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Factors associated with the presence of antibiotic residues in raw milk from cows in the canton of El Carmen, Manabí, Ecuador

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ABSTRACT

Milk, an essential component of human nutrition, can also serve as a vehicle for external contaminants such as antibiotics. To date, no data are available regarding the presence of antibiotic residues in raw milk from El Carmen (Manabí), the largest dairy basin on the Ecuadorian coast. Therefore, the objective of this study was to determine the presence of these residues and the associated risk factors. A total of 126 raw milk samples were collected from 42 dairy farms on three different occasions between September 2022 and January 2023. Of these, 78.57% (99/126) contained antibiotic residues below the maximum limits established by the Codex Alimentarius, whereas 21.43% (27/126) exceeded these limits. Sulfonamides were the most frequently detected residues (15.57%), followed by β-lactams (4.76%) and tetracyclines (0.79%). Among the risk factors analyzed, the frequency of antibiotic use in animals per year had the greatest influence (Odds Ratio: 0.001), compared with other covariates. The findings indicate a high prevalence of antibiotic residues in raw milk from the sampled region, primarily due to the indiscriminate use of these drugs by dairy farmers. Therefore, the role of regulatory agencies is crucial in preventing the commercialization of milk containing these residues. Additionally, training programs for producers are essential to mitigate this form of chemical contamination.

Keywords: antibiotics, raw milk, risk factors, Manabí.

Factores asociados a la presencia de residuos antibióticos en leche cruda de vacas en el cantón El Carmen, Manabí, Ecuador

RESUMEN

La leche, alimento esencial en la nutrición humana, también puede ser el vehículo de contaminantes externos, como los antibióticos. Al respecto, no se posee datos de la presencia de residuos de estos fármacos en la leche cruda del cantón El Carmen (Manabí), la mayor cuenca lechera de la costa ecuatoriana, por lo que el objetivo de la presente investigación fue determinar su presencia y los factores asociados. Se recolectaron 126 muestras de leche cruda de 42 ganaderías, por 3 ocasiones, entre septiembre 2022 y

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enero 2023, de las cuales el 78,57% (99/126) no presentaba presencia de residuos de antibióticos por encima de los límites permitidos por el Codex Alimentarius, frente a un elevado 21,43% (27/126) de muestras que sí presentaron, donde las sulfamidas mostraron mayor presencia (15, 57%), seguido de betalactámicos (4, 76%) y tetraciclinas (0, 79%). Entre los factores de riesgo, el de mayor influencia es la frecuencia al año con la que se usa los medicamentos en animales (Odd Ratio 0, 001), frente a otras covariables. Se concluye que existe alta presencia de residuos de antibióticos en la leche cruda del cantón muestreado, por uso indiscriminado de los ganaderos, por lo que el accionar de los organismos de control es fundamental a fin de impedir que se expenda el producto con estos residuos, así como se hace fundamental la capacitación a los productores para evitar esta contaminación química.

Palabras clave: antibióticos , leche cruda, factores, Manabí.

INTRODUCTION

Agriculture and livestock farming are among the most important economic sectors in Ecuador, ranking fourth and contributing 9.96% to the national gross domestic product (GDP). Dairy production is primarily carried out by small-scale farmers, who account for 80% of total milk production (Contero Callay *et al.* 2021; Ioneta 2022; Terán 2019). In 2022, Ecuador's daily milk production reached 5.5 million liters, with Manabí ranking as the country's fourth-largest milk-producing province, contributing 11.32% of the total. Manabí also represents the primary dairy basin of the coastal region, with El Carmen Canton being the area with the largest bovine population and the highest milk production (INEC-ESPAC 2023; CFN 2022).

One of the most critical challenges in the dairy industry is that raw milk and its derivatives often reach Ecuadorian households without undergoing quality analysis to ensure their suitability for human consumption or further processing (CIL 2015; Guapi Guamán *et al*, 2017; Terán 2019). In Manabí, bovine diseases such as mastitis and lameness are highly prevalent. Previous studies conducted in Chone Canton have reported the presence of various antibiotic residues in raw milk (Andrade & Saldarriaga 2018).

Cow's milk is an essential source of nutrients for consumers due to its high energy, protein, and fat content, which are fundamental for maintaining a healthy, balanced diet. Additionally, dairy farming plays a crucial role in employment generation, particularly for small-scale producers (FAO 2020 & 2023). However, its rich nutritional composition also makes it highly susceptible to contamination by external agents, including pathogenic microorganisms and chemical pollutants, which can lead to the development of diseases that pose health risks to consumers (Brown *et al.* 2020; Sachi *et al.* 2019; Terzi Gulel *et al.* 2020). Among chemical contaminants, antibiotics are of particular concern due to their widespread use in livestock farming for the treatment of bovine diseases. When withdrawal periods are not adequately observed, antibiotic residues can remain in raw milk and its derivatives, leading to significant health risks, such as antimicrobial resistance (Arrieta *et al.* 2019; Özdemir & Tuncer 2020) and hypersensitivity reactions in individuals with allergies (Díaz 2019).

Given the lack of recent data on the presence of antibiotic residues in raw milk from El Carmen Canton, this study aimed to determine the occurrence of these residues and identify the factors associated with their presence in raw milk from this region of Manabí Province, Ecuador.

MATERIALS AND METHODS

Population and sample

This study was a cross-sectional observational study conducted based on convenience sampling. Raw cow's milk samples were collected from various local dairy farms in the canton of El Carmen, Manabí province, specifically in the parishes of San Pedro de Suma, Wilfrido Loor Moreira, El Carmen, La Manga del Cura, La 14, and Santa María. Dairy farms in the region were selected through a census of local raw milk producers who expressed their willingness to participate in the study, with each collected sample representing an observational unit. Sample collection was carried out between September 2022 and January 2023 from 42 dairy farms at three different time points. By the end of the study, a total of 126 samples had been collected. Samples were obtained directly from either the refrigeration tank or storage containers at the farms, ensuring collection occurred immediately after the milking process.

To ensure proper sample collection, personal biosafety measures were observed, and sampling was conducted following the protocol for raw milk sample collection described by Agrocalidad (2020). A sterile ladle specifically adapted for cooling tanks and storage containers was used. The ladle was immersed twice into the container or tank, mixing the contents before drawing the sample from a depth of at least 15 cm below the milk's surface. The collected sample was then transferred into a pre-sterilized 500 mL polyethylene container. A representative sample volume of 30 mL of raw milk was taken, with agitation performed 5 to 6 times prior to sampling. In dairy farms with multiple aluminum or stainless-steel storage containers, the same milk volume was drawn from each container after agitating them 5 to 6 times. These subsamples were then pooled into a single container to ensure sample homogenization.

Sample analysis

After sample collection, the containers were labeled, and the milk was analyzed using a diagnostic test for antibiotic detection. The results were recorded on a control sheet, which included details such as location, sample volume, and time of sampling. Antibiotic detection was performed using the BIOEASY rapid test kit (Shenzhen Bioeasy Biotechnology Co., Ltd., China), a field test designed to detect β-lactams, sulfonamides, and tetracyclines, providing results in approximately nine minutes, following the manufacturer's instructions.

Coagulated milk was not permitted for testing, and tests exposed to inappropriate conditions (e.g., direct sunlight and humidity) were excluded. In cases of positive results, a second confirmatory test was conducted. For result analysis and interpretation, the presence of the upper control line was verified. Tests lacking this control line were deemed invalid and discarded. Upon detecting the control line, the test line signal was compared with the control line sequentially. A negative result was recorded if all lines exhibited uniform coloration. Conversely, if any line remained uncolored or was fainter than the others, the sample was considered positive for that antibiotic family.

Producer surveys

Surveys were conducted among all farm owners and/or individuals responsible for the selected herds to identify factors potentially contributing to the presence of antibiotics in raw milk. The survey included questions regarding the use of antibiotics to treat diseases affecting cattle on farms located in the canton of El Carmen, Manabí. Measurement variables were determined based on instruments designed in a similar study conducted in the Netherlands (Speksnijder *et al.* 2015) and an instrument used to assess antibiotic application in farms located in Bogotá, Colombia (Rodríguez & Acero Plazas 2014). The indicators assessed in these instruments were adapted to meet the research needs of the present study. The survey was validated by three faculty members from the Faculty of Veterinary Medicine at Universidad Central del Ecuador. Additionally, prior to survey administration, informed consent was obtained from the farm owners who participated in the study.

The variables analyzed in this study included the education level of the responsible personnel, age, common diseases affecting the dairy herds, medications used to treat these diseases (specifically antibiotics), dosage, withdrawal period, routes of administration, the individual or professional responsible for diagnosing the disease, and the frequency of medication use.

Statistical analysis

The data obtained from the test results and survey responses were recorded in a Microsoft Excel spreadsheet. Statistical processing was performed using the R Studio software, which was used to organize and classify the data into tables and figures reflecting the study's descriptive results. Frequency and percentage distributions were used to determine the number of cases in which antibiotic residues were detected in raw milk samples collected from the canton of El Carmen, Manabí.

Additionally, risk factor identification was analyzed using a binary logistic regression model to estimate the adjusted odds ratio or by fitting a linear model using weighted least squares with a 95% confidence interval. A bimodal regression was conducted, considering different factors (covariates) that could influence the detection of antibiotic residues in the analyzed raw milk samples (intercept). The covariates in this analysis corresponded to the data obtained from each survey question, while the intercept variable corresponded to the laboratory test results indicating the presence or absence of antibiotic residues. Using the R Studio software, the backward method was applied to eliminate covariates (i.e., risk factors influencing antibiotic residue presence) that had the least impact on the intercept or constant variable.

RESULTS AND DISCUSSION

Results

Presence of antibiotic residues in collected milk

Of the 126 raw milk samples analyzed from the dairy farms, a high proportion (21.43%; 27/126) contained antibiotic residues exceeding the limits established by the Codex Alimentarius, while the remaining 78.57% (99/126) tested negative for residues (table 1). This percentage is considered high, as antibiotic residues should not be present in the final milk supply intended for commercialization due to their implications for public health.

Regarding the types of antibiotics detected, sulfamide residues were found in 15.87% (20/126) of the total samples, β-lactam residues in 4.76% (6/126), and tetracycline residues in 0.79% (1/126) (table 1).

Risk factors

Educational level

The educational level of the farm owners and/or personnel responsible for the sampled farms was predominantly primary education (57.14%), followed by university degree holders (28.57%), and finally, secondary education (14.29%). These data suggest a moderate educational level among the respondents, considering that most have some technical knowledge (table 2).

Age

The age of the study participants (farm owners or workers from the sampled farms) ranged from 32 to 68 years, with a mean age of 44 years. Based on this distribution, participants were categorized into age groups, as detailed in table 3

TABLE 1. Presence of residues and families of antibiotics in raw milk from El Carmen, Manabí

Presence	n	%	Families	n	%
Negative	99	78.57%	Negative	99	78.57%
			Sulfamides	20	15.87%
Positive	27	21.43%	Beta-lactams	6	4.76%
			Tetracyclines	1	0.79%
Total	126	100%	Total	126	100%

Source: own elaboration.

TABLE 2. Educational level of those in charge of the farms

	Frequency	%
Technical	72	57.14%
Professional	36	28.57%
High school	18	14.29%
Total	126	100%

Source: own elaboration.

TABLE 3. Age group of farm managers

	Frequency	%
32 to 50 years	96	76.19%
Above 50 years	30	23.81%
Total	126	100%

Source: own elaboration.

These findings indicate that the personnel working on and managing the farms where samples were collected are relatively young, with 76.19% falling within the 32 to 50-year age range, while 23.81% were in the older age group (above 50 years).

Common diseases in dairy herds

Regarding the most frequently occurring diseases in dairy herds, the results indicated that mastitis was the most prevalent condition (26.19%), followed by infectious respiratory tract diseases with an incidence of 23.81%. Other identified diseases included bacterial infections (19.05%), digestive tract infections, bovine viral diarrhea, and atrophic rhinitis, each with a prevalence of 7.14% (table 4, figure 1).

Antibiotic use in disease treatment

Among the antibiotics used for disease management, tetracyclines were the most frequently administered, detected in 35.71% of the samples. This preference

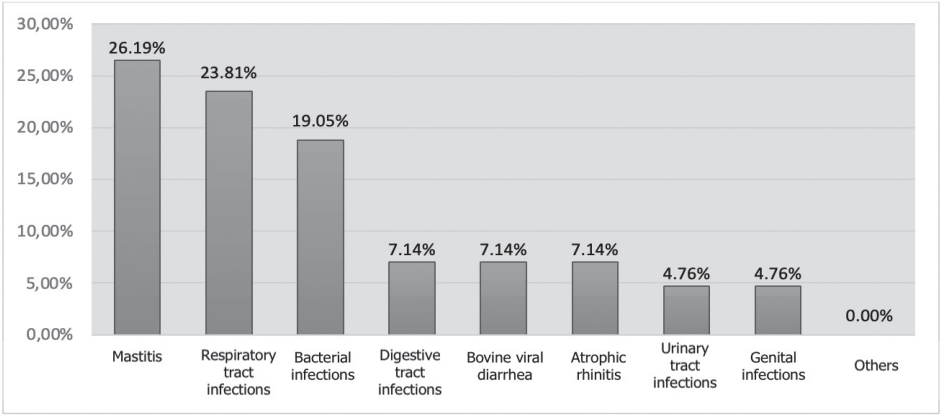

FIGURE 1. Common diseases in dairy herds.

Source: own elaboration.

TABLE 4. Common diseases in dairy herds.

	Frequency	%
Mastitis	33	26.19%
Respiratory tract infections	30	23.81%
Bacterial infections	24	19.05%
Digestive tract infections	9	7.14%
Bovine viral diarrhea	9	7.14%
Atrophic rhinitis	9	7.14%
Urinary tract infections	6	4.76%
Genital infections	6	4.76%
Total	126	100%

Source: own elaboration.

may be attributed to their relatively non-invasive characteristics and broad-spectrum efficacy in addressing multiple conditions affecting dairy herds. β-lactam antibiotics were also commonly used, reported in 35.71% of cases, followed by sulfonamides at 28.57% (table 5).

Dosage and withdrawal period

Respondents indicated that antibiotic administration was generally based on certain herd characteristics, such as weight and age. Specifically, the most commonly reported dosage ranged between 5 and 20 mL per adult cow (64.29% of respondents), while 16.67% reported administering 21–50 mL for more severe conditions, and 19.05% used doses exceeding 50 mL. Notably, some respondents did not adhere to the dosage recommendations provided by the manufacturers, which is a concerning finding.

Additionally, 64.29% of dairy farmers and farm managers reported implementing a withdrawal period of one to five days before selling milk from treated animals, while a smaller proportion observed a waiting period of 5 to 20 days. The antibiotic withdrawal period, as well as the waiting time before milk commercialization, may vary depending on the treated disease and the type of antibiotic used. However, these factors were often overlooked by some producers.

Dosage and withdrawal periods of the medications used

Analysis of the relationship between antibiotic dosage and withdrawal period revealed that 66.67% of dairy farmers adhered to the recommended waiting times before resuming milk production following antibiotic treatment. These respondents indicated that they followed prescribed treatment durations, including waiting at least 15 days after vaccinations or previous treatments before administering additional medication. However, 33.33% of respondents did not comply with these recommendations (table 6).

TABLE 5. Medications used in diseases of dairy herds.

	Frequency	%
Tetracyclines	45	35.71%
Beta-lactams	45	35.71%
Sulfamides	36	28.57%
Total	126	100%

Source: own elaboration.

TABLE 6. Dosage and withdrawal time of medication

	Frequency	%
Within the recommended periods	84	66.67%
Higher than recommended periods	42	33.33%
Total	126	100%

Source: own elaboration.

Route of drug administration

Regarding the administration route of antibiotics, 52.38% (66/126) of respondents reported using the intramuscular route, while 23.81% (30/126) primarily used intravenous administration. Additionally, 14.29% (18/126) applied intramammary administration, and 9.52% (12/126) used subcutaneous injection. The method of antibiotic administration largely depends on the drug type and manufacturer guidelines. However, adherence to these guidelines is critical, as proper administration ensures treatment efficacy and minimizes the risk of antimicrobial resistance.

Disease diagnosis

Concerning disease diagnosis, 64.29% of respondents reported that a veterinarian diagnosed illnesses in their herds, whereas in 35.71% of cases, diagnoses were made by the farm owner or milking personnel. The lack of professional veterinary involvement may lead to inappropriate medication use, including exceeding recommended dosages, prolonging treatment durations, or administering incorrect drugs for specific conditions.

To further investigate antibiotic usage frequency, respondents were asked to estimate the number of times they administered antibiotics to their dairy herds within a year. The results indicated an average of 22 antibiotic applications annually. Moreover, 76.19% of respondents reported administering antibiotics more than ten times per year. This frequency suggests that certain seasons exhibit higher disease incidence, prompting increased antibiotic use to mitigate potential outbreaks, prevent mass infections, and reduce livestock mortality.

Statistical analysis of risk factors

Statistical analysis determined that the frequency of antibiotic use was the only risk factor associated with the presence of antibiotic residues in raw milk from dairy herds.

Initial model including all variables

Initially, all identified variables from the dataset were entered into the R software for logistic regression analysis. This allowed for the identification of residual standard deviation to assess the relationship between two dissimilar variables and predict the probability of an increase or decrease in antibiotic residues in the raw milk samples (table 7):

Table 7 presents a summary of the estimation performed in R, using the appropriate function based on the obtained fit to calculate the residual variance. Upon analyzing table 8, it was observed that none of the variables were statistically significant in predicting the presence or absence of antibiotic residues in raw milk from the 126 samples, indicating a model error. Subsequently, the backward elimination method was applied using R software to gradually remove variables that do not contribute to predicting the presence or absence of antibiotics in raw milk.

TABLE 7. Residual deviation

Min.	1st Qu.	Median	3Q	Max.
-1.461e-06	-1.461e-06	-1.461e-06	-1.461e-06	1.461e-06

Source: own elaboration.

TABLE 8. Variables considered from the database.

	Estimation Std.	Error z	Value
(Intercept)	2.757e+01	1.037e+06	0
Educational level: high school	-1.334e-08	4.833e+05	0
Educational level: technical	-5.177e-11	1.900e+05	0
Years old	3.465e-12	161e+04	0
Diseases: bovine viral diarrhea	-3.747e-10	6.157e+05	0
Diseases: bovine viral diarrhea, mastitis	-6.272e-07	1.119e+06	0
Diseases: bacterial infections	-1.670e-09	5.223e+05	0
Diseases: urinary tract infections	-1.553e-09	6.881e+05	0
Diseases: genital infections	-1.249e-09	5.596e+05	0
Diseases: infections of the digestive tract	-1.188e-09	7.302e+05	0
Diseases: infectious digestive tract, mastitis	-6.379e-07	1.205e+06	0
Diseases: infectious diseases of the respiratory tract	-1.370e-09	5.818e+05	0
Diseases: infectious diseases of the respiratory tract, mastitis	8.919e-06	1.261e+06	0
Diseases: mastitis	-1.742e-09	4.569e+05	0
Diseases: atrophic rhinitis	-1.450e-09	6.999e+05	0
Antibiotics: penicillins	-2.807e-09	9.873e+05	0
Antibiotics: sulfamiridoxines	-3.273e-09	6.973e+05	0
Antibiotics: sulfamiridoxines , Beta-lactams	-8.741e-06	9.850e+05	0
Antibiotics: tetracyclines	-2.507e-09	6.720e+05	NA
Dose: higher	-1.284e-10	5.408e+05	0
Administration: intramuscular	4.423e-10	7.165e+05	0
Administration: intramuscular , intramammary	NA	NA	NA
Administration: intravenous	4.103e-11	2.539e+05	0
Administration: subcutaneous	NA	NA	NA
Diagnostician: owner	1.420e-07	9.599e+05	0
Diagnoser: veterinarian	1.419e-07	9.139e+05	0
Frequency	-1.098e-11	8.041e+03	0
Antibiotic type: none	-5.513e+01	3.246e+05	0
Antibiotic type: sulfonamides	7.204e-07	3.098e+05	0
Antibiotic type: tetracyclines	-8.260e-06	7.890e+05	0
(Dispersion parameter for binomial family taken to be 1)			
Null deviance: 1.3093e+02 on 125 degrees of freedom			
Residual deviance: 2.6892e-10 on 98 degrees of freedom			
AIC: 56			
Number of Fisher Scoring iterations: 26			

Source: own elaboration.

Selecting the best model using the backward method

The backward elimination method was applied using Akaike's Information Criterion (AIC), which enables the identification of variables that should be retained in the model until the optimal model is obtained. Based on AIC, after assessing each variable individually, it was determined that the best model included the *Coefficient, Intercept, and Frequency*. In other words, the covariate *frequency* was the only significant predictor of the presence or absence of antibiotic residues in raw milk.

Odds ratio analysis

The odds estimate the risk of the presence or absence of antibiotics in milk based on the cofactor of interest, such as an increase in the frequency of antibiotic use in dairy animals from the sampled herds. The OR analysis yielded a coefficient of 0.01103499, indicating a negligible effect size (0.011), which suggests an insignificant probability of detecting antibiotic residues in raw milk. This finding aligns with the overall negative results observed in most of the analyzed samples (table 9).

Model significance

The significance of the model was evaluated to determine whether the presence of antibiotic residues in raw milk was statistically meaningful, with the detailed results provided in table 10. The analysis revealed that the significance level for the variable frequency was 16, which falls within the 0.001 threshold, indicating that *frequency* is a statistically significant predictor of antibiotic presence in the milk samples collected from El Carmen, Manabí.

Moreover, the analysis showed that when antibiotic usage frequency reached an approximate or higher average of 90 doses per year, laboratory results were positive for sulfonamides and β-lactams (figure 2). This suggests that increased antibiotic administration is associated with a higher likelihood of detecting residues in raw milk.

TABLE 9. ODDS Ratio to express the possibility of an event

Variable	Coefficient	Odds_Ratio
1 Constant	-4.506684	0.01103499
2 Frequency	0.048392	0.01103499

Source: own elaboration.

TABLE 10. Model meaning

	Df	Residual desviation	Df	Resid. Dev	Pr (>Chi)
Null			125	130.934	
Frequency	1	70.88	124	60.054	<2.2e-16***
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1					

Source: own elaboration.

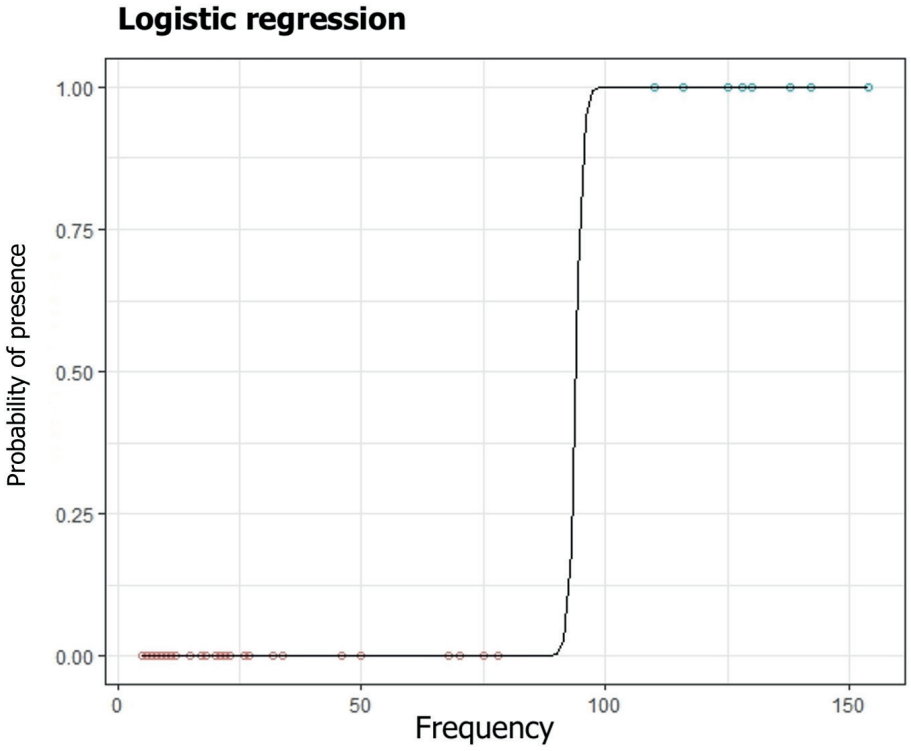

FIGURE 2. Logistic regression.

Source: own elaboration.

Discussion

Following the descriptive and statistical interpretation of the results obtained for raw milk from the canton El Carmen, Manabí, it was identified that 78.57% (99 out of 126 samples) tested negative for antibiotic residues. This finding aligns with reports from Colombia (Madera n.d.), where a study conducted on 40 samples from various farms found that 67.5% also exhibited non detectable antibiotic residues, meeting the national regulatory standards for commercialization and consumption. However, it is important to note that studies on raw milk in Ecuador have shown considerable heterogeneity, with results varying across locations and regions (Puga–Torres *et al.* 2022). For instance, a study conducted in collection centers and small farms in the cantons of Cayambe and Pedro Moncayo (Pichincha, Ecuador) found no antibiotic residues in any of the samples analyzed (Salguero *et al.* 2023).

One notable finding of this study was the prevalent use of sulfonamides (15.87%, 20 out of 126 samples) to treat diseases affecting dairy herds. This result is comparable with another study that reported a 19% prevalence (8 out of 100 samples) for sulfonamide use in dairy herds (Vásquez & Olivera 2012). However, our results do

not align with those of Rey et al. (2020), who found that penicillins were more frequently detected than sulfonamides in raw milk samples previously exposed to antibiotics (Rey *et al.* 2020).

Another key aspect to discuss is the prevalence of diseases in dairy herds, which directly influences antibiotic use. This study identified mastitis (26.19%) and infectious respiratory tract diseases (23.81%) as the most frequent health conditions (33 and 30 out of 126 samples, respectively). These findings are consistent with research conducted on dairy farms, where climatic and viral factors often trigger major disease outbreaks in dairy herds. If not addressed promptly, these conditions can lead to mass infections and high mortality rates, significantly impacting farmers' economic stability and threatening food security (Chiesa *et al.* 2021). Antibiotics are commonly used not only for disease treatment but also for preventive purposes and growth promotion. However, there is substantial scientific evidence indicating that food-producing animals (dairy, meat, and egg producers) serve as reservoirs of antimicrobial-resistant bacteria (Gutiérrez *et al.* 2020).

Regarding associated factors, the variable *frequency of antibiotic use* emerged as the most influential determinant. This finding is consistent with the study conducted by Vásquez and Olivera (2012), which reported that in over 50% of cases, annual antibiotic treatments exceeded 100 applications.

Similarly, our findings align with research conducted in Peru, which also identified mastitis as the most prevalent disease in dairy herds. This study further emphasized that antibiotic treatment for mastitis can lead to detectable residues in milk, as a portion of the administered antibiotics is excreted in the milk during the milking process. Additionally, cows with lower milk production tend to eliminate antibiotics over a longer period, prolonging the presence of residues in milk. The study also highlighted that failure to adhere to prescribed dosages and application times is a key factor contributing to antibiotic residues in raw milk and the development of bacterial resistance (Choque–Quispe *et al.* 2020).

Furthermore, it is crucial to ensure that treatment durations do not exceed recommended limits, as non-compliance with withdrawal periods increases the risk of antibiotic residues in raw milk. In the present study, it was observed that cases testing positive for antibiotic residues involved treatments that exceeded the recommended dosage and withdrawal times. Therefore, strict adherence to prescribed dosages and the minimization of antibiotic application are essential in managing diseases in dairy herds (Choque– Quispe *et al.* 2020; Reyna & Arteaga 2022).

CONCLUSIONS

This study determined the high prevalence of antibiotic residues in raw milk from the sampled canton, primarily due to the indiscriminate use of veterinary antibiotics by dairy farmers. A total of 21.43% of the analyzed samples contained antibiotic residues, with sulfonamides (15.87%) being the most frequently detected, followed by beta-lactams (4.76%). Statistical analysis performed using R software, based on Akaike's Information Criterion (AIC), identified *frequency of antibiotic use* as the most influential factor in the presence of antibiotic residues, compared to other covariables. However, the odds ratio (0.011) indicated that an increase in antibiotic usage frequency could raise the probability of detecting antibiotic residues in raw milk.

It is crucial to provide training to dairy farmers in this key milk-producing region of Ecuador regarding the risks associated with the indiscriminate use of antibiotics in livestock, particularly concerning public health implications when withdrawal periods are not respected. Additionally, governmental intervention is necessary to prevent contaminated milk from reaching consumers.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

ACKNOWLEDGMENTS

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FUNDING DECLARATION

This research was self-funded by the authors.

ETHICAL COMMITTEE APPROVAL

Ethical committee approval was not required for this study, as samples were collected directly from milk containers following the milking process performed by farm personnel. At no point did the researchers collect samples directly from the animals or have direct contact with them. Additionally, the survey conducted did not include personal or confidential questions directed at farm owners or animal caretakers.

DECLARATION ON THE USE OF ARTIFICIAL INTELLIGENCE

No artificial intelligence was used in this study.

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