



Prevalence of Subclinical Mastitis and Associated Risk Factors at Cow Level in Dairy Farms in Jhenaidah, Bangladesh

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Abstract | We conducted a cross-sectional study to determine the prevalence of subclinical mastitis (SCM) among medium to large scale household dairy farms in the southwestern district, Jhenaidah, Bangladesh from July to December 2019. A total of 78 (n=100) lactating cows from household dairy farms (N=32) were selected randomly as sampled populations. We screened the milk samples for SCM by using Surf Field Mastitis Test (SFMT). The prevalence of SCM varied among farm level [71.9% (95% CI: 53.3-86.3)], individual animal level [67.9% (95% CI: 56.4-86.3)] and quarter level [29.5% (95% CI: 24.5-34.9)]. Descriptive statistics represented the farmers and farm demography, characteristics of the sampled population, and overall management feature. In this study area, the farms followed the intensive rearing system where around 65.6% of the shed was concrete made. The farm animals were crossbred type where HF × Local was predominant. Random effect logistic regression calculated, body condition score (BCS) [OR=3.8 and 2.9, at cows level and quarter level respectively (BCS-2 vs. BCS-≥3)], and breed [OR=5.1 and 2.9, at cows level and quarter level respectively (HF× Sahiwal vs. HF × Local)] as potential risk factors. This study shows that SCM is highly prevalent in the study area, which is a major threat to the dairy industry's production performance. Regular screening by SFMT, proper hygiene, improve the management system, and farmer's awareness is required to control the disease.

Keywords | Subclinical Mastitis, Prevalence, Surf Field Mastitis Test, Jhenaidah

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INTRODUCTION

Dairy farming in Bangladesh getting popularity day by day and presently around 6 million dairy cattle of crossbred high yielding cows are distributed in both household and commercial farms across the country which produces around 9.4 million metric ton liters of milk per year (Alam and Sarder, 2010; DLS, 2019). Around 70% of farmers of Bangladesh are smallholders having 1-3 cows per farm and contribute to 70-80% of the total milk demand of the country (Hemme et al., 2008; Uddin et al., 2012). Dairy cows in the household farms produce around 200-250 liter of milk per 305 days lactation period (Hussain, 2013). Production disease like mastitis is the major

hindrance to getting the optimum benefit from a dairy farm (Odhong et al., 2014). Mastitis impedes the dairy sector's growth of this country due to affecting health, decreased production, and the welfare of dairy cows (Kee, 2012; Rahman et al., 2014). Mastitis generates a considerable loss to the dairy industry, which has been estimated for Bangladesh as Tk. 122.6 (US \$2.11) million per year (Bari et al., 2014).

Though, many of the research on mastitis in dairy cows has been conducted in developed countries across the globe where intensive rearing system with machine milking provision is common (Kumar et al., 2013). Considerably exploration of mastitis in dairy cows of least developed

countries like Bangladesh (Anon, 2020) is less likely, where the herd size is usually smaller with hand milking provision. Hence this study aimed to estimate the prevalence of subclinical mastitis among household farms of Jhenaidah, Bangladesh.

Clinical mastitis causes a negative economic impact on dairy farms through abnormal milk, deterioration of milk quality, reduced production (up to 70%), milk discharge after treatment (9%), treatment costs (7%), labor, premature culling (14%) and death (Halasa et al., 2007; Kee, 2012; Bari et al., 2014), whereas subclinical mastitis (SCM) is in the absence of clinical signs rather than an increase in somatic cell counts of the milk (Radostits et al., 2007; Kayesh et al., 2014). Subclinical mastitis is 15 to 40 times more prevalent than that of clinical mastitis, is of long duration (Almaw et al., 2008; Sarker et al., 2013; Kathiriya et al., 2014). Many earlier studies have been reported the SCM status in Bangladesh along with neighboring countries, including India, Sri Lanka, and Pakistan as well (Kathiriya et al., 2014; Qayyum et al., 2016; Sanotharan et al., 2016; Kabir et al., 2017). In Bangladesh, the prevalence of SCM in the crossbred dairy cows has been documented as 28.5-61.3% (Kayesh et al., 2014; Tripura et al., 2014; Badiuzzaman et al., 2015; Kabir et al., 2017). Unfortunately, no study has been found on SCM in the southwestern region, especially at the Jhenaidah district of Bangladesh.

Several techniques including California Mastitis Test (CMT), Surf Field Mastitis Test (SFMT), Somatic cell Count (SCC), White Slide Test (WST) are used in field condition for SCM diagnosis (Kathiriya et al., 2014) among which SFMT has possessed a sensitivity of 72.8% and specificity 87.1% and available in field condition at a very reasonable cost (Muhammad et al., 2010; Kabir et al., 2017). So, this study has used the SFMT technique to popularize it in field conditions.

Multiple studies have been carried out to identify the risk factors of SCM in lactating cows in different countries, including Bangladesh (Sarker et al., 2013; Qayyum et al., 2016). The most-reported predisposing factor for SCM at farm level was the unhygienic environment, abnormally large udder, teat injury, udder wound, unclean milker's hand, and mismanagement of milking machine (Bari et al., 2014; Kathiriya et al., 2014). Some other risk factors that can hardly be influenced through management are age, parity, stage of lactation, and housing; whereas breeding, teat shape, and body condition score (BCS) are included as manageable risk factors (Islam et al., 2011; Bari et al., 2014; Mpatswenumugabo et al., 2017). Again, teat end to floor distance, pregnancy, milk yield, ways of milking stimulation with milking technique, milk leakage, and type of floor has also been reported as the most significant risk

factors of mastitis by different authors (Tripura et al., 2014; Qayyum et al., 2016). The knowledge of SCM risk factors is needed to provide farmers with advice to prevent clinical mastitis in their cows.

Besides Jhenaidah, a district of Bangladesh, where livestock farming is more common which is affected by different production diseases like mastitis (Sayeed et al., 2020). Therefore, the present study has been conducted to estimate the prevalence and risk factors associated with sub-clinical mastitis (SCM) in the Jhenaidah district of Bangladesh.

MATERIALS AND METHODS

STUDY AREA

The study was conducted in the registered household dairy farms in three selected upazilas (Jhenaidah Sadar, Harinakunda and, Kotchadpur) of the Jhenaidah district of Bangladesh during the six months from July to December 2019. The study area is geographically located at 23.54°N and 89.00°E at the southwestern region of Bangladesh, surrounded by Jashore district at the south, Kushtia at the north, Rajbari, and Magura at the east, and Chuadanga at the western border however Indian border at the southwestern part of this area (Islam, 2003). The district comprises 6 upazila, located geographically at a tropical climatic area with an annual temperature ranging from 11.2 to 37.10° C, and average rainfall is 1467mm (Wikimapia, 2020) (Figure 1).

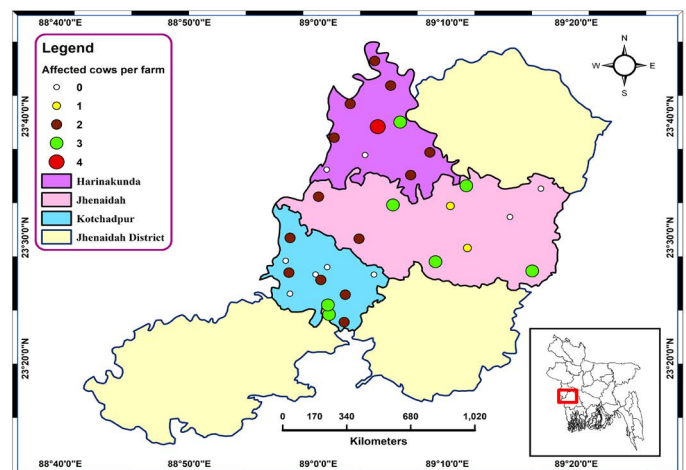


Figure 1: Spatial location of the study site with several positive samples per farm (some symbol of farm location is overlapped due to proximity with another farm)

REFERENCE POPULATION, SAMPLE SIZE, AND SAMPLING

Among six upazila of Jhenaidah district, we selected three upazila (sub-district) (Jhenaidah Sadar, Harinakunda, and Kotchadpur) based on easy accessibility to the field to conduct the study. All medium to large-sized commercial

household dairy farms recorded at District Livestock Office having at least one dairy lactating cow per farm during the study period in the study area were considered as the reference population. Thus, a complete list of household dairy farms (N=32 with 100 lactating cows) was prepared using the data obtained from the District Livestock Office of Jhenaidah.

To investigate the prevalence of SCM at the animal level, a total of 78 (N=100) individual lactating cows were required to evaluate considering 50% expected SCM prevalence (as no previous study has been reported SCM prevalence in this study area), $\pm 5\%$ precision, with a 95% confidence interval (<https://www.openepi.com/SampleSize>). The samples were collected from all the clinically healthy lactating cows under 32 commercial household dairy farms. Clinically healthy cows were defined as normal feeding behavior and body temperature and no visible changes in udder or milk.

REAGENT PREPARATION AND SFMT SENSITIVITY ANALYSIS

We prepared the reagent as the recipe described by Muhammad et al. (2010) and Bachaya et al. (2011). A, 3% surf solution ($P^H=10.3$) was briefly prepared by adding 3 gm detergent powder (Surf Excel®, Unilever, Bangladesh Ltd.) in 100ml of distilled water.

MILK SAMPLE EVALUATION FOR SCM

In the current study, we evaluated the milk samples based on the method described by Bachaya et al. (2011) in brief, after discarding 1-2 squirts of foremilk, 2ml milk and 2ml of SFMT solution were mixed to a CMT paddle, and it was twirled up to 20 to 30 seconds and observe the appearance of gel which was graded from 1 to 5 and interpreted as described by Iqbal et al. (2006) and Sharif et al. (2007). the normal consistency of milk was scored +1, indicating Negative (Fig. 2/A), whereas Light gel disappearing after stirring was scored +2, showing trace positive (Fig. 2/B). Again, light persistent gel short filaments were scored +3, indicating first degree positive for SCM (Fig. 2/C), Immediate thickening viscous cluster at the bottom of the well was scored +4, indicating second degree positive for SCM (Fig. 2/D). However, the thick gel consistency of egg albumin was scored +5, indicating third-degree positive SCM near clinical expression. No cows were found third-degree positive with a score of +5 in this study. The test score, 2 or more, was considered as SCM positive. If a sample from any quarter of four scored 2 or more, then that animal was treated as positive for SCM, and if any lactating cattle scored 2 or more, then the farm was defined as positive to SCM.

DATA COLLECTION

A pre-structured questionnaire was developed and reviewed under the Udder Health Development Project (<https://uhb.org.bd>) of Bangladesh run by Chattogram Veterinary and Animal Sciences University in collaboration with Utrecht University, The Netherlands and Swedish National Veterinary Institute, Sweden. The questionnaire was structured on farmers and farm demography, farm characteristics, and farm management. The data were recorded in the paper-based questionnaire by face to face interview and direct observation. The physical examination of cows recorded udder and teat shapes during sample collection. The udder and teat shapes were classified according to Bhutto et al. (2010). One hour was taken for each interview to fill the questionnaire. The cross-breed composition was recorded based on the history of artificial insemination in the previous parent stock of the sampled animals.

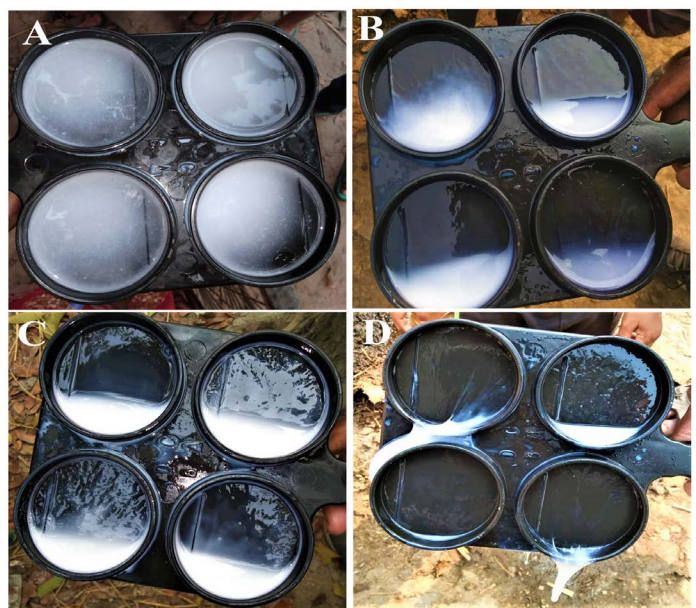


Figure 2: Milk sample evaluation using SFMT solution. A) Normal consistency of milk indicating negative, B) Light gel disappearing after stirring showing trace positive, C) Light persistent gel short filaments indicating first degree positive, D) Immediate thickening viscous cluster at the bottom of the well indicating second degree positive

STATISTICAL ANALYSIS

Sample evaluation output and field data were cleaned and entered into Microsoft Excel spreadsheet 2007, and then transferred to STATA/MP 14.0 (Stata Corp LP, Texas, USA). Different variables were filed to facilitate analyses as categorical variables where quantitative variables were categorized based on percentile. Body Condition Score (BCS) was evaluated, as described by Roche et al. (2009). Descriptive statistics were received on the data of farmer's demography, farm features, characteristics of sampled cows, and management features of farms. The prevalence of SCM was calculated at the farm level as well as the individual animal level and quarter level, and 95% Confidence

Table 1: Descriptive analysis of household dairy farm demography and basic management info in Jhenaidah district (N=32).

Variable	Category	Frequency (%)	95% Confidence Interval
Occupation	Farming	28 (87.5)	71.0-96.5
	Farming and small business	4 (12.5)	3.5-29.0
Educational status	Illiterate	3 (9.4)	2.0-25.0
	≤Class 8	20 (62.5)	43.7-78.9
	>Class 8 to SSC	6 (18.8)	7.2-36.4
	>SSC	3 (9.4)	2.0-25.0
Types of shed	Concrete	21 (65.6)	46.8-81.4
	Semi concrete	11 (34.4)	18.6-53.2
Boundary	Concrete	12 (37.5)	21.1-56.3
	Iron rod	1 (3.1)	0.08-16.2
	Absent	19 (59.4)	40.6-76.3
Floor	Concrete	25 (78.1)	60.1-90.7
	Brick	4 (12.5)	3.5-28.9
	Muddy	3 (9.4)	1.9-25.1
Bedding material	Rubber mat	24 (75.0)	56.6-88.5
	Absent	8 (25.0)	11.5-43.4
Drainage facility	Present	25 (78.1)	60.1-90.7
	Absent	7 (21.9)	9.3-39.9
Frequency of cleaning floor	Once	6 (18.8)	7.2-36.4
	Twice	19 (59.4)	40.6-76.3
	Thrice	7 (21.9)	9.3-39.9
Own fodder land	Yes	17 (53.1)	34.7-70.9
	No	15 (46.9)	29.1-65.3
Professional training on farming	Yes	14 (43.8)	26.4-62.3
	No	18 (56.3)	37.7-73.6
Breed	Cross	32 (100.0)	89.1-100*
Rearing system	Intensive	24 (75.0)	56.6-88.5
	Semi intensive	8 (25.0)	11.5-43.4
Semen Source	BRAC	15 (46.9)	29.1-65.3
	Government	17 (53.1)	34.8-70.9

Table 2: Descriptive analysis of household dairy farm animal's demography in Jhenaidah district (N=78).

Variable	Category	Frequency (%)	95% Confidence Interval
Crossbred	HF × Local	67 (85.9)	76.2-92.7
	HF × Sahiwal	11 (14.1)	7.3-23.8
Age (years)	≤3.5	30 (38.5)	27.7-50.2
	3.6-5.5	30 (38.5)	27.7-50.2
	≥5.5	18 (23.1)	14.3-34.0
Body weight (kg)	≤320	35 (44.9)	33.6-56.6
	≥321	43 (55.1)	43.4-66.4
BCS	≥3	47 (60.3)	48.5-71.2
	2	31 (39.7)	28.8-51.5
Parity	≤2	43 (55.1)	43.4-66.4
	≥3	35 (44.9)	33.6-56.5

Herd size (No. of milch cow)	≤2	28 (35.9)	25.3-47.5
	3-4	29 (37.2)	26.5-48.9
	≥5	21 (26.9)	17.5-38.2
Dry cow	≤2	31 (56.4)	42.3-69.7
	≥3	24 (43.6)	30.3-57.7
Pregnancy	Non-pregnant	25 (32.1)	21.9-45.6
	Pregnant	53 (67.9)	56.4-78.1
Lactation stage	Early	31 (39.7)	28.8-51.5
	Mid	31 (39.7)	28.8-51.5
	Late	16 (20.5)	12.2-31.2
Lactation period (month)	≤2	31 (39.7)	28.8-51.5
	3-5	28 (35.9)	25.3-47.5
	≥6	19 (24.4)	15.3-35.4
Udder shape	Bowl	10 (12.8)	6.3-22.3
	Cup	1 (1.3)	0.03-6.9
	Tight	57 (73.1)	61.8-82.5
	pendulous	10 (12.8)	6.3-22.3
Teat shape	Cylindrical	12 (15.4)	8.2-25.3
	Flat	3 (3.9)	0.8-10.8
	Pointed	61 (78.2)	67.4-86.8
	Round	2 (2.6)	0.3-8.9
Types of stimulation used during milking	Calf/suckling	75 (96.2)	89.2-99.2
	Hand	3 (3.9)	0.8-10.8
Frequency of milking	1	19 (24.4)	15.3-35.4
	≥2	59 (75.6)	64.6-84.7
Milk production (liter)	≤10	31 (39.7)	28.8-51.5
	11-20	39 (50.0)	38.5-61.5
	≥21	8 (10.3)	4.5-19.2
Calf suckling practices after milking	Calf not suckling	5 (6.4)	2.1-14.3
	Calf suckling	73 (93.6)	85.7-97.9
Previous history of mastitis	Yes	35 (44.9)	33.6-56.5
	No	43 (55.1)	43.4-66.4
Average daily milk yield before mastitis (liter)	≤10	6 (17.1)	6.6-33.6
	11-20	18 (51.4)	34.0-68.6
	≥21	11 (31.4)	16.9-49.2
Average daily milk yield after mastitis (kg)	≤2	7 (20.0)	8.4-36.9
	2.1-3.5	12 (34.3)	19.1-52.2
	≥4	16 (45.7)	28.8-63.3

Table 3: The Prevalence of subclinical mastitis in Jhenaidah district.

Level	Frequency (%)	95% Confidence Interval
Quarter (312)	92 (29.5)	24.5-34.9
Individual (78)	53 (67.9)	56.4-86.3
Farm (32)	23 (71.9)	53.3-86.3

Interval was estimated. The descriptive outputs were expressed in (CI) frequency value, percentage, and 95% CI.

A univariate chi-square test was performed to investigate the relationship between different variables and SCM. The variable indicating significant in univariate analysis ($P \leq 0.05$) was forwarded for multivariate analysis as described by Sayeed et al. (2017). The difference between likelihood-based standard error and robust (residual-based) standard error of logistic regression for both individual animal level and quarter level data indicate statistical clustering where particular farm and the individual animal was a cluster, a respectively. We fitted a random-effect logistic regression model using a backward elimination process for both individual and quarter-level analyses and checking for collinearity using chi-square statistics between independent variables. Hence two factors- frequency of milking (once and >once in a day) and previous history of mastitis (yes/no) was significant in univariate analysis. but due to collinearity with both body condition score (BCS) and crossbred, those two factors were dropped from multivariate analysis. We expressed the final outputs of the model in Odds Ratio (OR), wald test p-value, and 95% Confidence Interval (CI).

RESULTS

FARM DEMOGRAPHY

In Jhenaidah district, 87.5% of total farmers ($n=32$) responded to dairy farming as their main occupation. Maximum farmers were educated at a different level, where 9.4% of farmers were illiterate. Cow sheds were mostly made up of concrete (65.6%) and semi-concrete (34.4%), but 59.4% had no boundary/fence in their farms. Most of the farms had concrete floor (78.1%) followed by brick (12.5%) and earthen (9.4%) floor; rubber mattress was predominantly used in the farms (75.0%). Around 78.1% of farms had a well-constructed drainage facility, and 59.4% of farmers have cleaned the floor twice daily, followed by thrice daily (21.9%) and once daily (18.8%). About 53.1% of farms had their fodder land. Only 43.8% of farmers had professional training on dairy farming organized by the upazila livestock office. Most farmers were in the intensive rearing (75.0%), followed by semi-intensive farming (25.0%). For artificial insemination, 53.1% of farmers used the semen supplied by the government, whereas, 46.9% of farmers used the semen provided by Bangladesh Rural Advancement Committee (BRAC). Nevertheless, there had no quarantine and footbath facility on the farm (Table 1).

FARM ANIMAL DEMOGRAPHY

The studied farms were based on an intensive rearing system and preferred crossbred animals such as HF crossed with local indigenous cattle (85.9%) followed by HF crossed

with Sahiwal (14.1%). Around 37.2% of farms' herd size of milch cows was between 3 to 4, 35.9% of farms' herd size was ≤ 2 , and 26.9% of farms' herd size was ≥ 5 . Around 38.5% of animals were in ≤ 3.5 years old, whereas 38.5% and 23.1% of animals were between 3.6 and 5 and >5 years old, respectively. Again, 44.9% of animals' body weight was ≤ 320 kg, and 55.1% of animals' body weight was ≥ 321 kg. About 60.3% of animals were in $\text{BCS} \geq 3$, whereas 39.7% were in the BCS-2 category. Most of the lactating animals were in $\geq 3^{\text{rd}}$ parity (44.9%), whereas 55.1% of animals were in $\leq 2^{\text{nd}}$ parity. Among the lactating animals, around 67.9% were pregnant and 32.1% were non-pregnant. Almost 75.6% of farms practiced milking twice or more in a day. Overall, 39.7% of animals produced ≤ 10 liters of milk per day, whereas 50.0% of animals produced 11-20 liters of milk and around 10.3% of animals produced ≥ 21 liters of milk. About 44.9% of sampled animals were previously affected with mastitis (Table 2).

PREVALENCE OF SUB CLINICAL MASTITIS IN JHENAIDAH DISTRICT

The prevalence varied among farm level, individual animal level, and quarter level as well. The overall prevalence of SCM was 71.9% at the farm level, 76.9% at the cow level and, 29.5% at the quarter level (Table 3).

Geographically, farm-level SCM was more prevalent in Jhenaidah (90.0%) followed by Kotchadpur (83.3%) and Harinakunda (60.0%). Among individual animal levels, the highest prevalence was estimated in Kotchadpur upazila at 73.3% followed by Jhenaidah Sadar upazila (68.0%) and Harinakunda upazila (60.9%). Again, in quarter level, the highest prevalence was estimated in Jhenaidah Sadar upazila (42.0%) followed by Kotchadpur upazila (25.0%) and Harinakunda upazila (21.9%) (Table 4).

RISK FACTORS FOR SCM

The factors determined as significant in univariate analysis were nominated for multivariate analysis (Table 5). The estimated odds ratio of SCM was 3.8 times higher among individual animals having BCS-3 or more in contrast to BCS-2. In contrast, the odds ratio was 2.9 times higher at the quarter level of the same BCS group. Again, the odds of SCM was 5.1 times higher among the individual animals of having HF and Local crossed blood than the animals of HF and Sahiwal crossed blood. However, the odds of SCM was 2.9 times higher among the HF \times Local rather than HF \times Sahiwal at quarter level (Table 6).

DISCUSSION

FARMERS AND FARM DEMOGRAPHY

In Jhenaidah district, farmers reared crossbred animals following a predominantly intensive system of rearing

Table 4: The prevalence of subclinical mastitis in different Upazila under Jhenaidah district

Level	Upazilla	N	Frequency (%)	95% Confidence Interval
Quarter	Jhenaidah sadar	100	42 (42.0)	32.2-52.3
	Harinakunda	96	21 (21.9)	14.1-31.5
	Kotchadpur	116	29 (25.0)	17.4-33.9
Individual	Jhenaidah sadar	25	17 (68.0)	46.5-85.0
	Harinakunda	23	14 (60.9)	38.5-80.2
	Kotchadpur	30	22 (73.3)	54.1-87.7
Farm	Jhenaidah sadar	10	9 (90.0)	55.5-99.7
	Harinakunda	10	6 (60.0)	26.2-87.8
	Kotchadpur	12	10 (83.3)	51.6-97.9

Table 5: Univariable association between individual animal level and quarter level potential risk factors and SFT score

Variable	Category	Animal Level		Quarter Level	
		Positive (%)	P-value	Positive (%)	P-value
Breed	HF× Sahiwal	5 (38.5)	0.01	9 (17.3)	0.04
	HF × Local	48 (73.9)		83 (31.9)	
Body Condition Score (BCS)	≥3	39 (78.0)	0.01	71 (35.5)	<0.01
	2	14 (50.0)		21 (18.8)	
Udder Shape	Bowl and Cup	9 (81.8)	0.54	18 (40.9)	0.07
	Tight	37 (64.9)		59 (25.9)	
	Pendulous	7 (70.0)		15 (37.5)	
Teat Shape	Cylindrical	9 (75.0)	0.38	17 (35.4)	0.41
	Flat	1 (33.3)		2 (16.7)	
	Pointed and round	43 (68.3)		73 (28.9)	
Parity	1-3	43 (64.2)	0.08	76 (28.4)	0.28
	4-6	10 (90.9)		16 (36.4)	
Frequency of milking	Once in a day	7 (36.8)	<0.01	10 (13.2)	<0.01
	>Once	46 (77.9)		82 (34.8)	
Lactation stage	Early	20 (64.5)	0.76	31 (25.0)	0.36
	Mid	12 (75.0)		41 (33.1)	
	Late	21 (67.7)		20 (31.3)	
Pregnancy	Pregnant	38 (71.7)	0.30	68 (32.1)	0.14
	Non-pregnant	15 (60.0)		24 (24.0)	
Previous history of mastitis	Yes	30 (85.7)	<0.01	57 (40.7)	<0.01
	No	23 (53.5)		35 (20.4)	

Table 6: Multivariate association between individual animal level and quarter level potential risk factors and SFMT score

Factors	Category	Animal Level			Quarter Level		
		OR	95% CI	p-value	OR	95% CI	p-value
Body Condition Score (BCS)	2	Ref			Ref		
	≥3	3.8	1.3-11.1	0.01	2.9	1.3-6.7	0.01
Breed	HF× Sahiwal	Ref			Ref		
	HF × Local	5.1	1.3-18.9	0.01	2.9	0.9-9.1	0.06

supported by an earlier study (Hossain et al., 2004). Most of the dairy farms of Jhenaidah district had concrete floor (78.1%), and others had brick (12.5%) and muddy (9.4%) floor, which is in line with the study conducted by Hos-

sain et al. (2004) at Rangpur, Bangladesh. Almost all of the farmers were preferred to artificial insemination, and most of them used from government-supplied semen, which may be due to easy availability at a very reasonable price. However, the current study revealed that 53.1% of farmers have their fodder land supported by an earlier study (Hossain et al., 2005). Around 43.8% of dairy farmers received training at least once on dairying, which is much higher than the earlier study (Hossain et al., 2005) where about 17% of dairy farmers were reported on received training. This variation might be due to differences in the farmers' consciousness due to geographical location and socioeconomic status.

PREVALENCE OF SCM

The prevalence of SCM at the farm level was 71.9%, which might be a matter of significant anxiety for the dairy farmers of this region. However, the study revealed cow level prevalence of SCM (67.9%) is higher than the earlier reported prevalence in different district of Bangladesh including Sirajganj, Rajshahi, Chattogram, and Barisal (Barua et al., 2014; Kayesh et al., 2014; Badiuzzaman et al., 2015; Kabir et al., 2017). In the world context, the present study reported SCM prevalence is lower than the study conducted in Nigeria (85.3%) (Shittu et al., 2012), Uganda (86.2%) (Abrahmsén et al., 2014), and Vietnam (88.6%) (Östenson et al., 2013). On contrary, the estimated SCM prevalence is higher than the study conducted in Kenya (Mureithi and Njuguna, 2016), Ethiopia (Abebe et al., 2016), New South Wales of Australia (Plozza et al., 2011), Rwanda (Mpatswenumugabo et al., 2017) and Uruguay (Giannechini et al., 2002). The variation of cow level SCM prevalence among different studies conducted within the country and around the globe might be due to the difference in geographical location, climatic condition, farm composition, and overall husbandry practice (Barua et al., 2014).

Quarter level prevalence of SCM is in close agreement with the studies conducted in different parts of the country, including Barisal (Kayesh et al., 2014) Mymensingh and Tangail (Islam et al., 2011) but much lower than the study conducted in Rajshahi (Badiuzzaman et al., 2015) district of Bangladesh. Moreover, the estimated value is much higher than the reported prevalence in a neighboring country like Pakistan (Qayyum et al., 2016) and India (Joshi and Gokhale, 2006). The differences in prevalence between studies might be due to differences in milking practice, environmental conditions, and animals' immune status, which is supported by Qayyum et al. (2016).

RISK FACTORS OF SCM

The present study revealed a strong significant association between the prevalence of SCM and BCS, where the odds

ratio has been calculated as 3.8 and 2.9 among ≥ 3 BCS grouped animals at the individual level and quarter level respectively. This finding is in line with the study conducted by Sarker et al. (2013) who reported that animals with higher BCS might produce more milk that makes them prone to SCM. The present statement of this study is also supported by Haile-Mariam et al. (2001), who reported a higher infection rate among animals having higher BCS due to decrease lymphocytic functions (Banos et al., 2013). Besides, cows with higher BCS might have more chance of suffering from metabolic diseases (Roche et al., 2013), make them more susceptible to other infectious diseases like mastitis, supported by Chagunda et al. (2006) and Moyes et al. (2009). However, the present study estimated a significant relation of different categories of crossbred cows with the prevalence of SCM both in univariate and multivariate analyses. As a crossbred of HF and Local having higher OR (5.1) in contrast to a crossbred of HF and Sahiwal which can be supported by the study conducted in Chattogram, Bangladesh revealed a higher prevalence of SCM among the crossbred species of HF and Local (Barua et al., 2014). In contrast, the study conducted by Sanotharan et al. (2016) reported a comparatively lower prevalence of SCM among the Sahiwal breed in Srilanka. This statement can be concluded that Sahiwal is a temperate breed with more milk producibility and resistance to mastitis so, the cross of HF and Sahiwal are well tolerated than local, and HF crossbred progeny (Islam and Bhuiyan, 1997; Zafar et al., 2008).

CONCLUSIONS

Subclinical mastitis is considered to be an important challenge for dairy development. The prevalence of SCM is 71.9%, 67.9%, and 29.5% calculated at farm level, individual animal level, and quarter level, respectively. The cows with BCS ≥ 3 and crossbred cows of HF \times Local are more likely to be infected with SCM. To stop the progression of SCM to CM, it needs to control SCM at its early stage. So, the use of SFMT is a cost-effective and easily applicable technique for regular screening of SCM in field conditions. Care and management should be improved, and the farmers should be aware of the economic importance of the disease.

LIMITATIONS

The unwillingness of the farmer to test their cows fearing, reducing milk demand.

The majority of the farm does not have a proper record book, and they were not interested in disclosing the disease information.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS CONTRIBUTION

MAS designed the study. MAR and MAS did the fieldwork and data collection. MAS did the analysis. MAS prepared the draft manuscript with the help of MSB and AI. MMR and MAH reviewed the manuscript and supported the study by providing a questionnaire and field gears.

REFERENCES

- Abebe R, Hatiya H, Abera M, Megersa B, Asmare K (2016). Bovine mastitis: prevalence, risk factors and isolation of *Staphylococcus aureus* in dairy herds at Hawassa milk shed, South Ethiopia. *BMC Vet. Res.* 12: 270. <https://doi.org/10.1186/s12917-016-0905-3>
- Abrahmsén M, Persson Y, Kanyima BM, Båge R (2014). Prevalence of subclinical mastitis in dairy farms in urban and peri-urban areas of Kampala, Uganda. *Trop. Anim. Health Pro.* 46: 99-105. <https://doi.org/10.1007/s11250-013-0455-7>
- Alam M, Sarder M (2010). Effects of nutrition on production and reproduction of dairy cows in Bangladesh. *Bang Veterinarian.* 27: 8-17. <https://doi.org/10.3329/bvet.v27i1.5910>
- Almwaw G, Zerihun A, Asfaw Y (2008). Bovine mastitis and its association with selected risk factors in smallholder dairy farms in and around Bahir Dar, Ethiopia. *Trop. Anim. Health Pro.* 40: 427-432. <https://doi.org/10.1007/s11250-007-9115-0>
- Anon (2020) The World Bank In Bangladesh, available at: <https://www.worldbank.org/en/country/bangladesh/overview#:~:text=Bangladesh%20has%20an%20impressive%20track,than%20%2430%20billion%20in%20support.accessed%20on%20February%20,2020>
- Bachaya H, Raza M, Murtaza S, Akbar I (2011). Subclinical bovine mastitis in Muzaffar Garh district of Punjab (Pakistan). *J. Anim. Plant Sci.* 21: 16-19.
- Badiuzzaman M, Samad M, Siddiki S, Islam M, Saha S (2015). Subclinical mastitis in lactating cows: Comparison of four screening tests and effect of animal factors on its occurrence. *Bang. J. Vet. Med.* 13: 41-50. <https://doi.org/10.3329/bjvm.v13i2.26627>
- Banos G, Wall E, Coffey MP, Bagnall A, Gillespie S, Russell GC, McNeilly TN (2013). Identification of immune

traits correlated with dairy cow health, reproduction and productivity. *PloS One.* 8: <https://doi.org/10.1371/journal.pone.0065766>

- Bari M, Alam M, Uddin M, Rahman M (2014). Prevalence and associated risk factors of bovine clinical mastitis in Patiya upazila under Chittagong district of Bangladesh. *Int. J. Nat. Sci.* 5-9. <https://doi.org/10.3329/ijns.v4i1.28585>
- Barua M, Prodhan MAM, Islam K, Chowdhury S, Hasanuzzaman M, Imtiaz MA, Das GB (2014). Sub-clinical mastitis prevalent in dairy cows in Chittagong district of Bangladesh: detection by different screening tests. *Vet. World.* 7. <https://doi.org/10.14202/vetworld.2014.483-488>
- Bhutto AL, Murray RD, Woldehiwet Z (2010). Udder shape and teat-end lesions as potential risk factors for high somatic cell counts and intra-mammary infections in dairy cows. *Vet. J.* 183: 63-67. <https://doi.org/10.1016/j.tvjl.2010.08.007>
- Chagunda MG, Larsen T, Bjerring M, Ingvarstsen KL (2006). L-lactate dehydrogenase and N-acetyl-β-D-glucosaminidase activities in bovine milk as indicators of non-specific mastitis. *J. Dairy Res.* 73: 431-440. <https://doi.org/10.1017/S0022029906001956>
- DLS (2019) Livestock economy (2018-2019) at a Glance, DLS, available at <http://dls.portal.gov.bd>, accessed on February 20, 2020.
- Giannechini R, Concha C, Rivero R, Delucci I, López JM (2002). Occurrence of clinical and sub-clinical mastitis in dairy herds in the West Littoral Region in Uruguay. *Acta Vet. Scandi.* 43: 221. <https://doi.org/10.1186/1751-0147-43-31>
- Haile-Mariam M, Bowman P, Goddard M (2001). Genetic and environmental correlations between test-day somatic cell count and milk yield traits. *Lives Prod. Sci.* 73: 1-13. [https://doi.org/10.1016/S0301-6226\(01\)00232-9](https://doi.org/10.1016/S0301-6226(01)00232-9)
- Halasa T, Huijps K, Østerås O, Hogeveen H (2007). Economic effects of bovine mastitis and mastitis management: A review. *Vet. Quart.* 29: 18-31. <https://doi.org/10.1080/01652176.2007.9695224>
- Hemme T, Deeken E, Ramanovich M (2008). IFCN Dairy Report. International Farm Comparison Network IFCN Dairy Research Center Kiel Germany: 25-29.
- Hossain M, Alam M, Rashid M, Asaduzzaman M, Rahman M (2005). Small scale dairy farming practice in a selective area of Bangladesh. *Pak. J. Nutri.* 4: 215-221. <https://doi.org/10.3923/pjn.2005.215.221>
- Hossain Z, Hossain S, Rashid M, Sultana N, Ali M (2004). Study on the present management condition of private dairy farm at Rangpur Sadar Thana in Bangladesh. *J. Biol. Sci.* 3: 135-154.
- Hussain S (2013). The Bangladesh Dairy Market. Global Agricultural Information Network Report:
- Iqbal M, Amjed M, Khan M, Qureshi M, Siddique U (2006). Comparative efficiency of some indirect diagnostic tests for the detection of subclinical mastitis in cows and buffaloes. *Pak. Vet. J.* 26: 73-79.
- Islam M, Islam M, Rahman M, Islam M (2011). Prevalence of subclinical mastitis in dairy cows in selected areas of Bangladesh. *Bang. J. Vet. Med.* 9: 73-78. <https://doi.org/10.3329/bjvm.v9i1.11216>
- Islam S (2003). *Banglapedia: national encyclopedia of Bangladesh vol 3.* Asiatic society of Bangladesh, p
- Islam S, Bhuiyan A (1997). Performance of crossbred Sahiwal cattle at the Pabna milkshed area in Bangladesh. *Asian Austral. J. Anim. Sci.* 10: 581-586. <https://doi.org/10.5713/>

- ajav.1997.581
- Joshi S, Gokhale S (2006). Status of mastitis as an emerging disease in improved and periurban dairy farms in India. *Ann. N Y Acad. Sci.* 1081: 74-83. <https://doi.org/10.1196/annals.1373.007>
 - Kabir MH, Ershaduzzaman M, Giasuddin M, Islam MR, Nazir KNH, Islam MS, Karim MR, Rahman MH, Ali MY (2017). Prevalence and identification of subclinical mastitis in cows at BLRI Regional Station, Sirajganj, Bangladesh. *J. Adv. Vet. Anim. Res.* 4: 295-300. <https://doi.org/10.5455/javar.2017.d227>
 - Kathiriya J, Kabaria B, Saradava D, Sanepara D (2014). Prevalence of subclinical mastitis in dairy cows in Rajkot district of Gujarat. *Int. J. Sci. Nat.* 5: 433-436
 - Kayesh MEH, Talukder M, Anower A (2014). Prevalence of subclinical mastitis and its association with bacteria and risk factors in lactating cows of Barisal district in Bangladesh. *Int. J. Bio. Res.* 2: 35-38. <https://doi.org/10.14419/ijbr.v2i2.2835>
 - Kee C (2012). Bovine mastitis: an Asian perspective. *Asian J. Anim. Vet. Adv.* 7: 454-476. <https://doi.org/10.3923/ajava.2012.454.476>
 - Kumar P, Sharma N, Ranjan R, Kumar S, Bhat Z, Jeong DK (2013). Perspective of membrane technology in dairy industry: A review. *Asian-australas J. Anim. Sci.* 26: 1347. <https://doi.org/10.5713/ajas.2013.13082>
 - Moyes K, Larsen T, Friggens N, Drackley JK, Ingvarsen KL (2009). Identification of potential markers in blood for the development of subclinical and clinical mastitis in dairy cattle at parturition and during early lactation. *J. Dairy Sci.* 92: 5419-5428. <https://doi.org/10.3168/jds.2009-2088>
 - Mpatswenumugabo JP, Bebora LC, Gitao GC, Mobegi V, Iraguha B, Kamana O, Shumbusho B (2017). Prevalence of subclinical mastitis and distribution of pathogens in dairy farms of Rubavu and Nyabihu districts, Rwanda. *J. Vet. Med.* <https://doi.org/10.1155/2017/8456713>
 - Muhammad G, Naureen A, Asi MN, Saqib M (2010). Evaluation of a 3% surf solution (surf field mastitis test) for the diagnosis of subclinical bovine and bubaline mastitis. *Trop. Anim. Health Prod.* 42: 457-464. <https://doi.org/10.1007/s11250-009-9443-3>
 - Mureithi D, Njuguna M (2016). Prevalence of subclinical mastitis and associated risk factors in dairy farms in urban and peri-urban areas of Thika Sub County, Kenya.
 - Odhong C, Wahome R, Vaarst M, Kiggundu M, Nalubwama S, Halberg N, Githigia S (2014). Challenges of conversion to organic dairy production and prospects of future development in integrated smallholder farms in Kenya. *Lives. Res. Rural Dev.* 26: 14-21
 - Östensson K, Lam V, Sjögren N, Wredle E (2013). Prevalence of subclinical mastitis and isolated udder pathogens in dairy cows in Southern Vietnam. *Trop. Anim. Health Prod.* 45: 979-986. <https://doi.org/10.1007/s11250-012-0320-0>
 - Plozza K, Lievaart J, Potts G, Barkema H (2011). Subclinical mastitis and associated risk factors on dairy farms in New South Wales. *Aust. Vet. J.* 89: 41-46. <https://doi.org/10.1111/j.1751-0813.2010.00649.x>
 - Qayyum A, Khan JA, Hussain R, Avais M, Ahmed N, Khan A, Khan MS (2016). Prevalence and Association of Possible Risk Factors with Sub-Clinical Mastitis in Cholistani Cattle. *Pak. J. Zoo.* 48: 1-6.
 - Radostits OM, Gay C, Hinchcliff KW, Constable PD (2007). A textbook of the diseases of cattle, horses, sheep, pigs and goats. *Vet. Med.* 10: 2045-2050
 - Rahman S, Begum I, Alam M (2014). Livestock in Bangladesh: Distribution, growth, performance and potential. *Lives. Res. Rural Dev.* 26: Article # 173
 - Roche JR, Friggens NC, Kay JK, Fisher MW, Stafford KJ, Berry DP (2009). Invited review: Body condition score and its association with dairy cow productivity, health, and welfare. *J. Dairy Sci.* 92: 5769-5801. <https://doi.org/10.3168/jds.2009-2431>
 - Roche JR, Kay JK, Friggens NC, Loor JJ, Berry DP (2013). Assessing and managing body condition score for the prevention of metabolic disease in dairy cows. *Vet Clinics: Food Anim. Prac.* 29: 323-336. <https://doi.org/10.1016/j.cvfa.2013.03.003>
 - Sanotharan N, Pagthinathan M, Nafees M (2016). Prevalence of bovine subclinical mastitis and its association with bacteria and risk factors in milking cows of Batticaloa District in Sri Lanka. *Int. J. Sci. Res. Innov. Tech.* 3: 2313-3759
 - Sarker SC, Parvin MS, Rahman AA, Islam MT (2013). Prevalence and risk factors of subclinical mastitis in lactating dairy cows in north and south regions of Bangladesh. *Trop. Anim. Health Pro.* 45: 1171-1176. <https://doi.org/10.1007/s11250-012-0342-7>
 - Sayeed M, Islam B, Nahar N, Bari M, Sultana S, Arfin S, Haldar P, Islam A (2020). Epidemiology of livestock and poultry diseases in Jhenaidah district of Bangladesh. *Adv. Anim. Vet. Sci.* 8: 804-812. <https://doi.org/10.17582/journal.aavs/2020/8.8.804.812>
 - Sayeed MA, Smallwood C, Imam T, Mahmud R, Hasan RB, Hasan M, Anwer MS, Rashid MH, Hoque MA (2017). Assessment of hygienic conditions of live bird markets on avian influenza in Chittagong metro, Bangladesh. *Pre. Vet. Med.* 142: 7-15. <https://doi.org/10.1016/j.prevetmed.2017.04.009>
 - Sharif A, Ahmad T, Bilal MQ, Yousaf A, Muhammad G, Pansota F (2007). Estimation of milk lactose and somatic cells for the diagnosis of sub-clinical mastitis in dairy buffaloes. *Int. J. Agri. Bio (Pak):*
 - Shittu A, Abdullahi J, Jibril A, Mohammed AA, Fasina FO (2012). Sub-clinical mastitis and associated risk factors on lactating cows in the Savannah Region of Nigeria. *BMC Vet. Res.* 8: 134. <https://doi.org/10.1186/1746-6148-8-134>
 - Tripura T, Sarker S, Roy S, Parvin M, Sarker R, Rahman A, Islam M (2014). Prevalence of subclinical mastitis in lactating cows and efficacy of intramammary infusion therapy. *Bang J. Vet. Med.* 12: 55-61. <https://doi.org/10.3329/bjvm.v12i1.20464>
 - Uddin M, Sultana M, Bruermer B, Peters K (2012). Assessing the impact of dairy policies on farm-level profits in dairy farms in Bangladesh: Benchmarking for rural livelihoods improvement policy. *J. Rev. Global Econ.* 1: 124-138. <https://doi.org/10.6000/1929-7092.2012.01.11>
 - Wikimapia (2020) Jhenaidah District Town, available at <http://wikimapia.org/24901555/Jhenaidah-Dist-Town>, accessed on February 20, 2020.
 - Zafar A, Ahmad M, Rehman S (2008). Study of some performance traits in Sahiwal cows during different periods. *Pak Vet. J.* 28: 84-88