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Prevalence and consequences of bovine subclinical mastitis in hill tract areas of the Chattogram division, Bangladesh

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ABSTRACT

Mastitis is a common problem in Bangladeshi dairy cattle production. This study investigated the prevalence of subclinical mastitis (SCM), its impact on milk production and reproduction, and the resultant economic losses among dairy farms in the Chattogram division, Bangladesh. A cross-sectional study evaluated 161 farms, collecting 4,048 quarter milk samples from 1,012 cows to conduct a primary screening for SCM using the California mastitis test (CMT). Additional surveys were conducted on SCM-positive farms and animals to explore production and economic impacts. The prevalence of SCM at the farm level was 76.8% (43/56) in Chattogram and 69.5% (73/105) in Cox's Bazar. At the quarter level, the prevalence of SCM was found to be 32.29% (1307/4048; 95% CI: 30.9-33.8), while at the animal level, it was 41.3% (418/1012; 95% CI: 38.3-44.4). Crossbred cows exhibited a higher susceptibility to SCM at 47.2%, compared to indigenous breeds (31.2%). SCM significantly (p<0.05) altered milk's physicochemical properties, reducing fat, protein, and mineral content and negatively affecting milk quality, and market value along with some key reproductive parameters, particularly in Crossbred and Holstein Friesian cows. The economic losses due to SCM are substantial, primarily driven by decreased milk production and increased treatment costs. Crossbred and Holstein Friesian cows suffer the most significant financial losses from their higher milk production and treatment expenses. In conclusion, this study provided valuable insights for policymakers, veterinarians, and farmers, to adopt effective SCM control measures to improve the productivity of the dairy industry in Bangladesh.

INTRODUCTION

Dairy cattle production is vital to the global agricultural economy, providing essential nutrition and supporting millions of livelihoods. This industry stimulates economic activity in related sectors [1]. However, a significant threat to maximizing profits in milk production is mastitis, a prevalent disease in dairy cattle worldwide [2]. Mastitis, mammary gland inflammation, is one of the most common and costly diseases affecting dairy cattle worldwide [3]. It can be caused by a variety of pathogens, including bacteria, fungi, and viruses [4], leading to significant economic losses due to reduced milk yield, altered milk composition, increased veterinary costs, and premature culling of affected animals [5,6].

Mastitis represents a widespread problem in Bangladesh dairy cattle farming, manifesting in clinical (CM) and subclinical (SCM) forms, each markedly affecting the dairy sector. CM is identified by overt symptoms, including udder inflammation, erythema, increased temperature, pain, and alterations in milk appearance. SCM, however, is more common and economically damaging, occurring 15-40 times more frequently than CM [4,7–9]. This disease condition is asymptomatic but can be identified by increased somatic cell counts (SCC) in the milk, signifying an ongoing inflammatory response [10]. The prevalence of mastitis, particularly SCM, varies significantly across different regions and herd management practices, yet it consistently challenges dairy producers worldwide [11]. Often undetected without routine screening, SCM represents a considerable covert threat due to its chronic nature and detrimental effects on milk quality and yield [11].

The etiology of SCM is multifaceted, involving microbial virulence and load, treatment protocols, micro-environmental conditions, host characteristics, milking practices, potential vectors, immune response, and nutritional status [12]. Additionally, injuries to the teats or the udder, resulting from physical, chemical, or thermal factors, can also lead to SCM. Animals with SCM may act as reservoirs of infection, posing a risk to other herd members. In tropical climates, dairy animals experience a higher incidence of SCM due to environmental conditions that favor the proliferation of pathogenic microorganisms responsible for the disease [13,14]. SCM significantly affected reproductive parameters, including days open, calving intervals, service per conception, and conception rates. It primarily extended the duration of these parameters, leading to substantial economic losses for farmers [15]. SCM contributes to approximately two-thirds of the total economic losses in milk production [13]. Consequently, the routine application of on-site diagnostic tests is highly advantageous for the timely detection and management of SCM [16]. However, SCM imposes significant economic burdens on dairy farms through direct and indirect costs. Direct costs include expenditures for veterinary services, diagnostic testing, drugs, and labor, with treatment-related expenses such as veterinary visits ($50 per visit), medications ($100-$200 per case), and labor ($10-$20 per case), along with milk disposal losses due to antibiotic residues ($20-$50 per treatment) [17,18]. Indirect costs involve a 15-20% reduction in milk yield, resulting in an approximate annual loss of $110 per cow, and the prolonged effects of SCM, which incur additional costs of $200-$300 annually per cow [19,20]. Moreover, severe SCM can lead to early culling, imposing significant replacement costs of $1,200-$2,000 per cow, further impacting herd productivity [18].

Apart from the substantial economic losses associated with SCM, it has serious zoonotic potential and has been associated with the increasing development and the rapid emergence of multi-drug resistance strains globally [21,22]. The lack of proper identification of the causative agents of mastitis has contributed to the indiscriminate use of antibiotics, which accelerates the development and spread of MDR strains [23]. The dissemination of MDR pathogens can occur through multiple pathways, including poor hygiene practices, inadequate management, and transmission via milkers. Infected animals' shed resistant pathogens into their environment, contaminating milking equipment, bedding, and other surfaces, thereby increasing the risk of infection transmission within the herd [24]. Consequently, cases of antimicrobial-resistant SCM pose challenges in effective treatment, as these bacteria may not respond to standard antibiotic therapies, resulting in prolonged infections, decreased milk production, and economic losses for dairy farmers [25]. Therefore, prompt identification of SCM is crucial to address this issue effectively.

Despite its widespread presence and significant economic repercussions, a substantial gap exists in understanding mastitis prevalence, its effects on production, and its economic impacts in specific regions, particularly in developing countries like Bangladesh. The Chattogram division, a key area for dairy farming, needs comprehensive economic surveys assessing losses from SCM and changes in milk quality. Moreover, the status of SCM and its qualitative effects on milk remains poorly studied in this region. This study aimed to fill these knowledge gaps by thoroughly investigating SCM prevalence, its impact on milk production and quality, and the resultant economic losses among dairy farmers in Chattogram and Cox Bazar districts. This understanding is critical for developing targeted mastitis management strategies suited to local conditions, ultimately enhancing the financial viability and sustainability of dairy farming in the area.

MATERIALS AND METHODS

Ethical consideration

The study protocol was reviewed and approved by the Animal Experimentation and Ethics Committee (AEEC) of Sylhet Agricultural University under protocol number AUP#2022037.

Experimental design and location

The study was conducted in the hill tract areas of the Chattogram division, Bangladesh. The selected upazilas included Patia, Chandanaish, Satkania, and Lohagara from the Chattogram district, as well as Chakoria, Moheshkhali, Ramu, Ukhia, Teknaf, and Cox's Bazar Sadar from the Cox's Bazar district (Figure 1). The geographical coordinates of the study area are 22°20′06″N to 91°49′57″E (Chattogram) and 21°25′38″N to 92°00′18″E (Cox's Bazar). The study population required to estimate prevalence was calculated using an equation delineated by Naser *et al.* and Rahman *et al.* [26,27].

$$n = \frac{Z^2 \times Pexp \times (1 - Pexp)}{d^2}$$

Where n = Desired sample size, Z = 1.96 for a 95% confidence interval, and P_{exp} = 0.3595 (35.95%). Expected prevalence was considered from previously published work at Chattogram [28]; d = 0.05, Desired absolute precision (5%).

Based on the reference prevalence, the minimum required sample size was calculated to be 353.8. However, this study included 1,012 cows. From June to December 2023, a cross-sectional study was conducted on 161 dairy farms, where 4,048 quarter milk samples were collected for primary screening of SCM using the California Mastitis Test (CMT). The study included farms with varying herd sizes and management practices that had experienced cases of SCM in their cattle and met the inclusion criteria.

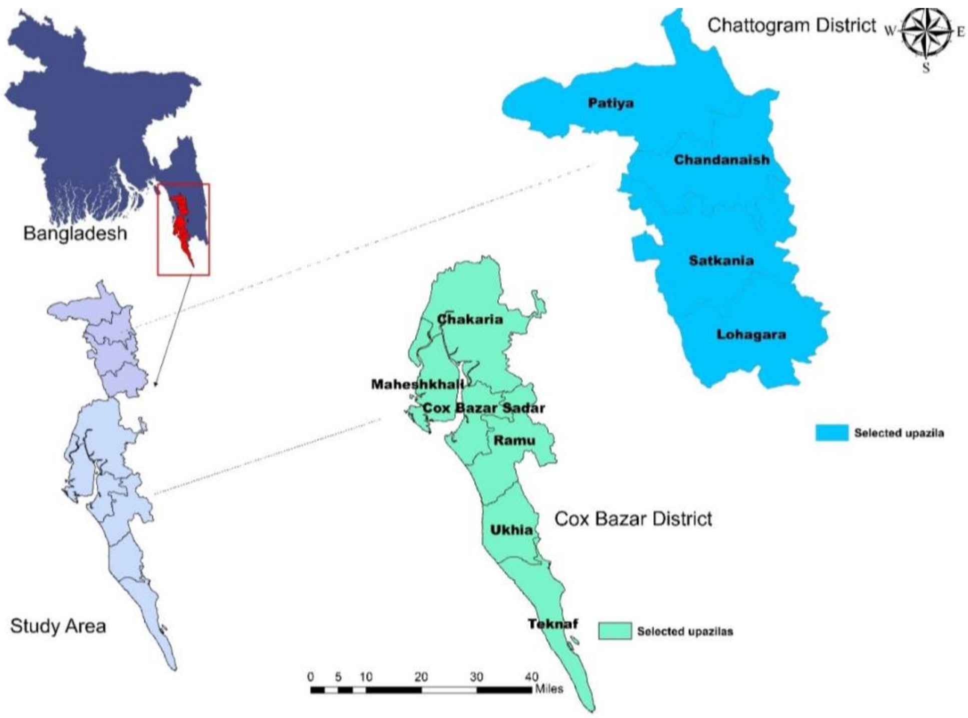

Figure 1. The study area map shows the selected district and sub-district of the experiment. The map was created using ArcMap 10.8.

Disease screening using CMT

The initial selection of cows involved a screening process using the CMT, following the manufacturer's instructions. Approximately 2 ml of milk was aseptically collected from each udder quarter, following the guidelines of the NMC protocols [29], and placed onto a CMT paddle. According to the manufacturer's instructions, an equivalent quantity of CMT reagent (Weizur CMT Test Kit, Gujarat, India) was applied to each well of the paddle. The "Eight-Not" method was applied gently for 15 seconds to achieve a homogeneous mixture. The resulting reaction was assessed using eye estimation, focusing on the formation of gel considering SCM positive and graded based on gel formation. The grading system ranged from negative (Grade 0), where no change in viscosity occurred and the milk remained liquid, to strong (Grade 3+), which was characterized by a very thick consistency and pronounced gel formation. Intermediate grades, such as slight thickening with no gel formation (Graded as Trace), noticeable thickening with slight coagulation (Grade 1+), and thick consistency with pronounced gel formation (Grade 2+), indicated varying levels of thickening and gel formation/ coagulation of SCM positive milk [30,31]. A total of 1012 dairy cows were screened for detection of SCM. Among them 547 cows were Crossbred, 112 were Holstein Friesian and 353 were Indigenous.

Chemical analysis of milk sample

The milk samples, collected by standard guidelines and under aseptic conditions, were transported to the laboratory. The chemical composition analysis of the milk, including parameters such as fat, protein, lactose, solids not fat (SNF), and mineral percentage, was conducted using an auto-milk analyzer through Near Infrared Spectroscopy using DA 7250 NIR Analyzer (PerkinElmer, Inc., Shelton, USA). All the chemical analyses of milk constituents (Physical properties, Chemical constituents such as: Calcium, Phosphorus, Sodium, Potassium, Albumin, Immunoglobulin, α-lactalbumin, β-lactoglobulin and Pre-albumin) and sample preparations were fully followed by the methods described by Gonçalves *et al.* [32].

Data collection

Each selected farm underwent a comprehensive assessment which involved a detailed evaluation of the farm management practices, herd composition, and health status of the cattle. Observations were made regarding the housing, feeding, and milking practices to understand the potential risk for SCM. Structured interviews were conducted with the farm owners or managers using a pre-tested questionnaire (Supplementary File). The questionnaire was designed to gather detailed information on the following aspects: Demographic information (Gender, Age, Education, Farmer type) of the farm owner; Farm management practices; History and prevalence of SCM in the herd; Treatment and control measures for SCM; Economic losses attributed to SCM.

The reproductive data was obtained from the farms' record-keeping repositories. For reproductive data analysis, a minimum of 10 SCM-positive cows and 10 healthy cows from each breed were randomly selected. The animals were chosen from the farms included in this study, ensuring that all selected cows had a minimum of two parties. The interviews were conducted in the local language, and the responses were recorded for subsequent analysis.

Assessment of production and economic loss

Our Research involved a meticulous process of evaluating cows diagnosed with SCM and an equal number of SCM-negative. A total of 836 cows were assessed, with a focus on comparing production loss, length of lactation, and average daily milk production. The cows were selected randomly irrespective of their age and breed, but mostly Crossbred, Indigenous (Deshi), and Holstein Friesian (HF) were observed. This thorough approach ensures the reliability and accuracy of our findings. The effects on production were measured using the following equations:

- Loss of milk production per day due to SCM = Average milk production/day (Normal healthy cow) – Average milk production/day (SCM positive cow)
- Production loss/Lactation (TK) = Loss of Production/Lactation × Unit price (Avg.) of Milk

Economic losses due to SCM were assessed, and the benefits of early diagnosis and treatment were determined through data collection on various economic attributes. To quantify the economic losses associated with SCM, data were gathered on the following parameters: the average price of milk per liter (TK), average loss of milk per day (liter), average days of suffering due to SCM, average days of treatment, treatment cost per day (TK), veterinarian consultation fee (TK), recurrence rate of SCM (Follow up process), decreased value of each recurrence rate (TK), and milk disposal due to antibiotic residues. The collected data were analyzed to estimate the direct and indirect economic losses per farm. The overall economic losses were calculated using a modified version of Jingar's formula [33]:

- Overall loss of Production during sufferings of SCM (TK) = Average price of milk per liter (TK) × Average loss of milk per day (liter) × Average days of suffering due to SCM

- Overall Medication expenditure (TK) = (Average days of treatment × Treatment cost per day (TK)) + Veterinarian consultation fee (TK)
- Loss of value/ Cow = Recurrence rate × Average number of quarters affected/SCM affected cow × Decreased value of recurrence (TK). [Recurrence rate = Number of quarters resulted in recurrent/Number of quarters affected]
- Total Expenditure/Cow during SCM (TK) = (Overall loss of Production during sufferings of SCM (TK) + Overall Medication expenditure (TK) + Loss of value/ Cow + Early diagnostic test fees). [Early diagnostic test fee (1000 TK/Cow) includes Test evaluation fees, Reagent fee, Labor cost, Transportation of evaluator, etc.]

  Finally, the benefits of early diagnosis and treatment of SCM were assessed through the following formula:
- Benefits from each cow per lactation = (Production loss/Cow/Lactation (TK) during undetected and untreated - Grand total Expenditure/Cow during Detected and Treated)

Statistical analysis

The data were entered into a statistical software package for analysis. Descriptive statistics were used to summarize the farm characteristics and management practices. The economic losses were calculated by aggregating the costs. The results were presented in terms of mean losses per farm and the overall economic benefits on the study population. The chi-square goodness of fit test was employed for the socio-demographic profile of the farmer, farm, and also animals. An independent sample t-test was conducted to compare the milk constituents of apparently healthy and SCM-affected cows, as well as to analyze the reproductive data between these two groups. *P* value <0.05 was considered as the level of significance. All the data were analyzed using SPSS version 26 software (IBM SPSS statistics 26). Finally, the data of milk constituents were visualized using GraphPad Prism 8.4.

RESULTS

Socio-demographic characteristics of farm owners

The socio-demographic characteristics of the 116 farm owners who participated in the cross-sectional survey on the economic and production effects of SCM in Chattogram and Cox's Bazar districts, Bangladesh, were presented in Table 1. The demographic profile included variables such as gender, age, education level, and type of farming operation.

The majority of participants were male, comprising 81.9% (95% CI: 73.7-88.4%) of the total sample, with 34 males from Chattogram (CTG) and 61 from Cox's Bazar (CB). Female participants accounted for 18.1% (95% CI: 11.6-26.3%) of the sample, with nine from CTG and twelve from CB. The age of farm owners varied significantly ($P < 0.001$). The largest age group was 46-60 years, comprising 48.3% (95% CI: 38.9-57.7%) of the total participants, with five from CTG and fifty-one from CB. The 31-45 age group represented 31.9% (95% CI: 23.5-41.2%), with 23 from CTG and 14 from CB. Participants aged 61 years and above constituted 10.3% (95% CI: 5.5-17.4%), with seven from CTG and five from CB. The youngest group, 15-30 years old, made up 9.5% (95% CI: 4.8-16.3%) of the sample.

The education levels of the farm owners also varied significantly (P = 0.029). The majority had higher secondary education, accounting for 52.6% (95% CI: 43.6-61.9%) of the sample, with 18 from CTG and 43 from CB. Those with graduation or higher education comprised 31.0% (95% CI: 22.8-40.3%), with 15 from CTG and 21 from CB. Farm owners with secondary education made up 10.3% (95% CI: 5.5-17.4%), with 4 from CTG and 8 from CB. The least educated group with primary education represented 6.0% (95% CI: 2.5-12.0%), with six from CTG and one from CB.

Regarding the type of farming operation, significant differences were observed (*P* < 0.001). Medium-sized farms (11-30 cows) were the most common, representing 48.3% (95% CI: 38.9-57.7%) of the total, with 32 from CTG and 24 from CB. Small farms (less than ten cows) comprised 44.0% (95% CI: 34.8-53.5%) of the sample, with eight from CTG and 43 from CB. Large farms (above 30 cows) were the least common, comprising 7.8% (95% CI: 3.6-14.2%) of the participants.

Table 1. Socio-demographic characteristics of farm owners.

Demographic Characters	Explanatory variable	Participant From CTG	Participant From CB	Total (%)	(95% CI)	*P*-value
Gender						0.54
	Male	34	61	95 (81.9)	73.7-88.4	
	Female	9	12	21 (18.1)	11.6-26.3	
Age (Years)						<0.001
	15-30 Years	8	3	11 (9.5)	4.8-16.3	
	31-45 Years	23	14	37 (31.9)	23.5-41.2	
	46-60 Years	5	51	56 (48.3)	38.9-57.7	
	Above 61 Years	7	5	12 (10.3)	5.5-17.4	
Education						0.029
	Primary	6	1	7 (6.0)	2.5-12.04	
	Secondary	4	8	12 (10.3)	5.5-17.4	
	Higher Secondary	18	43	61 (52.6)	43.61.9	
	Graduation & Higher	15	21	36 (31.0)	22.8-40.3	
Farmer Size						<0.001
	Small (<10 cows)	8	43	51 (44.0)	34.8-53.5	
	Medium (11-30 cows)	32	24	56 (48.3)	38.9-57.7	
	Large (Above 30)	3	6	9 (7.8)	3.6-14.2	

n = 116, regarding the cross-sectional Survey on SCM economic and production effects evaluation in Chattogram, Bangladesh. CI: Confidence Interval, χ^2: Chi-square Goodness of Fit Test, CTG: Chattogram, CB: Cox's Bazar

Prevalence of SCM

The prevalence of SCM was assessed at both farm and animal levels, as detailed in Tables 2 and 3. In the Chattogram district, the overall SCM prevalence in cattle was 76.8% (43/56, 95% CI: 63.6-87.0). The highest prevalence was observed in Chandanaish at 82.4%, while the lowest was in Satkania at 72.2%. In the Cox's Bazar district, SCM prevalence was slightly lower at 69.5% (73/105, 95% CI: 59.8-78.1). The highest prevalence in this district was recorded in Cox's Bazar Upazila at 82.4%, with the lowest in Chokoria Upazila at 55.6%. Farm management practices significantly influenced SCM prevalence (*p*<0.001). Fully intensive management systems had a significantly higher prevalence of 84.5% (87/103), while semi-intensive systems showed a lower incidence of 47.7% (21/44). Regarding flooring types, SCM prevalence was highest on concrete floors at 86.5% (32/37), and lowest on cemented floors at 66.7% (78/117).

At the quarter level, the prevalence of SCM was found to be 32.29% (1307/4048; 95% CI: 30.9-33.8), while at the animal level, it was 41.3% (418/1012; 95% CI: 38.3-44.4). In farm/herd level the prevalence was 72.05% (116/161; 95% CI: 64.4-78.8). The prevalence of SCM varied significantly across different animal-level characteristics (Table 3). Among age groups, the highest prevalence was observed in cows aged 5–8 years 44.9% (282/627), while the lowest was 29.9% (44/147) in those aged above 8 years (p=0.002). Crossbred cows had a higher prevalence of 47.2% (258/547), while indigenous cows showed a prevalence of 31.2% (110/353). For lactation stages, SCM prevalence was highest in the late stage (58.1%; 198/341) and lowest in the early stage (28.0%; 107/382) (p<0.001). Parity also influenced SCM prevalence, with multiparous cows showing a higher prevalence (43.7%; 351/803) compared to primiparous cows (32.1%; 67/209) (p=0.003).

Table 2. Prevalence of SCM on farm-level characteristics.

Characteristics	Explanatory variable	No. of farm tested (N)	No. of positive (x)	Prevalence (%), 95% CI	p- value
Location: Chattogram	Patia	11	8	72.7 (39.0-93.9)	
	Chandanaish	17	14	82.4 (56.6-96.2)	
	Satkania	18	13	72.2 (46.5-90.3)	0.88
	Lohagara	10	8	80.0 (44.4-97.5)	
Cox's Bazar	Chokoria	18	10	55.6 (30.8-78.5)	
	Moheshkhali	19	15	78.9 (54.4-93.9)	
	Ramu	15	11	73.3 (44.9-92.2)	0.40
	Ukhia	17	12	70.6 (44.0-89.7)	
	Teknaf	19	11	57.9 (33.5-79.8)	
	Cox's Bazar	17	14	82.4 (56.6-96.2)	
Farm Management (Feeding)	Intensive	103	87	84.5 (76.0-90.9)	
	Semi-intensive	44	21	47.7 (32.5-63.3)	<0.001
	Open house	14	8	57.1 (28.9-82.3)	
Type of flooring	Concrete	37	32	86.5 (71.2-95.5)	
	Cemented	117	78	66.7 (57.4-75.1)	0.046
	Muddy	7	6	85.7 (42.1-99.6)	
Overall	Herd Level	161	116	72.05 (64.4-78.8)	

CI: Confidence Interval, χ^2: Chi-square Goodness of Fit Test

Table 3. Prevalence of SCM on some animal-level characteristics.

Characteristics	Explanatory variable	Animal tested (N)	No. of positive (x)	Prevalence (%), 95% CI	p-value
Age (Years)					0.002
	3.5-5	238	92	38.7 (32.4-45.2)	
	5-8	627	282	44.9 (41.0-49.0)	
	8 above	147	44	29.9 (22.7-38.0)	
	Total	1012	418	41.3 (38.3-44.4)	
Breed of cows					<0.001
	Crossbred	547	258	47.2 (42.9-51.5)	
	Holstein Friesian	112	50	44.6 (35.2-54.3)	
	Indigenous (Deshi)	353	110	31.2 (26.4-36.3)	
Lactation stage (Days)					<0.001
	Early (<90)	382	107	28.0 (23.0-32.8)	
	Middle (90-180)	289	113	39.1 (33.4-44.9)	
	Late (>180)	341	198	58.1 (52.6-63.4)	
Parity					0.003
	Primiparous	209	67	32.1 (25.8-38.9)	
	Multiparous	803	351	43.7 (40.3-47.2)	

CI: Confidence Interval, χ^2: Chi-square Goodness of Fit Test

CMT grading of SCM-positive samples

The CMT results for SCM-positive milk samples are presented in Table 4 and Figure 2. The majority of samples (67.74%) were graded as negative, showing no viscosity changes. A small percentage showed trace thickening (5.16%) or noticeable thickening with some gel formation (10.30%). More severe grades were also observed, with 9.31% in Grade 2 and 7.51% in Grade 3, indicating increasingly thick consistency and strong gel formation (Figure 2B).

The frequency of SCM-positive quarters revealed that all four quarters were identified in 162 animals, while only one quarter was affected in 29 animals. The majority of SCM-positive cases involved three affected quarters, totaling 176 animals, as shown in Figure 2A.

Table 4. CMT results grading of SCM positive samples.

Grading/Category	CMT findings	Results	%	95% CI
Negative (Zero)	No changes in viscosity, milk remains liquid	2741	67.74	66.3-69.2
Trace	Slight thickening but no gel formation	209	5.16	4.5-5.9
+ (Grade-1)	Noticeable thickening, some gel formation	417	10.30	9.4-11.3
++ (Grade-2)	Thick consistency and pronounced gel formation	377	9.31	8.4-10.3
+++ (Grade-3)	Very thick and strong gel formation	304	7.51	6.7-8.4

CI: Confidence Interval

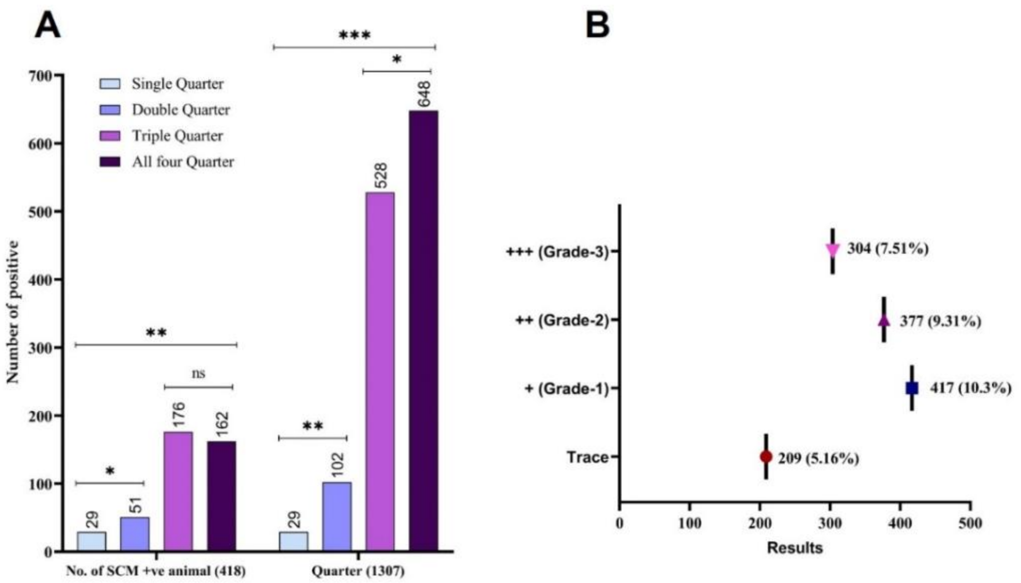

Figure 2. A) Frequency of SCM on CMT positive quarters. Ns: Non-significant, *P<0.05, **P<0.01, ***P<0.001. B) SCM positive results on the basis of CMT grading.

Effect of SCM on physical and chemical properties of milk

The physicochemical properties of milk change significantly due to the effects of SCM in cattle. The chemical components (Fat, SNF, Lactose, Protein, and Minerals) and the protein factions (Immunoglobulin, α-lactalbumin, β-lactoglobulin, albumin and Pre-albumin) and minerals (Calcium, Phosphorous, Sodium, and Potassium) had shown significant deteriorations due to the effects of SCM (Figure 3).

The physical properties of milk from healthy cows showed slight alterations due to SCM (Figure 3A). While pH and freezing point remained relatively stable, significant variations were observed in density, specific gravity, and acidity (%). The density of milk significantly increased (p<0.01) from 30.6 in normal milk to 38.5 in SCM milk. Similarly, the specific gravity was higher in SCM milk than normal milk (1.02 vs 1.04). Additionally, overall milk acidity (%) saw a significant increase (p<0.05) in SCM milk (0.20 vs 0.35).

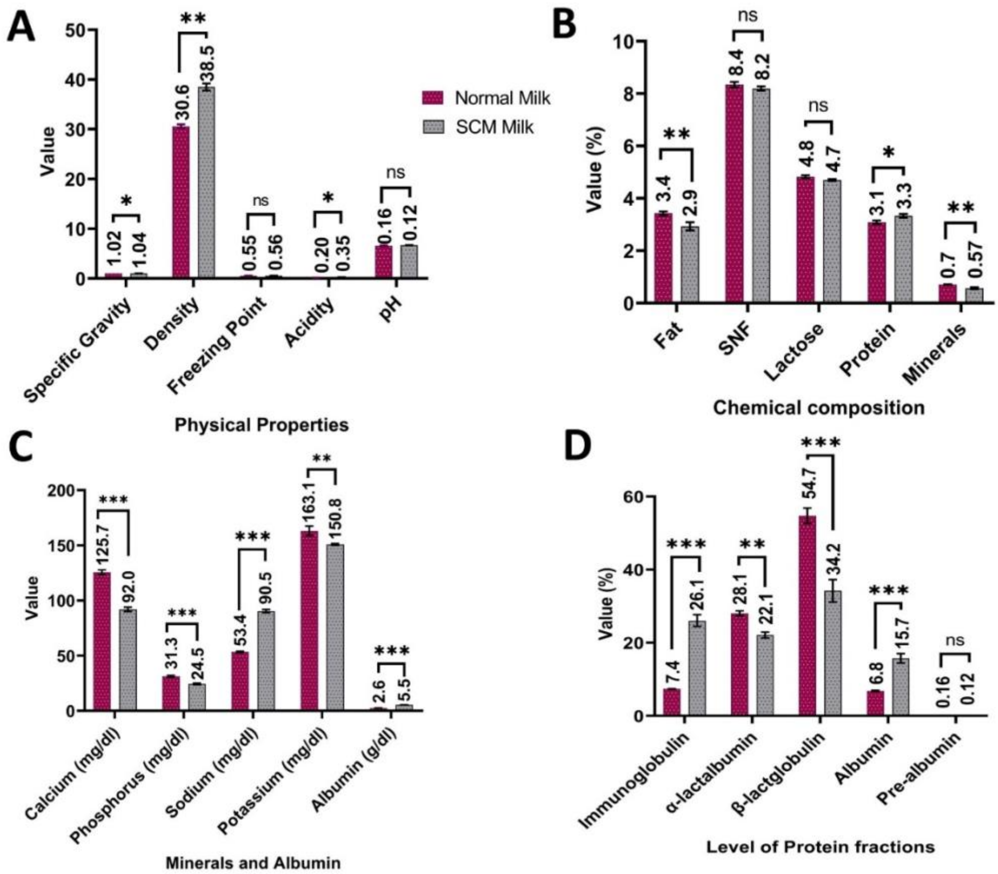

Figure 3. Comparative assessment (Normal vs SCM milk) of milk due to the effects of SCM. A) Changes of physical properties of milk due to SCM compared with normal healthy milk. B) Changes of chemical properties of milk due to SCM. C) Minerals and albumin. D) Level of protein fractions. [ns: non-significant, *p<0.05, **p<0.01, ***p<0.001, Independent sample t-test]

SCM significantly alters the chemical components of milk (Figure 3B). The fat (%) significantly (p<0.01) reduced from 3.4 to 2.9. All other components like Solid not fat (8.2), lactose (4.7), and minerals (0.57) were reduced due to the effect of SCM in cows. The impact of SCM on the mineral and albumin content in milk results in significant alterations compared to normal milk (Figure 3C). The calcium in the normal milk of cows was reduced from 125.7 mg/dl to 92.0 mg/dl while a cow was affected in SCM. Notably, there was a marked decrease (p<0.001) in the phosphorus level, with SCM milk showing 24.5 mg/dl compared to 31.3 mg/dl in normal milk. Conversely, the sodium content in SCM milk rises substantially, registering at 90.5 mg/dl versus 53.4 mg/dl in normal milk. Potassium levels exhibit a slight decrease in SCM milk (150.8 mg/dl) compared to normal milk (163.1 mg/dl). Immunoglobulin levels increase dramatically from 7.4 to 26.1, indicating a higher immune response. α-lactalbumin levels decreased from 28.1 to 22.1, and β-lactoglobulin levels also dropped significantly from 54.7 to 34.2, reflecting disruptions in protein synthesis. Albumin content rises from 6.8 to 15.7, signaling inflammation and tissue damage. Finally, the pre-albumin levels slightly decreased from 0.16 to 0.12 (Figure 3D).

Consequence of SCM on reproductive parameters

The comparative assessment of reproductive parameters between apparently healthy (AH) cows and SCM-positive cows across different breeds was explained in detail in Table 5.

The calving interval was significantly higher in SCM-positive cows (414.5 days) compared to AH cows (398.8 days) for Crossbred and Holstein Friesian breeds (*P* < 0.01), but no significant difference was observed in Indigenous breeds (*P* = 0.16). Similarly, days open were significantly longer in SCM-positive cows of Crossbred and Holstein Friesian breeds (*P* < 0.01), while Indigenous breeds showed no significant difference. Service per conception did not differ significantly (*P* > 0.05) between AH and SCM-positive cows across all breeds. However, conception rates were significantly reduced in SCM-positive Holstein Friesian cows (*P* = 0.04), with no significant differences in Crossbred or Indigenous cows.

Table 5. Comparative assessment of reproductive parameters between AH and SCM-positive cows across different breeds.

Reproductive Parameters	Breeds of Cow	AH cow		SCM (+ve) cow		t-score	*P*-value
		Mean	SEM	Mean	SEM		
Calving Interval (Day)	Crossbred	398.8	2.3	414.5	3.89	3.47	0.003
	Holstein Friesian	388.8	2.30	418.2	3.69	6.74	<0.001
	Indigenous	371.6	4.38	383.7	6.95	1.47	0.16
Days Open (Day)	Crossbred	117.7	1.41	128.1	3.01	3.13	0.006
	Holstein Friesian	108.2	1.26	120.4	3.44	3.33	0.004
	Indigenous	95.1	2.36	101.6	3.37	1.57	0.13
Service Per Conception	Crossbred	2.10	0.23	2.50	0.31	1.04	0.31
	Holstein Friesian	2.30	0.21	2.50	0.27	0.58	0.57
	Indigenous	1.90	0.18	2.20	0.20	1.12	0.28
Conception Rate (%)	Crossbred	58.0	4.16	51.0	2.76	1.40	0.18
	Holstein Friesian	65.0	2.68	56.0	3.06	2.12	0.04
	Indigenous	72.0	4.16	64.0	4.0	1.38	0.18

SEM: Standard Error of Mean, apparently healthy (AH).

Effect of SCM on production loss in different breeds of cattle

Lactation length and production of crossbred cows

SCM significantly impacts milk production across different cattle breeds, leading to substantial economic losses in per-laboration. In crossbred cows, the average lactation period for healthy cows was 236.7 days, but this decreased to 228.9 days in SCM-positive cows, resulting in a loss of 7.8 days. Additionally, milk production per day drops from 9.2 liters in healthy cows to 7.3 liters in SCM-positive cows. Consequently, the overall production per lactation falls from 2177.6 liters to 1670.9 liters, amounting to a production loss of 506.7 liters and a financial loss of 44,893.6 TK ($ 382.17) per lactation (Table 6).

Table 6. Production effects of SCM among the different breeds of cows in Chattogram and Cox's Bazar district of Bangladesh.

Variables	Breed type	Normal cow (A)	SCM positive cows (B)	Loss due to SCM (C= A-B)	Production loss/Lactation (TK)
No. of lactating cow studied		418	418		
(D) Average lactation period (Days; Mean ± SD)	Crossbred	236.7 ± 11.7	228.9 ± 13.9	7.8	
	Holstein Friesian	301.9 ± 9.1	296.1 ± 7.1	5.8	
	Indigenous	209.3 ± 15.0	205.8 ± 9.3	3.5	
(E) Average milk production/ Day (Liter)	Crossbred	9.2 ± 1.5	7.3 ± 1.3	1.9	
	Holstein Friesian	22.5 ± 2.8	18.3 ± 3.6	4.2	
	Indigenous	2.6 ± 0.7	2.3 ± 0.7	0.3	
(F=D×E) Overall production per lactation (Liter)	Crossbred	2177.6	1670.9	506.7	44893.6 ($ 382.17)
	Holstein Friesian	6792.8	5418.6	1374.2	121754.1 ($ 1036.46)
	Indigenous	544.2	473.3	70.9	6281.7 ($ 53.47)

SD: Standard deviation; Conversion rate: 1 TK = 0.00851 USD (Conversion based on 24 August 2024)
Production loss/Lactation (TK) = Loss of Production/Lactation × Unit price (Avg.) of Milk (88.6 TK)

Lactation length and production of Holstein Friesian cows

For Holstein Friesian cows, the average lactation period decreases from 301.9 days in healthy cows to 296.1 days in SCM-positive cows, resulting in a loss of 5.8 days in undetected cows per lactation. Their milk production drops significantly from 22.5 liters per day in healthy cows to 18.3 liters in SCM-positive cows, causing a daily loss of 4.2 liters. This results in a decrease in overall production per lactation from 6792.8 liters to 5418.6 liters, with a production loss of 1374.2 liters and a financial loss of 121,754.1 TK ($ 1036.46) per lactation (Table 6).

Lactation length and production of Indigenous cows

In indigenous cows, the average lactation period for healthy cows was 209.3 days which decreases to 205.8 days in SCM-positive cows, leading to a loss of 3.5 days per lactation. Daily milk production drops slightly from 2.6 liters in healthy cows to 2.3 liters in SCM-positive cows, resulting in a daily loss of 0.3 liters. Overall production per lactation decreases from 544.2 liters to 473.3 liters, with a production loss of 70.9 liters and a financial loss of 6,281.7 TK ($ 53.47) per lactation (Table 6).

Economic losses due to SCM in dairy cows

The economic losses incurred due to SCM in dairy cows in Chattogram and Cox's Bazar districts can be substantial, affecting milk production, treatment costs, and overall cow health. This analysis examines the economic parameters contributing to the total loss and highlights the benefits of early detection and treatment of SCM (Table 7).

Production losses during suffering from SCM

The average price of milk per liter is 88.6 TK. Crossbred cows suffer an average milk loss of 1.9 liters daily for 16.8 days due to SCM, resulting in an overall production loss of 2828.1 TK. Holstein Friesian cows lose 4.2 liters per day for 18.1 days, leading to a higher production loss of 6735.4 TK. Indigenous cows, with a daily milk loss of 0.3 liters for 14.2 days, incur a production loss of 377.4 TK.

Table 7. Loss of economic indices (Due to SCM) in dairy cows of Chattogram and Cox's Bazar.

Sl. No.	Economic parameters	Crossbred (Mean)	Holstein Friesian (Mean)	Indigenous (Mean)
1	Avg. price of milk/Liter (TK)	88.6		
2	Avg. loss of milk/Day (Liter)	1.9	4.2	0.3
3	Avg. days of suffering due to SCM	16.8	18.1	14.2
4	Overall loss of Production during sufferings (TK) {4= (1×2×3)}	2828.1	6735.4	377.4
5	Avg. days of treatment	10.8	12.6	8.2
6	Treatment cost/Day (TK)	87.5	93.1	67.9
7	Veterinarian consultation fee (TK)	1008.3	1253.7	880.7
8	Overall Medication expenditure (TK) {8= (5×6) +7}	1953.3	2426.8	1437.5
9	Recurrence rate	0.07	0.06	0.07
10	Average number of quarter affected/SCM affected cow	1.76	1.62	1.30
11	Decreased value due to recurrence rate (TK)	2381.6	2308.3	1639.7
12	Loss of value/ Cow {12= (9×10×11)}	293.4	224.4	149.2
13	Grand total Expenditure/Cow during SCM (TK) {13= (4+8+12) + Early diagnostic test fees}	6074.8	10386.6	2964.1

Early diagnostic test fee (1000 TK/Cow) includes Test evaluation fees, Reagent fee, Labor cost, Transportation of evaluator, etc.

Treatment costs

Treatment costs also add to the economic burden. Crossbred cows require an average of 10.8 days of treatment at a daily cost of 87.5 TK, plus a veterinarian consultation fee of 1008.3 TK, resulting in an overall medication expenditure of 1953.3 TK. Holstein Friesian cows, with a 12.6-day treatment period, a daily cost of 93.1 TK, and a consultation fee of 1253.7 TK, have a higher treatment cost of 2426.8 TK. Indigenous cows have the lowest treatment cost at 1437.5 TK, with an 8.2-day treatment period at 67.9 TK per day and an 880.7 TK consultation fee.

Loss due to recurrence rate

The recurrence of SCM decreases the value of affected quarters. The recurrence of SCM and the average number of affected quarters per SCM-affected cow were similar across breeds, but the economic impact varies. The decreased value of recurrence was the highest in crossbred cows (2381.6 TK), resulting in a loss of 293.4 TK per cow. Holstein Friesian cows lost 224.4 TK, and indigenous cows incurred the lowest loss at 149.2 TK.

Grand total expenditure

The total expenditure per cow during SCM, including production losses, treatment costs, and recurrence rate of SCM, plus an early diagnostic test fee of 1000 TK ($ 8.51), amounts to 6074.8 TK for crossbred cows, 10386.6 TK for Holstein Friesian cows, and 2964.1 TK for indigenous cows (Table 7).

Benefits of early diagnosis and treatment

The benefits of early detection and treatment of SCM in cows were significant, as illustrated in Table 8. For crossbred cows, the production loss per lactation during undetected and untreated SCM was 44893.6 TK, compared to a total expenditure of 6074.8 TK when SCM was detected and treated, resulting in a benefit of 38818.8 TK ($ 330.348) per cow per lactation. Holstein Friesian cows benefit even more, with a production loss of 121754.1 TK versus an expenditure of 10386.6 TK, yielding a benefit of 111367.5 TK ($ 947.737). Indigenous cows, though having lower overall production losses, still benefit by 3317.6 TK ($ 28.233) per cow per lactation.

Table 8. Economic benefits from early diagnosis and treatment of SCM in cows.

Types of breeds	Production loss/Cow/Lactation (TK) during undetected and untreated (X)	Grand total Expenditure/Cow during Detected and Treated (Y)	Benefits from each cow per lactation (Z = X - Y)
Crossbred	44893.6 ($ 382.045)	6074.8 ($ 51.697)	38818.8 ($ 330.348)
Holstein Friesian	121754.1 ($ 1036.127)	10386.6 ($ 88.389)	111367.5 ($947.737)
Indigenous	6281.7 ($ 53.457)	2964.1 ($ 25.224)	3317.6 ($28.233)

Conversion rate: 1 TK = 0.00851 USD (Conversion based on 24 August 2024)

DISCUSSION

The results of this study highlighted the significant socio-demographic and economic impacts of SCM on dairy farming in the Chattogram and Cox's Bazar districts of Bangladesh. Our analysis revealed substantial differences in SCM prevalence based on farm management practices, cow breeds, and farming operations. We explored the severe production and financial losses associated with SCM, with notable variations in milk constituents across different cow breeds. Furthermore, the study demonstrates the considerable economic benefits of early diagnosis and treatment of SCM, emphasizing the critical need for improved management and detection strategies in the region.

The male participant at the farm ownership level stands at 81.9%, significantly surpassing female ownership, contrary to previous findings indicating a notable rise in female involvement in livestock farming, increasing from 43% to 69% [34]. This surge in female empowerment is largely attributed to substantial support from various NGOs and governmental initiatives to enhance women's societal status. In this study, the largest age group was identified as those between 46-60 years, accounting for 48.3% (95% CI: 38.9-57.7%) of the total participants. This finding aligns closely with research conducted in the Barind area, Dinajpur, and Khagrachari districts of Bangladesh, where the predominant age group was approximately 31-50 years [35,36]. The education levels among the farm owners in this study varied significantly (P = 0.029), with the majority holding higher secondary education, comprising 52.6% (95% CI: 43.6-61.9%) of the total sample, compared to those with graduation or primary education. This result contrasts with findings from other studies in the Barind area and Panchagarh district, where the majority of farm owners had only completed primary education, and in Khagrachari district, where most farm owners were illiterate [35–38]. Educated farmers are more likely to adopt improved management practices, such as regular cleaning and disinfection of milking equipment, proper milking techniques, and timely identification and treatment of mastitis cases. Jeelani *et al.* [39] found a significant association between udder hygiene scores and management practices in dairy farms. Proper udder hygiene and management were linked to lower somatic cell counts, indicating better udder health and reduced incidence of SCM. Additionally, small (Farms having less than 10 cows) and medium (Farms having less than 11-30 cows) sized farms were the most prevalent, comprising 44% and 48.3%, respectively, closely aligning with findings from other studies in Bangladesh [35,36,40].

In this study, the prevalence of SCM in cattle was found to be highest in the Chattogram district, at 76.8% (43/56, 95% CI: 63.6-87.0). This finding closely aligns with a previous study in Chattogram, which reported a prevalence of 70% [41]. Other studies have reported varying prevalence rates: 32.43% and 34.2% in Chattogram, and 53% in Jhenaidah, which do not align with our findings [42–44]. Internationally, SCM prevalence has been reported at 53% in Kenya, 36.4%-50.2% in China, 50% in Colombia, and 62% in Rwanda, showing some variation from our study results [45–48]. Intensive management systems demonstrated a significantly higher prevalence of SCM in dairy cattle at 84.5% compared to semi-intensive farming, consistent with findings from a study in Welimada, Sri Lanka, where intensive farms had a prevalence of 61% [49]. Intensive management system causes an increased frequency of SCM due to increased stress and overcrowding, which can compromise the immune system and create an environment conducive to bacterial growth [50]. Additionally, the frequent and close contact among animals in these systems facilitates the transmission of mastitis-causing pathogens. Concerning flooring types, SCM prevalence was highest on concrete floors at 86.5%, contrasting with another study where soil-type floors were found to increase SCM occurrence in cattle [51]. Additionally, crossbred cows exhibited a higher prevalence of SCM at 47.2% compared to indigenous breeds, aligning with the findings of other studies [52,53].

Our study revealed significant alterations in milk constituents due to SCM in cows from the Chattogram and Cox's Bazar districts of Bangladesh. These alterations encompassed both physico-chemical properties and chemical components of milk. Mammary gland inflammation can lead to alterations in milk composition due to localized effects. Serum components may enter milk, and specific milk constituents typically migrate from the alveolar lumen to the perivascular area leading to changes in milk components [9]. Physically, SCM increased in milk density, specific gravity, and acidity, indicating potential changes in milk composition influenced by inflammatory processes. Chemically, SCM decreased fat, solid-not-fat (SNF), lactose, and mineral contents in milk. The findings of this study are aligned with the findings of another study [54]. The decrease in fat percentage from 3.4% to 2.9%, and reductions in calcium, phosphorus, and potassium levels underscored the metabolic shifts and inflammatory responses associated with SCM. Similar findings were also reported previously [32]. Similarly, a study was conducted in Egypt on Holstein Friesian cows and observed similar kinds of changes due to the effects of SCM [55].

The changes in protein fractions observed were also remarkable, with significant ($P<0.001$) increases in immunoglobulin with concomitant reductions of α-lactalbumin and β-lactoglobulin levels discovered from the present study. The level of immunoglobulins in SCM milk increased significantly (7.4 to 26.1). The increased level is due to the immune response caused by infection [56]. The SCM pathogens provoke an immune response leading to a larger production of immunoglobulins (antibodies), in the bovine milk, following the body’s defense mechanism [57]. In SCM milk, the level of α-lactalbumin decreases to 22.1 in our study. Specifically, regarding proteins, a reduction in the key lactose-synthesis protein alpha-lactalbumin during infection could indicate impaired synthetic activity in the mammary gland [58] . This impairment may lead to lower lactose production and milk quality issues. Our study showed a big drop in beta-lactoglobulin (from 54.7 to only 34.2). As a key whey protein, significantly reduced beta-lactoglobulin expression was observed and may be associated with alterations in milk protein synthesis and secretion pathways during SCM [59]. Inflammation and tissue damage may also influence the secretion of this protein [59,60]

This rise was closely linked to the high albumin concentrations, which are a pro-inflammatory factor targeted at repairing damaged tissue [48, 50]. One of these functions is that albumin works as a carrier protein, it can be upregulated by inflammatory stimuli to help in repairing tissues and immune responses [32,55–57].

These changes reflect the immune system's response to SCM, affecting milk quality and potentially compromising its nutritional value. The elevated albumin content further indicates tissue damage and inflammation within the mammary gland, contributing to overall milk quality deterioration. The slight reduction in pre-albumin further reflects the broader impact of SCM on the protein synthesis machinery of the mammary gland

[32,55]. Our findings aligned with previous research highlighting the detrimental effects of SCM on milk quality. Studies by Goncalves *et al.* [32] and Dufour *et al.* [9] similarly reported reductions in fat and protein contents, alongside changes in mineral composition in SCM-affected milk. These consistent findings emphasize the universal impact of SCM on milk composition across different geographical regions and cattle breeds [32,55–57]. In Crossbred and Holstein Friesian cows, SCM led to considerable negative impacts on key reproductive metrics, as evidenced by increased calving intervals and days open, along with reduced conception rates in Holstein Friesian cows. The significant reduction in milk progesterone levels across all breeds suggests a possible hormonal imbalance associated with SCM, which may contribute to impaired reproductive performance. Indigenous breeds showed relatively minimal effects, highlighting potential breed-specific resilience to SCM-related reproductive challenges. Similar findings were also observed by Waseem *et al.* [15].

The findings of our study showed the significant economic impact of SCM on dairy production, highlighting the need for effective management strategies. Effective measures to address SCM include improving diagnostic techniques, adopting better farm management and hygiene practices, and promoting farmer awareness programs. Additionally, prioritizing studies on antimicrobial resistance, breed susceptibility, nutritional interventions, and sustainable farming practices can significantly enhance disease prevention and control. SCM affects various breeds differently, with Holstein Friesians experiencing the highest production losses, followed by crossbred and indigenous cows [4].

Early diagnosis and treatment of SCM present clear economic advantages, as the study details. For crossbred cows, early intervention prevents a substantial production loss of 44,893.6 TK, with treatment costs amounting to only 6074.8 TK, resulting in a net benefit of 38,818.8 TK per cow per lactation. Holstein Friesian cows exhibit even greater benefits, avoiding a loss of 121,754.1 TK with an expenditure of 10,386.6 TK, yielding a benefit of 111,367.5 TK per cow per lactation. Though incurring lower absolute losses, Indigenous cows still benefit from early treatment, with a net gain of 3317.6 TK per cow per lactation.

The analysis highlights that the financial benefits of early diagnosis and treatment of SCM far outweigh the costs. Proactive measures include regular screening using CMT and MWST, maintaining strict milking hygiene, ensuring proper under-health management, providing balanced nutrition, and implementing targeted antimicrobial therapies based on susceptibility testing to reduce the overall economic burden, enhance milk production efficiency, and improve the profitability of dairy farming [33]. The reduction in production loss, coupled with manageable treatment costs, underscores the importance of early detection systems and prompt veterinary intervention [8,33]. Additionally, addressing SCM early prevents the long-term damage caused by recurrent SCM, preserving the health and productivity of dairy herds [33].

The economic analysis reinforces the critical need for early diagnosis and effective treatment protocols for SCM in dairy cows [61]. The substantial financial benefits justify the investment in early intervention, making it a cost-effective strategy for sustaining dairy production and profitability [62]. This study limits the hill tract area only which did not generalize the whole scenario of SCM in Bangladesh. We overlooked the economic analysis of CM. The economic losses due to CM should be explored in further investigations.

CONCLUSIONS

This study reveals the high prevalence of SCM in dairy farms across Chattogram and Cox's Bazar districts, with variations linked to socio-demographic, farm management, and animal-level factors. Key influences include age, breed, lactation stage, and parity, with medium-sized and intensively managed farms showing higher SCM rates, necessitating targeted interventions. SCM significantly alters milk's physical, chemical, and mineral properties, diminishing its economic and nutritional value. Additionally, it negatively affects milk protein fractions along with some key reproductive parameters, posing health and economic challenges for farmers. The findings highlight the need for improved farm management, awareness campaigns, and routine diagnostic practices to mitigate SCM. Region-specific strategies and policies are essential to reduce SCM prevalence, limit economic losses, and ensure the sustainability of Bangladesh's dairy sector.

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AUTHOR CONTRIBUTIONS

MPM, HH, KAB– Conceptualization, Methodology, Data Curation, Software, Formal Analysis, Investigation, Writing – Original Draft, Writing – Review and Editing; MI, MR, SIR, MM, MSH, – Data Curation, Investigation, Laboratory work, Formal Analysis, Writing-reviewing and editing, MSRC and MA- Data Curation, Formal Analysis, Writing – Original Draft, Writing – Review and Editing; MNH, AA, MMR and MMR – Conceptualization, Methodology, Data Curation, Software, Formal Analysis, Investigation, Validation, Visualization, Resources, Project Administration, Supervision, Writing – Original Draft, Writing – Review and Editing. All authors have approved the final version of the manuscript.

CONFLICTS OF INTEREST

There is no conflict of interest among the authors.

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