



# Prevalence of Bovine Subclinical Mastitis and Associated Risk Factors at Sylhet District in Bangladesh

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## Authors' contributions

This work was carried out in collaboration among all authors. Authors TZT and MSI designed the study, performed the statistical analysis and wrote the original draft of the manuscript. Author MAL wrote the protocol and first draft of the manuscript. Author DAM and FB managed the literature searches. All authors read and approved the final version of the manuscript.

## Article Information

### Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: <https://www.sdiarticle5.com/review-history/118197>

**Original Research Article**

**Received: 02/04/2024**

**Accepted: 06/06/2024**

**Published: 08/06/2024**

## ABSTRACT

Subclinical mastitis is a prevalent issue in dairy farming, leading to significant economic losses. This study aims to determine the prevalence of subclinical mastitis in the Sylhet district and identify associated risk factors. Understanding these factors is crucial for developing effective control strategies. A total of 114 dairy cows from 22 dairy farms were sampled for this study, and the milk of the cows was screened for SCM using the California Mastitis Test (CMT). Results reveal that the

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**Cite as:** Tanvi, Tanzila Zafrin, Md. Sahidul Islam, Md. Asaduzzaman Lovelu, Dilruba Akter Mir, and Fowzia Bahar. 2024. "Prevalence of Bovine Subclinical Mastitis and Associated Risk Factors at Sylhet District in Bangladesh". *Asian Journal of Research in Animal and Veterinary Sciences* 7 (3):208-15. <https://journalajravs.com/index.php/AJRAVS/article/view/305>.

prevalence of SCM at the individual cow and farm level was 25.43% and 31.81%, respectively. Nevertheless, a chi-square ( $\chi^2$ ) statistical analysis showed that the incidence of SCM was significantly correlated ( $p < 0.05$ ) with variables like milk yield and the educational status of farmers. Contrarily, the factors such as the type of house, adequate drainage facilities, floor type, udder washing before milking, farmer's occupation, professional experience, milking method, breed, age, and lactation stage showed no significant association ( $p > 0.05$ ) with the prevalence of SCM. In conclusion, the results of this study demonstrate the high prevalence of SCM, which poses a serious risk to the productivity performance of the dairy business. To combat the disease, CMT screenings on a regular basis, good hygiene, improved management practices and farmer awareness are recommended in the studied areas.

**Keywords:** Subclinical mastitis; California mastitis test; prevalence; risk factors; Bangladesh.

## 1. INTRODUCTION

The dairy sector of Bangladesh is becoming more and more well-known every day. Currently, 6 million crossbred, high-yielding cows are raised on both commercial and household farms nationwide, producing 9.4 million metric tons of milk annually [1,2]. Dairy cows frequently get mastitis, which causes numerous physical, chemical, pathological and bacterial changes in the glandular tissue and milk [3]. This has had an impact on the dairy industry worldwide, with direct and indirect financial ramifications for farms in the form of decreased milk output and quality, a high rate of culling, subpar productivity, volatile milk prices, costs associated with replacement, treatment and other factors [4,5]. Mastitis is difficult to diagnose, dairy farmers may underestimate these costs [6]. Among bovine diseases, mastitis has a significant negative influence on both the economy and animal welfare [7].

Clinical mastitis can be detected by the history of animals and clinical signs; laboratory tests are required to diagnose subclinical mastitis. Studies show that there is a notably greater incidence of subclinical mastitis (SCM) than clinical mastitis. With a frequency that is 15–40 times higher than that of clinical mastitis, subclinical mastitis is much more prevalent [8]. Most of the dairy farms suffer large financial losses as a result, and the dairy business in Bangladesh is particularly hard hit [9]. According to the California Mastitis Test (CMT), the prevalence of SCM in cows in Bangladesh varies between 20 to 44% [10,11]

Recently, CMT was used in a study to determine whether a patient has subclinical mastitis [12]. According to various research, the prevalence of SCM in crossbred dairy cows in Bangladesh has been reported to range from 28.5% to 61.3%

[13-16]. Unfortunately, little research has been done on SCM in the eastern side of Bangladesh, especially in the district of Sylhet. Cattle's farming is more prevalent in Sylhet district and impacted by several production diseases such as mastitis [17]. Thus, the current study aims to determine the prevalence and risk factors related to sub-clinical mastitis in the Sylhet district of Bangladesh.

## 2. MATERIALS AND METHODS

### 2.1 Study Area and Animals

The study was carried out in the registered medium to large-sized commercial household dairy farms of Sylhet district in Bangladesh during the period from January 2021 to October 2021. In order to investigate the prevalence of SCM at the farm and animal level, 114 dairy cows were selected at random as sample populations from a total of 22 dairy farms. Thus, a detailed list of household dairy farms was created using the data obtained from the Sylhet district livestock office.

### 2.2 Data Collection Procedure

A structured questionnaire was given to the livestock farmers to gather information on the cattle breed (cross/indigenous), age of the cow (<5 years/5 to 7 years/>7 years), production system (intensive, semi-intensive and extensive), floor type (concrete/semi-concrete/muddy), washing of udder before milking (yes/no), lactation stage (early/mid/late), professional experience of farmers (yes/no), milking techniques (hand milking/machine milking), and milk yield (<4 liters /4 to 10 liters/>10 liters). Farmers' approval was obtained prior to data collection and every questionnaire question lasted for about 15 minutes. Interviews with the farm owners and, in certain situations, an

abstraction of the farm records were used to collect data.

### 2.3 Detection of Subclinical Mastitis by California Mastitis Test (CMT)

Firstly, 2 ml of milk was manually collected in each cup while the paddle was held almost horizontally. Next, a polyethylene wash bottle was used to dispense an approximate equivalent volume of CMT reagent, which is made up of 3% sodium hydroxide, 1.5% bromocresol purple, and 1.5% alkyl acryl sulfonate. The paddle was used to mix the ingredients gently for a short period in a horizontal plane. The high somatic cell concentration in the milk caused the response to occur relatively instantly. Within ten seconds, the reaction peaked and was scored. Before being utilized for the following test, the paddle was thoroughly cleaned with water. Depending on the strength of the response, the CMT test findings were categorized as either positive or negative. Gel formation was used to score and record the CMT outcome [18]. Based on the physical examination of the udder, the type and appearance of milk output, and the animal's response to CMT, SCM was identified [19].

### 2.4 Data Analysis

The data collected from the questionnaire survey and CMT analysis were loaded into Microsoft Excel 2010, a spreadsheet tool, for data summarization and analysis. The prevalence of SCM was calculated using the Microsoft Office Excel worksheet 2010. The prevalence was determined by dividing the number of positive samples by the total number of samples tested. In order to evaluate the significant influence of parameters, the data were additionally examined using Pearson's Chi-square ( $\chi^2$ ) test with Statistical Package for Social Sciences (SPSS) version 25 (SPSS Inc., USA). A p-value ( $p < 0.05$ ) was regarded as statistically significant.

## 3. RESULTS

### 3.1 Overall Prevalence of SCM at Sylhet District in Bangladesh

In this study, a total of 114 dairy cattle were examined using the California mastitis test, out of which 25.43% ( $n = 29/114$ ) were recorded as positive for mastitis. Out of 22 farms, 7 farms

were affected with mastitis which was 31.81% ( $n=7/22$ ) of the total farms (Table 1).

### 3.2 Household Dairy Farm Level Prevalence of SCM at Sylhet District in Bangladesh

According to this study, the prevalence in extended, semi-intensive, and intensive housing systems was determined to be 36.36%, 28.57%, and 25%, respectively, the difference was not statistically significant ( $p > 0.05$ ). In the house with a proper drainage system, SCM was recorded at 33.33%, whereas in the house with a defective drainage system, it was recorded at 28.57%. The occurrence of SCM was found to be the highest at concrete flooring (41.67%) followed by semi-concrete and muddy floor with 25% and 16.67%, respectively. It was observed that the prevalence of SCM noted insignificantly ( $p > 0.05$ ) higher (35.29%) on farms where the udder was cleaned prior to milking, while on farms where this practice was not followed, the prevalence was 20%. The farms with the highest frequency (35.71%) of SCM were operated by farmers who primarily depended on animal husbandry for their livelihood, whereas farms with farmers who considered animal husbandry as a subsidiary occupation had the lowest prevalence (25%). Compared to farmers without experience (44.44%), those with professional experience had a lower frequency of SCM (23.08%). There is a significant association ( $p < 0.05$ ) between the educational status of farmers and the prevalence of SCM. The percentage of SCM in illiterate, SSC, HSC and graduate farmers were recorded at 44.44%, 25%, 33.33% and 0%, respectively. The farms with hand milking had the insignificantly ( $p > 0.05$ ) highest frequency (38.46%), whereas farms with machine milking had the lowest prevalence (22.22%) of SCM (Table 2).

### 3.3 Individual Cow Level Prevalence of SCM at Sylhet District in Bangladesh

The prevalence of SCM varies according to breed, age, lactation stage and milk yield, as Table 3 illustrates. The crossbred cows had an insignificantly ( $p > 0.05$ ) higher frequency of SCM (26.14%) in the area compared to native cows (23.08%). There were three observed prevalence rates of SCM: 22.92% (5 to 7 years), 28.57% (>7 years), and 25.81% (<5 years). Mastitis is more common in cases of early lactation (33.96%) than in cases of mid- and late-lactation (19.15%)

and 14.29%, respectively. Furthermore, a statistically significant ( $p < 0.05$ ) correlation exists between milk yield and prevalence. The highest percentage (36.84%) of SCM was observed in

cows generating 4 to 10 liters of milk per day, followed by 33.33% and 16.39% in cows producing more than 10 liters and less than 4 liters of milk per day, respectively.

**Table 1. Overall prevalence of SCM at Sylhet district in Bangladesh**

Level	Number of Tested (N)	Number of Affected (n)	Prevalence (%)
Farm	22	07	31.81
Individual	114	29	25.43

(N) = Number of Tested Farm / Animal, (n) = Number of Affected Farm / Animal, (%) = Percentage of Prevalence

**Table 2. Household dairy farm level prevalence of SCM at Sylhet district in Bangladesh**

Risk Factor	Categories	Farms Tested		SCM Positive		$\chi^2$	p-Value
		N	%	n	%		
Type of housing	Intensive	11	50.00	04	36.36	0.224	0.894
	Semi intensive	07	31.82	02	28.57		
	Extensive	04	18.18	01	25.00		
Proper drainage facilities	Present	15	68.18	05	33.33	1.455	0.228
	Absent	07	31.82	02	28.57		
Type of floor	Muddy	04	18.18	01	25.00	0.672	0.715
	Concrete	12	54.55	05	41.67		
	Semi concrete	06	27.27	01	16.67		
Udder washing before milking	Yes	17	77.27	06	35.29	0.417	0.519
	No	05	22.73	01	20.00		
Farmer's occupation	Dairy farming	14	63.64	05	35.71	0.604	0.490
	Farming and business	08	36.36	02	25.00		
Professional experience	Yes	13	59.09	03	23.08	1.119	0.290
	No	09	40.91	04	44.44		
Educational status	Illiterate	04	18.18	04	44.44	11.757	0.003
	SSC	09	40.91	01	25.00		
	HSC	06	27.27	02	33.33		
	Graduate	03	13.64	00	0.00		
Milking methods	Hand milking	13	59.09	05	38.46	0.269	0.604
	Machine milking	09	40.91	02	22.22		

(N) = Number of Tested Farm / Animal, (n) = Number of Affected Farm / Animal, (%) = Percentage of Prevalence, (SSC) = Secondary School Certificate, (HSC) = Higher Secondary Certificate, ( $\chi^2$ ) = Chi Square Value

**Table 3. Individual cow level prevalence of SCM at Sylhet district in Bangladesh**

Risk factor	Categories	Sample Tested		SCM Positive		$\chi^2$	p-Value
		N	%	N	%		
Breed	Indigenous	26	22.81	06	23.08	0.099	0.753
	Cross	88	77.19	23	26.14		
Age	<5 years	31	27.19	08	25.81	0.344	0.842
	5 to 7 years	48	42.11	11	22.92		
	>7 years	35	30.70	10	28.57		
Stage of lactation	Early	53	46.49	18	33.96	3.679	0.159
	Mid	47	41.23	09	19.15		
	Late	14	12.28	02	14.29		
Milk yield	<4 liters	61	53.51	10	16.39	6.085	0.048
	4 to 10 liters	38	33.33	14	36.84		
	>10 liters	15	13.16	05	33.33		

(N) = Number of Sample Tested, (n) = Number of SCM Positive Sample, (%) = Percentage, ( $\chi^2$ ) = Chi Square Value

#### 4. DISCUSSION

The prevalence of subclinical mastitis 25.43% obtained in this study is lower than the 51.3% reported by Rahman et al. [20] in cows using a modified white slide test. Sayeed et al. [17] stated that around 71.9% farm of the Jhenaidah district was affected by SCM. The overall prevalence of SCM at Bijoy Nagar upazila under the Brahmanbaria district of Bangladesh was 28.75% which was almost similar to our findings [21]. On a global scale, the current study found that the prevalence of SCM is lower compared to studies conducted in Nigeria (85.3%) [22], Uganda (86.2) [23], and Vietnam (88.6%) [24]. On the other hand, the estimated prevalence of SCM is more than that found in the studies conducted in Ethiopia [25] and Rwanda [26]. The disparity in the prevalence of subclinical mastitis in cows across various studies conducted within the country and worldwide can be attributed to differences in geographical location, meteorological conditions, farm makeup, and overall husbandry practices [27].

According to Islam et al. [28], the prevalence of SCM in local breed cows was 22.22%, 27.94%, 21.21%, and 18.18%, respectively, for the age groups of 3 to 5 years, >5 to 8 years, >8 to 12 years, and >12 years. Simultaneously, the age groups of 3 years to 5 years, >5 years to 8 years, >8 years to 12 years, and >12 years in cross-breed cows showed 33.33%, 40.90%, 28.57%, and 0.00% frequency of SCM, respectively. Our study is more or less supported by these results. The high incidence of SCM in the initial phase of lactation can be attributed to the susceptibility of high-yielding cows with larger udders to infection and inflammation. This observation confirms the findings of Pal and Verma [29], who reported that there is a higher occurrence of mastitis in cows producing up to 9 kg of milk, and a lower occurrence of the disease in later stages of lactation (after five months).

From the results of this investigation, there was a tendency for the prevalence of SCM to decline from early (45.83%) to mid (31.58%), with a greater incidence in the late stage than in the mid (33.33%) stage [28]. However, Rahman et al. [30] found that the highest occurrence of subclinical mastitis is during the third month of lactation (34%), while the lowest occurrence is during lactation periods of five months or longer (2%). Islam et al. [12] and Sinha et al. [31] found a greater occurrence of subclinical mastitis

during the later stages of lactation in Bangladesh.

Sohidullah et al. [32] and Almaw et al. [33] showed a higher prevalence of SCM in cross-breed dairy cows, in an investigation of the prevalence of the condition based on breed type. Our results were nearly identical to those of these investigations.

Moreover, the prevalence of SCM in local breed was found to be 13.04%, 20.75%, 30.95%, 37.5% and 50%; in cross breed, the corresponding rates were 28.57%, 35.48%, 40.00%, and 42.85% in >1-2 liters, >2-5 liters, >5-10 liters, and >10 liters daily milk producing cows respectively [28]. This finding is similar to the results of our study.

The type and cleanliness of the floor are crucial risk factors for the occurrence of SCM. Previous research has shown that cemented floors and their cleanliness play a significant role in increasing the risk of SCM elsewhere [34,32]. Nearly the same results were also seen in our investigation [35-37].

#### 5. CONCLUSION

It can be concluded that subclinical mastitis poses a significant obstacle to the advancement of dairy farming in Bangladesh. At the farm and individual animal levels, the prevalence of SCM is determined to be 31.81%, and 25.43%, respectively. It is necessary to control subclinical mastitis in its early stages to prevent its advancement to clinical mastitis. Thus, using CMT is an affordable and practical method for routinely checking subclinical mastitis at field level. Farmers should be aware of the economic impact of the disease and take steps to improve care and management practices.

#### ACKNOWLEDGEMENTS

The Department of Poultry Science at the Faculty of Veterinary, Animal and Biomedical Sciences, Khulna Agricultural University, Khulna-9100, Bangladesh provided support for this study.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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