganado, que incluyen los métodos de almacenamiento de alimentos, la aplicación de protocolos de cuarentena para animales infectados, la introducción de nuevos animales de engorde al hato, la frecuencia de pesaje en báscula, el manejo adecuado del almacenamiento de medicamentos, la segregación de animales enfermos, la detección de estros, el lavado de manos al interactuar con el hato y el uso de instrumentos limpios utilizados en los procedimientos habituales. Además, se encontraron factores protectores como tener vías internas bien definidas y mezclar la producción con aves. Comprender estos factores de riesgo es de gran utilidad desde una perspectiva de Una Salud, ya que permite la implementación de medidas para proteger tanto la salud animal como la humana.
Palabras clave: *Leptospira* spp., aborto, ELISA, ganado, salud humana.

INTRODUCTION

Zoonotic diseases are infections that vertebrate animals can transmit to humans. Under natural conditions, the infectious agents responsible for these diseases include bacteria, viruses, parasites, fungi, rickettsia, among others (Tomori & Oluwayelu 2023). Among these, the bacteria causing leptospirosis have garnered significant interest due to the disease's notable impact on public health and its contribution to both reproductive and non-reproductive losses in cattle production (O'Doherty *et al.* 2014). While the disease predominantly affects beef and dairy cattle production in tropical regions, it remains a concern of global significance (Bharti *et al.* 2003). Mammals, including humans, are the most frequent hosts in endemic areas. The challenge of clinical diagnosis is further compounded by the similarity of leptospirosis symptoms to those of other diseases such as dengue, parvovirus, and hepatitis (Faine 1994; Romero *et al.* 2010). It is crucial to recognize that animal species maintain a relationship with specific *Leptospira* serovars. Advances in isolation methods from environmental samples have enabled the identification of 68 *Leptospira* species to date (Arent *et al.* 2022).

Leptospirosis in beef cattle significantly compromises livestock production due to its severe reproductive consequences, including stillbirths and abortions (Sohm *et al.* 2023). This disease remains a critical subject of study and evaluation to guide future strategies for its control, prevention, and potential eradication. In 1980, leptospirosis accounted for approximately 40.8% of abortions in beef cattle in Venezuela, while in Ireland, this figure reached 49.7% (Ellis *et al.* 1982). Despite its impact, leptospirosis has not been classified as a notifiable disease by the Colombian Agricultural Institute (ICA, by its initials in Spanish), as it is not mandated by the World Organization for Animal Health (WOAH) (Instituto Colombiano Agropecuario 2015). Consequently, there is limited official information on the economic burden of leptospirosis on Colombia's national dairy and beef production.

Several studies conducted in Colombia identified a total of 2,426 serovars from 1,586 positive cases, with *L. hardjo* being the most frequently detected, reflecting its status as a reservoir in ruminants, pigs, and equines (Ensuncho–Hoyos *et al.* 2017; Olivera *et al.* 2018; Pulido *et al.* 2017; Estupiñán *et al.* 2018; Vargas *et al.* 2018; Baena *et al.* 2018; Guzmán–Barragán *et al.* 2022). Among bovines, the most common reservoirs identified were *L. hardjo*, *L. tarassovi*, *L. pomona*, and *L. grippotyphosa* (Balamurugan 2016).

Humans can acquire *Leptospira* spp. either directly or indirectly through the skin or via the ocular, oropharyngeal, and nasopharyngeal mucosa (Suárez Conejero *et al.* 2015). The primary reservoirs of this disease are rodents and dogs, which excrete the bacteria in their urine, thereby contaminating both indoor and outdoor environments (Abgueguen 2014). In beef cattle, transmission typically occurs indirectly through contact with contaminated water or soil, as well as through uterine secretions, semen, and urine (Munoz–Zanzi *et al.* 2020). Consequently, the infection enters the body via mucous membranes or damaged skin (Ellis 2015).

Leptospirosis progresses through distinct phases. The disease begins with an acute or septicemic phase, lasting approximately one week. This is followed by the immune phase, during which antibodies are produced, and *Leptospira* spp. is excreted in the urine for a brief period. The quantity of bacteria excreted during this phase is minimal, and the bacteria are typically non-viable, classifying the host as accidental (Monte *et al.* 2015). In the chronic stage, the pathogen primarily localizes in the kidneys and the genital tract of carrier animals (Faine 1994).

Leptospirosis has been extensively studied across various species worldwide; however, research focusing on beef cattle remains insufficient (Goarant *et al.* 2019). As noted by Sohm *et al.* (2023), it is critical for public health to increase awareness and attention to leptospirosis due to its significant economic impact. Collaborative efforts between health professionals and veterinarians can assist farmers in preventing the spread of the disease, particularly in regions not yet classified as endemic. The growing practice of open grazing in Europe has further contributed to the resurgence of leptospirosis, underscoring the importance of studies on beef cattle for improving epidemiological understanding of the disease (Sohm *et al.* 2023).

Risk factors such as geographic location, inadequate management practices, animal purchasing, livestock movement, and biosecurity measures significantly influence the presence of leptospirosis (O'Doherty *et al.* 2014). Accordingly, the present study aims to analyze the risk factors associated with leptospirosis in the southern Andean region of Colombia.

MATERIALS AND METHODS

This study was conducted with the approval of the Ethics, Bioethics, and Scientific Integrity Committee of the Colombian Agricultural Research Corporation (Agrosavia, by its acronym in Spanish), as per Act N.° 2 of 2021. All biosecurity measures established on the farms were strictly followed, and sampling procedures adhered to a standardized protocol.

A cross-sectional investigation was undertaken in collaboration with the University Corporation of the Huila Region (Corhuila, by its acronym in Spanish), the Colombian Agricultural Research Corporation (Agrosavia, by its acronym in Spanish), the Committee of

Livestock Farmers of Huila (CGH, by its initials in Spanish), and the Governorate of Huila. Cattle were randomly selected from livestock farms located in the department of Huila. Out of the 37 municipalities in the department, 24 were randomly chosen and grouped into four geographic zones: the northern zone (Aipe, Algeciras, Baraya, Campoalegre, Colombia, Palermo, Rivera, Tello, Villavieja, and Yaguará); the central zone (Altamira, El Pital, Garzón, Gigante, Suaza, and Tarqui); the western zone (La Plata, Nátaga, Paicol, and Tesalia); and the southern zone (Acevedo, Pitalito, San Agustín, and Timaná).

The criteria for selecting the farms included membership in a livestock association, availability of suitable land for livestock use, maintenance of production records, and possession of sufficient economic resources for disease treatment. A total of 150 farms meeting these inclusion criteria were selected, and 360 cows were randomly chosen for the study. Data collected for each animal included a problem-oriented clinical record, an epidemiological survey, and a socioeconomic survey. Clinical examinations of all cattle across the herds were performed by veterinary medicine and zootechnics professionals. Clinical parameters evaluated included heart rate, respiratory rate, temperature, body condition, and weight. Additionally, the morphophysiological integrity of various systems (cardiovascular, respiratory, digestive, reproductive, and integumentary) and body parts was assessed. Blood samples were collected via jugular vein puncture from each bovine, and the presence of bovine antibodies was determined using the commercial Cow Leptospira spp. Antibody ELISA kit (Abbexa LTD, Cambridge, UK). The ELISA kit has a reported sensitivity of 94% and specificity of 99%, as per the manufacturer.

To identify risk factors associated with leptospirosis, both epidemiological and socioeconomic surveys were administered. These surveys comprised 230 potential risk factors, including biosecurity, sanitation, transportation, traceability, medication use, animal health, and feeding practices (epidemiological survey).

ELISA protocol

Blood samples were collected in 10 mL red-cap tubes and centrifuged at 3,070 Relative Centrifugal Force (RCF) for 5 minutes to separate the serum. To ensure preservation, the samples were stored at -20 °C to prevent protein degradation and denaturation.

Prior to processing, ELISA kit components and serum samples were thawed to room temperature. Serum samples were diluted with Sample Diluent Buffer at a 1:5 ratio, while the Wash Buffer was prepared by diluting it with distilled water at a 1:30 ratio, following the manufacturer's instructions.

A total of 50 µL of positive, negative, and blank controls (Sample Diluent Buffer) were aliquoted in duplicate, alongside 50 µL of each diluted sample, into the respective wells of the ELISA plate. The plate was incubated for 30 minutes at 37 °C in a dry oven (ARI Medical Technologies). After incubation, the plate was washed five times using 1X Wash Buffer. Next, the Detection Reagent was added, and the plate was incubated again at 37 °C for 30 minutes, followed by additional washes as specified by the manufacturer.

To develop the reaction, TMB Substrate A and TMB Substrate B were added to each well, and the plate was incubated at 37 °C for 15 minutes. Subsequently, 50 µL of Stop Solution was added to inactivate the enzymatic reaction. The optical density

(OD) of the samples was measured within 3 minutes after adding the Stop Solution, using a BOECO BMR-100 Microplate Reader equipped with a 450 nm filter.

STATISTICAL ANALYSIS

A Pearson correlation coefficient was calculated to evaluate the relationship between variables, focusing on the association between socioeconomic and demographic factors and the presence of the pathogen *Leptospira* spp. Binary logistic regression models were developed, where the dependent variable represented the ELISA test results (0: negative; 1: positive for leptospirosis). Risk factors were analyzed, and odds ratio (OR) were calculated for each, with a 95% confidence level (P-value < 0.05) to ensure the validity of the identified risk factors.

Descriptive analysis was performed using the statistical software R (version 4.3.3). This approach facilitated the determination of logistic regression models, and the identification of risk factors associated with the occurrence of the disease. Risk factors with significant associations, whether with the presence or absence of leptospirosis, were identified and highlighted.

RESULTS AND DISCUSSION

Out of the 360 samples analyzed, 177 (49.2%) tested positive for leptospirosis. The seroprevalence across different age groups was as follows: 12-45 months, 39% (23/59); 46-65 months, 49.6% (57/115); 66-85 months, 54% (67/124); 86-105 months, 50% (12/24); and ≥106 months, 40% (12/30) (table 1).

The highest prevalence by region was observed in farms located in the northern zone, with 52.5% (93/177), followed by the western zone at 48.3% (29/60), the central zone at 46.6% (27/58), and the southern zone at 43.1% (28/65) (table 2).

Out of the 360 cows analyzed, 85 (23.6%) had contact with canines, and 42 of these (49.4%) tested positive for leptospirosis. Similarly, 101 cattle (29.1%) had contact with equines, with 52 (51.5%) testing seropositive. Additionally, 28 cattle (7.8%) had contact with ovines, among which 13 (46.4%) tested positive for leptospirosis (table 3).

TABLE 1. Seroprevalence of leptospirosis according to different age groups

Age groups (months)	Frequence	Seroprevalence	95% Confidence interval	
			Lower limit	Upper limit
12-45	23/59	39.0	26.5	51.4
46-65	57/115	49.6	40.4	58.7
66-85	67/124	54.0	45.3	62.8
86-105	12/24	50.0	30.0	70.0
≥106	12/30	40.0	22.5	57.5
not received	6/8	75.0	45.0	105.0
χ^2: 6,781; **gl:** 5; **p:** 0.237.				

Source: own elaboration.

TABLE 2. Seroprevalence of leptospirosis according to the different regions of the department of Huila.

Region	Frequence	Seroprevalence	95% Confidence interval	
			Lower limit	Upper limit
Northern	93/177	52.5	45.2	59.8
Western	29/60	48.3	35.9	61.0
Central	27/58	46.6	33.9	59.6
Southern	28/65	43.1	32.7	54.8
χ^2: 1,947; **gl:** 3; **p:** 0,584.				

Source: own elaboration.

TABLE 3. Contact with other domestic species associated with the seroprevalence of leptospirosis in cattle from the department of Huila

Contact with other species		Frequence	Seroprevalence	χ^2	p	OR ICI	IC OR 95%	
							ICS	
Canines	Yes No	42/85 135/275	49.4 49.1	0.002	0.959	1.01	0.62	1.65
Equines	Yes No	52/101 125/259	51.5 48.3	0.302	0.583	1.14	0.72	1.80
Sheep	Yes No	13/28 164/332	46.4 49.4	0.091	0.763	0.89	0.41	1.92
Birds	Yes No	10/42 167/318	23.8 52.5	12.82	0.001	0.28	0.13	0.59

Source: own elaboration.

Additional clinical and morphophysiological integrity parameters were assessed in the cows, including heart rate, respiratory rate, temperature, body condition, and weight. The integrity of various systems (cardiovascular, respiratory, digestive, reproductive, and integumentary) was also evaluated, along with other body parts. However, no statistically significant differences were observed.

Of the 230 variables analyzed through independent logistic regression models, only 12 were identified as statistically significant, either as protective or risk factors (table 4). A multiple logistic regression model incorporating these significant variables was subsequently developed, resulting in 4 variables that demonstrated statistical significance ($p \leq 0.05$) (table 5).

TABLE 4. Independent variable logistic regression of risk factors with the seroprevalence of leptospirosis

Variables		β	gl	p	OR	IC OR 95%	
						ICI	ICS
Purchase cattle for fattening	No		-	-	-	-	-
	Yes	0.665	1	0.013*	1.94	1.15	3.28
Have defined internal pathways	No			-	-	-	-
	Yes	-1.33	1	0.011*	0.26	0.095	0.73
Storage of concentrate	No			-	-	-	-
	Barrel	0.574	1	0.037*	1.78	1.03	3.05
	Pallet	0.351	1	0.264	1.42	0.77	2.63
	Pallet/ Barrel	2.170	1	0.049*	8.76	1.01	75.77
	Floor	0.27	1	0.942	1.03	0.50	2.11
Weigh on scale	No			-	-	-	-
	Periodically	0.984	1	0.008*	2.67	1.29	5.53
	Sporadically	0.427	1	0.126	1.53	0.89	2.65
Rodent control program	No			-	-	-	-
	Yes	0.523	1	0.016*	1.69	1.10	2.58
Medications stored and classified	No			-	-	-	-
	Yes	0.674	1	0.008*	1.96	1.19	3.22
Quarantine	No			-	-	-	-
	Yes	0.493	1	0.023*	1.64	1.07	2.50
Isolation	No			-	-	-	-
	Yes	0.748	1	0.009*	2.11	1.20	3.72
Heat detection in cows	No			-	-	-	-
	Yes	0.569	1	0.008*	1.77	1.16	2.69
Handwashing and disinfection	No			-	-	-	-
	Yes	0.554	1	0.014*	1.74	1.12	2.70
Prior cleaning of objects	No			-	-	-	-
	Yes	1.188	1	0.000*	3.28	1.71	6.27

Note: * = variable with significance p ≤0.05;–= reference variable

Source: own elaboration.

TABLE 5. Multiple logistic regression analysis of risk factors with the seroprevalence of leptospirosis.

Independent variables	Coefficients β	gl	p	OR	IC OR 95%	
					ICI	ICS
Intercept	0.56	1	0.31	1.76		
Mix cattle production with birds	-1.28	1	0.00	0.28	0.13	0.59
Have defined internal pathways	-1.39	1	0.01	0.25	0.09	0.7
Isolation	0.59	1	0.05	1.8	1	3.3
Heat detection in cows	0.66	1	0.04	1.9	1.2	3.0
Omnibus test p: 0.000			**Nagelkerke R^2:** 0.124			

Source: own elaboration.

The One Health approach emphasizes the interconnectedness of human health, animal health, and ecosystem integrity, advocating for collaborative, multidisciplinary efforts to achieve balance within this complex system (Roberts 2019). Furthermore, many emerging zoonotic diseases originate from wildlife due to ecosystem degradation (Taddei 2021). Leptospirosis, a zoonotic disease, affects humans as well as wild and domesticated animals (Vera *et al.* 2019). However, the epidemiological role of wild animals in disease transmission remains a subject of debate (Pal *et al.* 2021).

Studying the ecosystem dynamics of bovine leptospirosis, including its epidemiological, demographic, and social determinants within specific populations, will enhance public health strategies aimed at reducing human transmission. Additionally, identifying risk factors associated with bovine disease will support the development of prevention and control measures to mitigate its adverse effects on human health.

The prevalence of leptospirosis in the tested individuals was 49.2% (177/360), with *Leptospira* seropositivity analyzed using the ELISA assay. According to the literature, no studies on leptospirosis in cattle have been conducted at the regional level. However, at the national level, research is scarce, and the seroprevalence observed in this study is considerably higher than the average reported. At the international level, several studies have investigated leptospirosis in livestock due to its significant impact. For instance, a study in southern Andaman reported a seroprevalence of 42.15% (180/247), which is similar to the findings of the present study; however, the study used the MAT technique and identified serovars such as *L. hardjo*, *L. australis*, *L. pomona*, and *L. canicola* (Sunder *et al.* 2017). Similarly, in Japan, a study identified 44 (12.8%) dairy cows as seropositive using the TMA technique, with *L. sejroe* being the most prevalent serovar. Additionally, PCR testing of urine culture samples revealed a prevalence of 5.12% (Koizumi & Yasutomi, 2012). In another study conducted in a Nigerian abattoir, a 3.5% seroprevalence was found, with a preference for the *L. hardjo* serovar (Ngbede *et al.* 2012). Furthermore, in Monte Negro, Brazil, the prevalence was 52.8% (1,114/2,109) (Aguiar *et al.* 2006).

At the national level, in the department of Caquetá, the Colombian Cattle Ranching Association (Fedegan, by its acronym in Spanish) analyzed data from several ICA diseases and determined a prevalence of 46% (Fedegan 2010). In Mexico, a study using the ELISA technique found a seroprevalence of 8.58% (17/198) (Andrade *et al.* 2001). Similarly, in Egypt, a 2022 study using the ELISA technique found a seroprevalence of 39.33% (236/600) (Ibrahim *et al.* 2022).

The TMA technique is traditionally considered the gold standard for diagnosing leptospirosis; however, its use is limited to certain laboratories, which complicates its widespread application (Hartskeerl & Smythe 2014). On the other hand, PCR testing detects the presence of the antigen, and protocols have demonstrated its sensitivity (100%) and specificity (99%), making it a reliable, fast, and accurate method (Hernández–Rodríguez *et al.* 2011).

The technique employed in the present study was ELISA, which does not detect the antigen but instead identifies reactive antibodies to *Leptospira* spp. Therefore, the results do not necessarily indicate that the cattle were diseased at the time of sampling. However, the ELISA technique provides valuable information regarding the effectiveness of preventive and biosecurity measures implemented across different cattle herds. A positive result suggests that these measures were breached at some point, leading to infection.

Culturally, the implementation of rodent control programs is often perceived as necessary when there is a high level of infestation. Rodents, the primary reservoir for *Leptospira* spp., are commonly found in production environments. As such, rodent control programs are typically enacted in response to their presence. These animals act as significant vectors of contamination due to their behavior, such as urinating and deliberately walking in various areas. Consequently, the presence of rodents is closely associated with one of the most important risk factors for disease transmission. According to Garoussi *et al.* (2006), because rodents have direct contact with cattle feed, cattle are particularly susceptible to infection (Garoussi *et al.* 2006).

This research also identifies the storage of bovine concentrate in bins as a risk factor, due to the ease with which rodents can access the feed. The inadequate and insecure storage methods allow rodents to infiltrate the concentrate, facilitating the spread of the pathogen. This finding is consistent with Fávero *et al.* (2017), who reported a positive correlation between rodent access to cattle concentrate and the occurrence of leptospirosis (Fávero *et al.* 2017).

Moreover, medicines are at risk of contamination by *Leptospira* spp. through the urine left behind by rodents as they move around the storage areas. As a result, farmers may believe they are storing medicines appropriately without recognizing the influence of rodents on disease transmission (Mwachui *et al.* 2015). In the present study, this variable is also associated with leptospirosis as a risk factor (OR = 1.96), with statistical significance ($p = 0.008$).

In the present investigation, the mixing of poultry with cattle was identified as a protective factor (OR = 0.283), as rodent populations may be specifically concentrated near the poultry houses within the herd (Miño *et al.* 2007). Consequently, the separation of livestock and poultry may reduce the risk of leptospirosis contamination in cattle. However, according to Castillo (2014), the mixing of poultry could also pose a risk factor or potential source of infection, as birds may act as vectors by ingesting infected rodents (Castillo Hernández 2014). Additionally, strains of *Leptospira* spp. that infect cattle have been identified in chickens (Bracken 1955). Despite this, Castillo suggests that birds may help control the disease by preying on rodents, which are the primary reservoir for *Leptospira* spp.

The introduction of external or replacement animals into the herd is considered a risk factor (OR = 1.94), as cattle themselves can act as reservoirs for the disease. This finding aligns with the work of Williams and Windens (2014), who identified the introduction of external cattle as a predisposing factor for the presence of *Leptospira* spp. in herds. Therefore, the concept of a *closed farm*, which enforces strict biosecurity measures to prevent the introduction of external diseases, is essential (Van Schaik *et al.* 2002). In this context, it is also critical to adopt good husbandry practices (GHP), including quarantine protocols and comprehensive diagnostic screening for all diseases prior to the introduction of new cattle.

On the other hand, internal roads were identified as a protective factor. This is due to the organization of cattle herds and their mobility, which reduces contact between cattle and potential reservoirs such as stagnant water (e.g., puddles, floods). Several studies have shown that contaminated water and rainfall are the primary sources of infection in both animals and humans (Barbagelata *et al.* 2013; Romero–Vivas *et al.* 2013). Additionally, water helps maintain the viability of the antigen over extended periods by preventing desiccation (Andre–Fontaine *et al.* 2015).

Weighing animals is a routine practice on farms, and it is common for farmers not to clean the scale thoroughly between weighing different animals. In this study, this lack of cleaning is considered a risk factor for the transmission of leptospirosis, as animals carrying the antigen can contaminate the scale, facilitating the spread of the disease. Furthermore, when animals are weighed, they are typically arranged sequentially before reaching the scale. This practice increases the likelihood of disease transmission among the animals. According to Loureiro and Lilenbaum (2020), *Leptospira* spp. can form biofilms to enhance their survival, and high concentrations of these bacteria may be present in the vaginal discharge of cows, even in asymptomatic individuals (Loureiro & Lilenbaum 2020). In a similar vein, rectal palpation and/or ultrasonography are commonly used to verify estrus. However, if these procedures are not performed properly and proper sanitation measures are not followed, particularly in the herd at the time of the procedure, poor management practices could contribute to disease spread (Barrett *et al.* 2018).

The implementation of quarantine and/or the separation of sick animals was identified as a risk factor (OR=1.64; OR=2.11, respectively) in the present investigation. This is generally associated with poor management practices and a lack of awareness of the disease by the farmers. Tinoco Rangel (2024) reports that a farm located in the department of Cesar has a sanitary plan that includes preventive measures (quarantine guidelines) as well as control measures, which include instructions for the separation of sick animals, particularly for officially controlled diseases such as brucellosis and tuberculosis (Tinoco Rangel 2024). On the other hand, quarantine measures are intended to reduce the spread of diseases between farms (Becker *et al.* 2020). Santos *et al.* (2019) analyzed risk factors associated with leptospirosis in pigs and found that quarantine had no significant effect

(P=0.117) (Santos *et al.* 2019). However, in farms where quarantine was implemented, the disease prevalence was 44.3% (35/79), compared to 54.8% (102/186) in those that did not practice quarantine. In contrast, Valença *et al.* (2013) found that quarantine was a protective factor (OR=0.37) with statistical significance (P=0.004), with prevalence rates of 13.9% and 30.4%, respectively, for farms that did and did not perform quarantine (Valença *et al.* 2013). However, effective quarantine requires obtaining negative results from detection tests, which incurs higher costs and labor, especially when considering the number of animals entering the farm (Van den Brink *et al.* 2023). Similarly, the separation of animals is commonly used as a preventive measure to manage diseases, but it primarily helps improve the signs and symptoms without eliminating the possibility that the animal remains a carrier (Moreira *et al.* 2019).

Finally, to maintain a healthy sanitary environment within the facilities, daily processes are implemented, including the cleaning of objects to be used with the herd, the farm itself, and the respective washing and disinfection of hands. However, statistical analysis reveals these variables as risk factors (OR=3.28; OR=1.74, respectively) with statistical significance. This suggests that the processes associated with these variables are either being performed incorrectly or not at all. For instance, inadequate management of excreta contributes to the spread of diseases, as it serves as a reservoir for *Leptospira* spp. and other pathogens (Contreras–Gómez *et al.* 2017). Additionally, utensils and equipment commonly used in bovine management (e.g., transducers, palpation gloves, intravaginal devices) may not be properly cleaned. Similarly, proper milking practices require hand washing before milking to reduce the risk of bacterial infection (Cerón–Muñoz *et al.* 2015). These findings highlight the need for training in good husbandry practices, with a particular emphasis on adequate hygiene standards for farmers. Although certain variables are expected to prevent disease, they may, in fact, contribute to its occurrence.

CONCLUSIONS

It is crucial to promote and train personnel responsible for herd management to enhance livestock practices, including animal welfare, food quality and safety management, environmental sustainability, and health and disease control, in order to prevent the emergence of diseases such as leptospirosis, which can lead to significant economic losses. The study concluded that the seroprevalence of leptospirosis in the department of Huila is high. However, the primary goal was to identify the key risk factors influencing the dissemination of the pathogen. The presence of cattle in areas frequented by rodents was associated with other risk factors and is considered a major trigger for the disease's onset. Additionally, the design and management of internal pathways are critical for maintaining herd health, as they acted as a protective factor by reducing the likelihood of puddles, which, along with rodents, serve as a reservoir for the disease.

Finally, the factors evaluated in this study are not the only potential triggers; further research is needed to investigate and identify additional risk factors for the occurrence of leptospirosis. Given the lack of alternative solutions for infected individuals and the zoonotic risk posed to public health, prevention remains the most effective mitigation strategy. In this way, progress can be made toward achieving the goals of the One Health approach, as cattle production is directly linked to human consumption and, therefore, plays a vital role in public health.

CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

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DECLARATION OF USE OF ARTIFICIAL INTELLIGENCE

We declare that no artificial intelligence was used in the preparation of this work.

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