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STUDY OF PREVALENCE AND CAUSES OF SUB-CLINICAL MASTITIS IN GOATS OF GANDAKI PROVINCE, NEPAL

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Abstract: A study was conducted to determine the prevalence and causes of subclinical mastitis in goats at Goat Research Station (GRS), Bandipur, and National Livestock Breeding Office (NLBO) farm Lampatan, Pokhara. Altogether 61 samples including 43 from GRS and 18 from NLBO were screened for subclinical mastitis using the California Mastitis Test (CMT) reagent, of which positive samples were then aseptically collected in a sterile 10 ml plastic tube and cultured in nutrient agar. Culture-positive samples were Gram-stained to determine their type (gram-positive or gram-negative). In the case of mixed colonies, the culture was sub-cultured. The data were entered and tabular and graphical representation was done in MS-Excel 2010. Results revealed that 26.23% of samples were positive for CMT and of 122 active teats tested, 19 (15.57%) were positive for CMT. Similarly, 23.3% of samples from goats of GRS, and Bandipur were CMT positive whereas 33.3% of samples from goats of NLBO, and Pokhara were positive. On the other hand, the prevalence of subclinical mastitis was 27.27%, 31.03%, and 19.04% for Saanen, Boer, and cross-bred goats, respectively. Considering the samples collected from GRS, 75% were culture-positive of total cultured samples while those from NLBO showed 57.14 % culture positive. In the meantime, overall samples showed 68.42% of total cultured samples to be culture positive. Out of 12 culture-positive samples, 92.30 % (11/12) were gram-positive and only 7.69 % (1/12) were gram-negative.

Keywords: Subclinical Mastitis, California Mastitis Test

I. INTRODUCTION

Background of study

Goat is the major source of income for marginalized people in Nepal and now, with modern techniques, it is attracting big capital investors. Goats traditionally had a strong influence on

the socio-economic life of human populations, especially in rural and less favored regions of the world. In these regions, goat constitutes an important source of proteins by converting different natural resources of lower quality to high-quality protein (Dubeuf et al 2004) (1). Goats utilize household by-products, farm byproducts, and grass and fodder from community forests for the production of meat, milk, and manure. Goats can acclimatize to harsh climatic situations and can be reared in all three geographical regions of the country i.e. Terai, Hills, and Mountains. In Nepal goats are mostly raised for meat and goat milk is rarely consumed. But in recent times rearing goats for milk purposes has flourished with raised knowledge about its nutritive value. With time, rearing goats for milk purposes has been flourishing in Nepal with the introduction of foreign milk-yielding breeds like Saanen. Saanen goats are reared for milk on government farms like Goat research station, Bandipur, Goat Development Farm, and Chitlang.

Mastitis is one of the biggest challenges in the Nepalese dairy industry. The Nepalese dairy industry has seen a recent expansion- from merely cows and buffaloes are considered dairy animals to the inclusion of sheep and goats in the industry. For this reason, some exotic milch goat breeds have been introduced and with them have entered the risk of goat mastitis. Mastitis is known to decrease both quantity and quality of production. The bacterial contamination of milk from the affected animal renders it unfit for human consumption as well as for its offspring and provides a mechanism for the spread of diseases like tuberculosis, sore throat, Q-fever, brucellosis, leptospirosis, etc. and has zoonotic importance (Sharif et al 2009) (2). Worst case scenario is projected by subclinical mastitis. Not only is this form difficult to detect it also lowers production. Subclinical mastitis can only be detected by measures of the milk's cellular content (somatic cells) with all other visible signs of inflammation being absent. An easier method for detection of



this form is the California Mastitis Test (CMT) which helps to detect the increased leucocyte count in milk.

Objectives:

General objectives: To study the prevalence and causes of subclinical mastitis in goats of Goat Research Center, Bandipur, and NLBO farm Lampatan, Pokhara
Specific objectives: To identify major bacteria type (gram-positive or gram-negative) causing mastitis in goats

II. LITERATURE REVIEW

Mastitis is a disease of the mammary gland that negatively influences the quantitative and qualitative parameters of milk. At the same time, the milk from a mastitic gland is a source of infection to both other animals and the consumer and therefore a direct threat to human and animal health. This determines the importance of an early and accurate diagnosis of the various forms of inflammation of the mammary gland in goats (Dimitrov et al 2018) (3). In a study of the status of subclinical mastitis (SCM) in Jamunapari and Barbari goats in Indian organized farms it was found that overall, the prevalence of SCM in the goats was assessed as 19.89% (36/181). The prevalence of SCM in Barbari and Jamunapari goats was found as 24.21% (23/95) and 15.12% (13/86), respectively. (Mishra et al 2018) (4)

In a Study in Pakistan Tandojam, A total of 76 (38%) milk samples were found positive for CMT, those also showed bacterial growth on culture media. Among those 43 (56.58%) were from right quarters whereas the remaining 33 (43.42%) were detected with sub-clinical mastitis from left quarters (Pirzada et al 2016). In research in Tiaret Region (Western Algeria), milk samples were collected from 298 lactating goats in which the Californian Mastitis Test showed a prevalence of 33.9% among the tested goats (Bourabah et al 2013) (6). McDougall et al (2002) (10) reported a prevalence of SCM at 19.0% and a similar result was obtained by Contreras et al (2007) (11) who noticed a prevalence of SCM from 5-30% in goats.

III. METHODS AND METHODOLOGY

Study Area

The study was conducted from December 2018 to February 2019 at GRS Bandipur and NLBO farm, Pokhara.

Sample size

Whole herd sampling was done for the study. A total of 122 milk samples were collected from 61 lactating goats.

Sample Collection

The positive samples from GRS have collected aseptically in the 10 ml sterilized plastic tube which was labeled and then transported to the GRS microbiology laboratory and the samples of NLBO farm Pokhara, were similarly transported to the Livestock Disease Investigation Laboratory Pokhara in an ice box.

Method of test

California Mastitis Test

- Before conducting the CMT test, washing of teats with 70% ethyl alcohol using cotton was done.
- Milk was drawn directly into the CMT paddle from the teats after discarding some initial streaks of milk.
- Left and right teat milk sample was drawn in respective chambers noted L and R. Any physical abnormality present in milk from each teat was observed for its color, consistency, flakes, etc.
- Then the reagent was added to the chamber (1:1 ratio).
- The milk and reagent were rotated by movement of the paddle and the reaction was observed immediately (Chakrabarti A. 2007) (7)
- A change in viscosity indicated an increase in quarter SCC, with the CMT reaction being visually scored by the investigator at 45 seconds after adding the reagent by a 5- point scale as follows: negative, the mixture remains liquid with no evidence of the formation of the precipitate; trace, a slight precipitate evident which tends to disappear with continued movement of the paddle; 1 positive (1), a distinct precipitate but no tendency toward gel formation; 2 positive (2), the mixture thickens immediately with some gel formation, and with motion, the mixtures tend to move in toward the center leaving the bottom of the outer edge of the cup exposed, and out again covering the bottom of the cup if the motion stopped; 3 positives (3), a distinct gel forms which tend to adhere to the bottom of the paddle and a distinct central peak forms during swirling. (Kandeel et al 2018) (8)

Laboratory Examination

Milk samples were maintained at room temperature for 1 hour. Then, they were streaked on a Nutrient agar plate by regular streaking method. Then, those plates were incubated at 37°C for 24 hours. After that, the growth was noted and sub-culture was done if non-uniform colonies were obtained. The plates were reincubated for 24 hours at 37°C. Eventually, the cultural isolates were identified based on colony characteristics and Gram's staining (Bouchard et al 2006) (9).

Analysis

The data collected in the field on the data collection sheet were entered into a Microsoft Excel spreadsheet. Similarly, the data from the test result was also drawn together in the excel sheet. The pictorial and tabular forms of data were made using MS- Excel. To analyze the association SPSS software was run.

IV. RESULT AND DISCUSSION

In this study, of the total of 61 goats, 57 (26.23%) goats were positive for CMT, and of 122 active teats, 19 (15.57%) were positive for CMT. Out of 19 CMT-positive samples, 16 goats samples from 10 goats are found culture-positive (Culture on nutrient agar).



1. Prevalence of Subclinical Mastitis based on location

Table1: Interpretation of result of CMT

location	Sample Size(goats)	CMT positive	Prevalence
GRS Bandipur	43	10	23.3%
NLBO	18	6	33.3%
Overall	61	16	26.23%

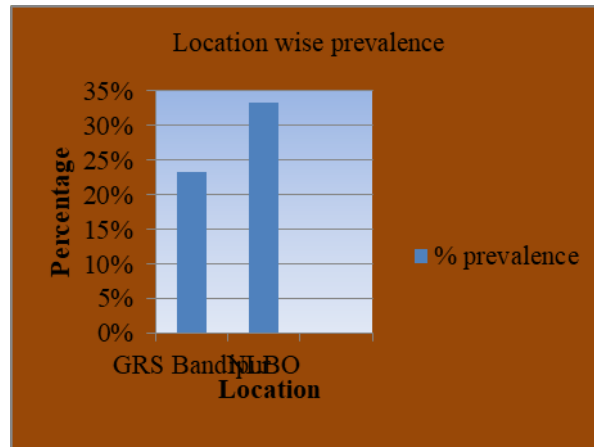


Figure1: Graph presenting prevalence of Subclinical Mastitis based on location

2. Breed wise overall prevalence of subclinical mastitis

Breed	Total goat screened	CMT Positive	Prevalence
Saanen	11	3	27.27 %
Boer	29	9	31.03%
Cross	21	4	19.04%

Table2: Breeds-wise overall prevalence of subclinical mastitis

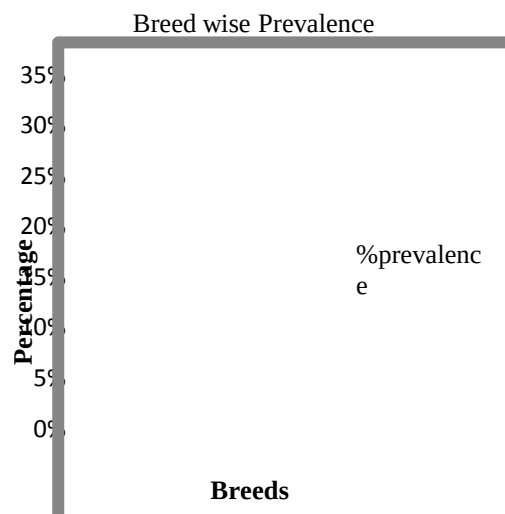


Figure2: Graph presenting breed-wise overall prevalence of subclinical mastitis

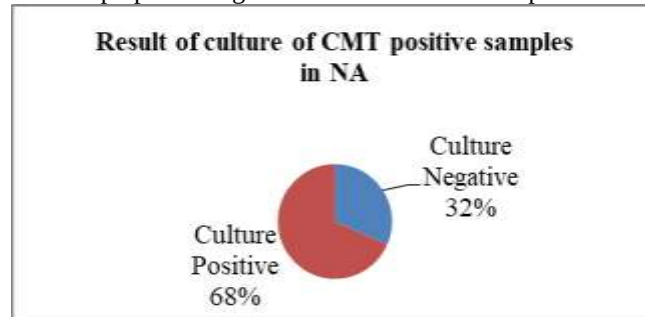


3. **Culture Result of Mastitis positive samples (Culture on nutrient medium):**

Table3: Culture Result of Mastitis positive samples (Culture on nutrient medium)

Location	Cultured Sample	Culture Positive	Percentage evaluation
GRS Bandipur	12	9	75%
NLBO	7	4	57.14%
Overall	19	13	68.42%

Figure1: Graph presenting culture Result of Mastitis positive samples



4. **Result of Gram staining of culture-positive samples:**

Table4: Result of Gram staining of culture-positive samples:

Gram Staining Result (Total 12)	Percentage Evaluation
Gram-Positive	92.30%
Gram-Negative	7.69%

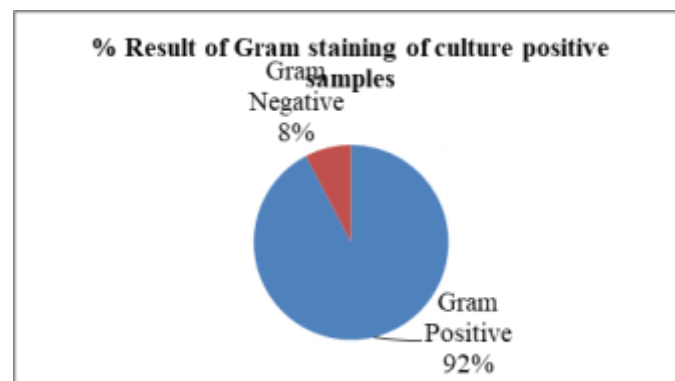


Figure4: Graph presenting the result of Gram staining of culture-positive samples

5. **CMT results based on teats:**

Table5: CMT result based on teats

Teat	CMT positive	Evaluation
Right only	5	31.25%
Left only	8	50%
Both	3	18.75%

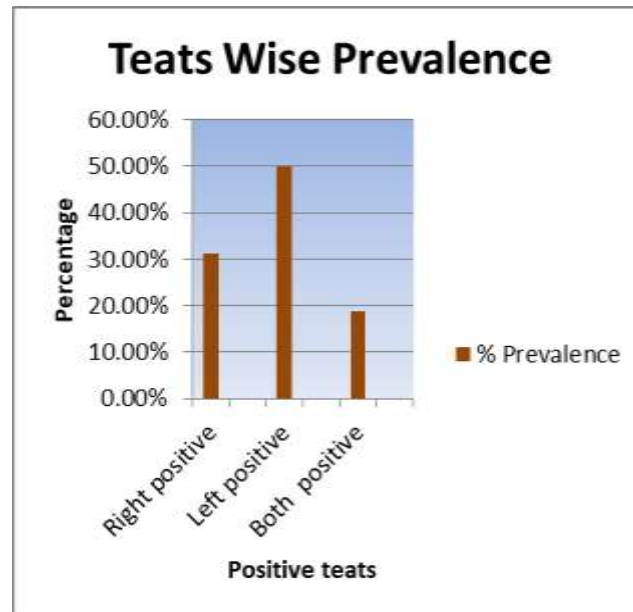


Figure5: Graph presenting CMT result based on teats

In this study, it is revealed that out of a total of 61 goats, 16 (26.23%) were positive for CMT, and of 122 active quarters, 19 (15.57%) were positive for CMT. Out of 19 CMT-positive samples, 13 were found culture positive (Culture on nutrient agar). Location-wise prevalence was 23.3% and 33.3% for GRS Bandipur and NLBO farms respectively. Breed-wise prevalence of SCM mastitis was found to be 27.27 % for Saanen, 31.03 % for Boer, and 19.04 % for cross breeds. 75% of total samples collected from GRS were found to be culture-positive while 57.14 % of samples from NLBO farm were culture positive and overall 68.42 % of the sample was culture positive. Out of 12 culture-positive samples, 92.30% were gram-positive and only 7.69 % were gram-negative.

In this study overall prevalence was found to be 26.23 % (Prevalence of SCM in GRS Bandipur is 23.3 % and that of NLBO farm is 33.3 %) which supports the findings of McDougall et al 2002 and Contreras et al 2007. The Pearson chi-square test (i.e. $p=0.634$) showed the association between the Location Of and prevalence of subclinical mastitis is non-significant. This means that location could not be a determining factor for subclinical mastitis in Government Farm. Breed-wise prevalence suggests Boer has a higher prevalence and crossbreed has the least prevalence of SCM.

V. CONCLUSION AND RECOMMENDATION

Conclusion

From this study, it is found that subclinical mastitis is prevalent in government-organized goat farms. NLBO farm has a higher prevalence rate of SCM than GRS Bandipur. The result obtained shows subclinical mastitis is present even in a well-organized farm which should be minimized soon as these farms are considered as best and managed farms in Nepal.

GRS Bandipur supplies Saanen milk to the consumers and prepares cheese from leftover milk. Inclusion of mastitis-infected milk for distribution to the consumer might act as a source of transfer of animal pathogens to human beings. Statistically, no significant difference in the prevalence of the sub-clinical mastitis in Saanen, Boer, and Crossbreed goats was found as determined by the Chi-square test, probably due to the low sample number. The potential variability in the prevalence among the breeds may be attributed to the difference in genetic resistance, hygiene, milking practices, and management systems along with methods used for the diagnosis of the disease (Amin et al 2011) (12).

Recommendation

- Discard milk from infected goats
- Dipping of all teats following each milking for milking goats. Treatment of all quarter of all goats at drying off should be done
- Regular screening for early detection and treatment should be done as soon as detected
- Udder of all lactating goats should be monitored regularly and treatment should be done if any abnormality and external injury is found
- Infected goats should be separately treated
- Regular sanitation and disinfection of farm.

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