Prevalence and risk factors of bovine mastitis in intensively managed dairy farm

A clinical report submitted in partial satisfaction of the requirement for the Degree of Doctor of Veterinary Medicine (DVM)

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Session: 2017-2018

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November 2023

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November 2023

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Abstract

The bovine mastitis continues to pose a major problem for the dairy sector, affecting both milk production as well as overall animal health. This clinical report investigated the prevalence of mastitis in dairy herds focusing on the associations with management and hygiene practices of farms. Key risk factors associated with mastitis were identified as bedding materials post-dipping and udder hygiene. A total of 357 lactating cow from 15 different dairy farms in Chattogram district were investigated. The duration of the study was from February to July, 2023. The study revealed that 93.33% of the dairy farms had at least one cow affected by mastitis. The prevalence of mastitis in 357 lactating cows was 12.89%. Of 5.4% cows with bedding materials and 18.19% of cows without bedding materials suffered from mastitis. Mastitis was observed in 11.87% and 30% respectively when the cows were milked under full hand milking and knuckling method. Almost similar rate of mastitis were observed whether the floor was cleaned 1 to 3 times or 4 to 6 times per day. Farms with concrete floors had 13.63% of cows with mastitis, while those with brick floors had 9.85% affected by mastitis. Post-dipping, bedding materials, udder hygiene were found significantly associated with mastitis ($P<0.05$). There was no significant relationship between mastitis and both pre-dipping and floor type were found ($P>0.05$). These findings provide valuable insights into addressing mastitis issues in the dairy industry. Improvement of management and hygienic practices is one of the most effective ways that dairy farmers can reduce the incidence of mastitis and gain significant benefits..

Keywords : Mastitis, risk factors, CMT, udder hygiene.

Chapter-I
Introduction

Mastitis, an inflammatory disease of the mammary glands, is a widespread and complex issue in the dairy sector. It is a prevalent disease that has a global impact due to its prevalence and economic impact (Santman et al., 2016) . Dairy cow mastitis is a multi-faceted illness with a long-standing history, resulting in economic losses due to a decrease in milk production, additional treatment expenses, additional labor, and premature culling of cows that are chronically infected (Omer et al., 2014). With the increasing global demand for dairy products, there is a growing need to understand the complex interplay between mastitis, external factors, and dairy hygiene. As the dairy industry continues to evolve and respond to changing market demands, the need for a comprehensive approach to mastitis understanding and management is essential. Mastitis is a complex condition with multiple causes and evolving dimensions, and the pathogenic agents responsible for it must be addressed on an ongoing basis. This is due to the fact that mastitis can be induced by a variety of environmental conditions and practices in the udder.

Mastitis is found in two types; clinical mastitis and subclinical mastitis. Subclinical mastitis is distinguished from acute mastitis by the lack of visible changes in the milk or udder, while clinical mastitis is characterized by gross abnormalities. The majority of subclinical cases of mastitis are preventable through effective farming management practices, such as stress-free conditions, proper maintenance of milking equipment and milking procedures, and the use of various rapid and easy diagnostic methods such as California Mastitis Test (CMT) (Abebe et al., 2016). In recent years, a variety of studies have examined the management mechanisms associated with subclinical mastitis (Breen et al., 2009; Devries et al., 2012); as well as clinical mastitis (Barnouin et al., 2005; Jansen et al., 2009; Richert et al., 2013). It is of paramount importance to maintain the health of dairy cows' udder in order to prevent the development of mastitis. This necessitates regular monitoring of the physical condition of the cow's udder, the early identification of any signs of injury or abnormality, and the prompt provision of appropriate treatment. Utilizing techniques such as teat dipping and spraying disinfectants following milking can help to reduce the risk of bacterial invasion. Furthermore,

proper nourishment and hydration of the cows is necessary to maintain the integrity of their udder.

Mastitis is a condition in which the udder is exposed to pathogens at the time of milking, which is a critical stage in the development of the infection. Adhering to strict udder hygienic practices, such as pre and post-dip, is essential for the health of the udder and the overall health of the herd and, consequently, of the dairy operation. According to numerous studies, chapped teat skin promotes colonization of bacteria which may eventually result in mastitis (Agger et al., !986). The role of milking personnel in the prevention of mastitis cannot be overstated. It is essential that they are adequately trained in the proper milking practices. This includes proper hand washing prior to milking, the proper cleaning and disinfection of milking machinery, and the application of disposable gloves. Failure to maintain proper hygiene while milking can lead to the spread of bacteria to the udder. Therefore, it is essential that farm workers are educated and regularly monitored to ensure that these practices are adhered to.

Additionally, the farm environment can have a significant influence on the occurrence of mastitis. The microbial numbers of milk are affected by the cow's health and hygiene, the environment in which the cow is housed and milked, and the hygiene of milking equipment and storage facilities (Omer et al., 2014). A well-maintained and dry barn environment reduces the exposure of the cow's udders to external pathogens. Regularly removing of manure and appropriate bedding management are important. Proper ventilation of the barn helps to regulate humidity and the amount of bacteria present in the air, thus decreasing the likelihood of contamination. Additionally, farmers should be mindful of the overall welfare of the cows. Stress and discomfort can weaken the cow's immune system, making them more prone to mastitis. Therefore, there is a need for continuous research and regular evaluation of preventive and control strategies to effectively address this condition. It is a condition in which the udder is exposed to pathogens at the time of milking, which is a critical stage in the development of the infection. Adhering to strict udder hygienic practices, such as pre and post-dip, is essential for the health of the udder and the overall health of the herd and, consequently, of the dairy operation.

Mastitis is a crucial topic for research since it presents significant obstacles for both the economy and animal welfare. It has wide-ranging effects on environmental elements like housing, milking methods, insect control initiatives, and udder hygiene routines like pre- and post-dipping. The creation of evidence-based prevention and treatment strategies depends on the identification of risk factors and how they affect the incidence of mastitis. Furthermore, maintaining optimal udder health is crucial to guaranteeing sustainable and effective production as the dairy industry works to satisfy the growing demand for milk and dairy products worldwide. The objective of this clinical report was to have a thorough understanding of the variables affecting the incidence of mastitis in dairy cattle. To be more precise, the objective of the present study was;

1. To evaluate the prevalence of mastitis associated with management and hygiene practices of dairy farms
2. To investigate the risk factors associated with mastitis in lactating dairy cows.

Chapter-II
Materials and Method

2.1 Study design :

The study was conducted in two different area of Chattogram district namely Chattogram metropolitan area and Karnaphuli. The areas were chosen for the study due to the abundant number of dairy farms present in those regions. Data was collected from February to July, 2023 in the study area.

2.2 Study population :

A total of 15 dairy farms were randomly selected from the dairy farms located in the study areas. The herd size ranged from 2 to 60 animals. Each farms consist of at least 2 lactating cows. In total, 357 lactating cows were selected for the study.

2.3 Data collection :

A questionnaire with closed-ended questions was created to gather information about different herd and animal-specific factors that are thought to play a role in the spread of mastitis in dairy herds. A pre-made questionnaire was filled out in person by asking questions. The questionnaire provided the following details i.e. total animals, milking cows, floor type, bedding materials, breed, milking techniques, milking type, pre dipping, post dipping, floor streaming frequency, CMT test, udder lesions.

2.4 California Mastitis Test (CMT) :

The California Mastitis Test was employed to screen for subclinical mastitis. It was carried out ac- cording to the procedure described by Quinn et al., 1999. Squeezing approximately 2 ml of milk from each quarter into each of the four shallow cups of the CMT paddle was then supplemented with an equal amount of the commercially available CMT reagent. A circular motion of the mixtures was applied in a horizontal direction for a period of 15s. The results of the test were classified as 0 (Negative), trace, 1 (Moderate Positive), 2 (Significant Positive) and 3 (Strong Positive) based on the gel formation.

2.5 Studied farms :

In table-1, farms location and number of lactating animals of each farm were described. The 15 farms included in the study varied in the number of lactating cows, with the lowest farm having two cows lactating and the highest farm having sixty cows lactating.

Table 1: Distribution of the farms in the studied area

Farm name	Location	No of milking animal
SK agro dairy farm	Dalipara, Changaon	11
JN dairy farm	East mohra, Chandgaon	60
Mahar & agro dairy farm	Jahangir abashik,Hathazari	25
Sam agro farm	Uttar gate, Muradnagar	4
Hazi dairy farm	Hamidpur,Bayezid	16
Ikhlas rijik dairy farm	Rupnagar Abashik	20
Shahjahan dairy farm	Maizpara, Chadgaon	44
Alia dairy farm	Jalalabad Abashik	7
Ajad dairy farm	Jalalabad, West Khulshi	2
Bandhan dairy farm	Karnaphuli	35
Shahnaz dairy farm	Karnaphuli	28
Yusuf dairy farm	Mid Shikolbaha	8
Eva dairy farm	Gheshia para, Khalpar	27
Homeland dairy farm	Khandakar para, Bayezid	60
Nur dairy farm	Mokbul society, Steel mill	10
		N = 357

2.6 Data analysis :

The collected data was analyzed in a comprehensive manner using a tabular approach, and accurate calculations and analysis were conducted using Microsoft Excel software. Calculations of frequency and percentage were used to provide a comprehensive scientific understanding. Associations between different variables were examined through a t-test, by STATA-13. The significance level for probability was set at $p<0.05$.

Chapter-III

Results

The study was conducted on 357 lactating cows from 15 different dairy farms of Chattogram district to determine the prevalence of mastitis and its associated risk factors.

Table 2: Prevalence of cow level mastitis in relation to different risk factors (N=357)

Variable		Number	Positive	Prevalence (%)
Bedding material	Yes	148	8	5.4
	No	209	38	18.19
Milking type	Full hand milking	335	40	11.87
	Knuckling	20	6	30
Floor cleaning frequency	1-3	66	8	12.12
	4-6	291	38	13.05
Floor type	Concrete	286	39	13.63
	Brick	71	7	9.85

In Table 2 result shows that the prevalence was higher in farms that did not use bedding materials (18.19%) on the floor compared to those that used bedding materials (5.4%). A variation was found in prevalence based on milking type. Mastitis was more prevalent in case of knuckling (30%) than full hand milking (11.87%). The prevalence was almost similar in case of floor cleaning frequency whether it had been cleaned 1-3 times/day or 4-6 times/day. A variation was found in prevalence based on floor type. Mastitis was more prevalent in case of concrete floor (13.63%) than brick floor (9.85%).

Table 3: Prevalence of mastitis in studied lactating cows

Observation	Overall mastitis		
	No of examined	Positive	Prevalence (%)
Herd level	15	14	93.33
Cow level	357	46	12.89

The incidence of mastitis at the herd and cow level is shown in table 3. Based on the CMT and clinical examination, mastitis was positive in 93.33 percent of the herds and in 12.89 percent of the individual cows.

Table 4: Regression analysis of the association of cow-level mastitis with different risk factors

Variable	Category	Mean ± SD	p value
Pre-dipping	Yes	4.6 ± 6.94	0.34
	No	2.3 ± 2.27	
Post-dipping	Yes	1.22 ± 0.67	0.03
	No	5.83 ± 5.94	
Bedding materials	Yes	1 ± .53	0.04
	No	5.42 ± 5.5	
Udder hygiene	Slightly dirty	.88 ± .35	0.02
	Moderate to very dirty	5.58 ± 5.38	
Floor type	Concrete	3.25 ± 4.77	0.75
	Brick	2.33 ± 1.52	

SD= Standard Deviation

Table 3 shows the relationship of some variables with the occurrence of mastitis. In the present study, the presence of mastitis was significantly influenced by post-dipping, bedding materials and udder hygiene ($p<0.05$). On the other hand, this study did not find any significant link between pre-dipping and floor type with mastitis occurrence.

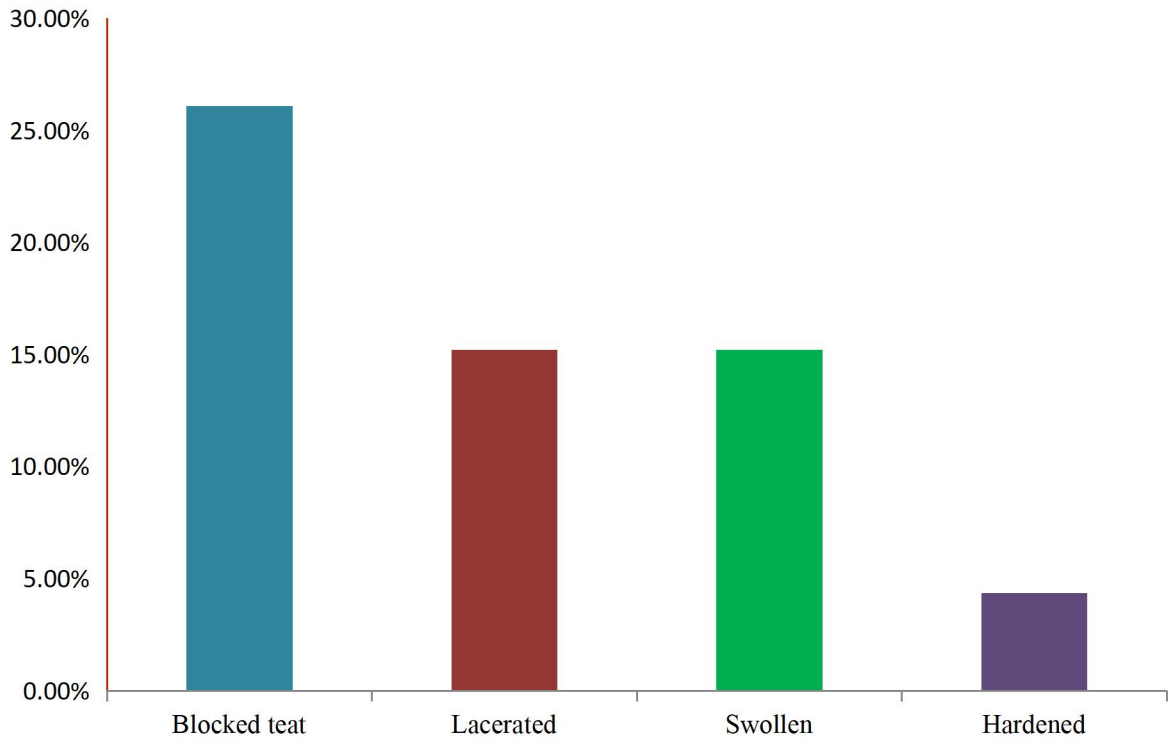

Fig 1: Types of udder lesions found on cow affected with mastitis

Figure 1 shows the frequency of udder lesions in mastitis affected cows. The prevalence was highest in blocked teat (26.09%) followed by lacerated (15.22%), swollen (15.22%) and hardened udder, respectively.

Chapter-IV

Discussion

The aim of this study is to explore the complex mechanisms that influence the incidence of mastitis on dairy farms. The study looks at a broad spectrum of farm management and hygiene factors to better understand the relationship between mastitis and dairy cattle. The study showed that the majority of herds 93.33 had at least one cow that had mastitis. Unfortunately, there weren't a lot of similar studies done in Bangladesh, so it was hard to compare the results at the herd level to other studies on mastitis. About 12.89% of cows were reported to have mastitis. This percentage represents the percentage of cows with mastitis within the study population. It is evident that the prevalence of herd level mastitis in this study was very high. A prevalence of 40 % or higher within a single herd should be a cause for concern for the producer. The prevalence of high mastitis in the study area is not only detrimental to milk production, but also adversely affects milk quality, resulting in economic losses and a public health risk. It is possible that the prevalence of high prevalence could be due to the lack of routine prevention and control strategies, other than treatment of clinical cases, in the dairy herds. This assumption is supported by the fact that no herds in the study implemented routine prevention measures, such as teat infection after milking, the wearing of gloves while milking, non-malignant mastitis treatment during the non- lactating period, the culling of infected animals, and others. Moreover, the farmers did not conduct any routine examinations or tests to determine whether the cow was suffering from subclinical mastitis. On the other hand, the risk factors associated with mastitis were more prevalent. These were bedding materials, milking type, floor type, post dipping and dirty udder.

In the present day, the primary focus of dairy producers is the implementation and maintenance of a comprehensive mastitis control program. Despite the prevalence of such control programs among dairy herds, such as the use of teat dipping, and machine milking maintenance, mastitis cases remain a recurring issue (Todhunter et al., 1995, Milne et al., 2003). Mastitis is a condition that is primarily caused by the accumulation of infection by a variety of mastitis pathogens. Factors that influence the successful implementation of a mastitis control program include the type of milking (hand or machine milking), the environmental conditions, and hygiene practices (Sergeant et al., 2001; Radostits, 2007).

Teat dipping is one of the best mastitis control programs and is one of the most popular because it is easy and affordable. The study revealed that there was no significant relation between the occurrence of mastitis and pre-dipping. The results of this study is consistent with Kamal et al., 2015 ; Oliver et al., 1993. This study further revealed that post dipping of teat after milking reduces the occurrence of mastitis in farm. It is a way to prevent the entry of bacteria into udder. This study is consistent with Khasanah et al., 2021; Kamal et al., 2015, Singh et al., 2018. On the other hand, the result of this study is in contrast to the findings of Schukken et al., 1990; Elbers et al., 1998; Barkema et al., 1999; Peeler et al., 2000 Santman-Berends et al., 2016. In their study they revealed that post-dipping is a risk factor for mastitis.This could be due to the fact that post milking teat infection prevention led to a decrease in infections with small pathogens, resulting in an increase in infections with major pathogens (Lam et al., 1997). Such differing findings highlight the complexity of the subject and the need for a comprehensive understanding of the factors influencing mastitis in dairy cows.

The study shows there was no correlation between the type of floor (concrete and brick) and occurrence of mastitis. No other study was found that is related to this factor. The study also shows that prevalence of mastitis is higher in knuckling method of milking than full hand milking. The condition of the cows' udder was also included in the risk factors that increase the incidence of mastitis. All cows in the study had slightly to severely dirty udders, and the incidence of mastitis increased significantly as the degree of discoloration increased. The dirt on the cows' udder was due to poor hygiene on dairy farms. Out of the total number of farms studied, 46.67% per cent were classified as poor hygiene due to inadequate waste drainage systems and the build-up of waste and urine on the udder. Similar to this study, other studies have also shown a strong correlation between poor hygiene on udder and an increased risk for mastitis (Abebe et al., 2016, Iraguha et al., 2015).

The results of the study also indicated that the absence of bedding material in cow shed had a significant correlation with the occurrence of mastitis, with cows in herds that had not been provided with bedding material exhibiting a 3.3-fold higher risk of developing

mastitis compared to those that had been provided with it. It was suggested that the floor of the shed may be a potential entry point for mastitis-causing organisms, particularly those associated with environmental pathogens, which could enter the cow's udder via the teat area. In response to this, Radostitis et al.,2004, stated that the provision of adequate bedding material would reduce the incidence of mastitis significantly.

Chapter-V

Conclusion

The study found key associations between certain risk factors and the prevalence of mastitis in dairy farm. In particular, post-dipping proved to be an effective preventive measure, significantly reducing the occurrence of mastitis, while pre-dipping did not show the same effect. The presence of bedding materials has been identified as a protective factor against mastitis. The knuckling method of milking showed a higher prevalence of mastitis, which emphasized the importance of milking techniques in mastitis control. Udder hygiene also played a key role, and as udder cleanliness worsened, the incidence of mastitis clearly increased, which was largely due to poor hygiene practices on the dairy farms studied. In addition, the study looked at the possible influence of floor type and cleaning frequency but there was no significant association with mastitis incidence. This information directs the specific areas that require additional attention and action to improve mastitis management. Taken together, the results of this study provide important insights into the urgent problem of mastitis and the need for preventive strategies to mitigate its effects. The focus should be on preventative measures such as post-dipping, maintaining good udder hygiene and providing bedding materials. These initiatives will not only improve the quality and production of milk, but will contribute significantly to the overall welfare of the dairy industry and the communities it serves in this region of Bangladesh.

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Appendix

```
. ttest Positive, by(udder)

Two-sample t test with equal variances
```

Group	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
0	7	5.571429	2.033729	5.380742	.5950725	10.54778
1	8	.875	.125	.3535534	.579422	1.170578
combined	15	3.066667	1.106116	4.283968	.6942843	5.439049
diff		4.696429	1.896657		.5989508	8.793906

```
    diff = mean(0) - mean(1)                                      t =   2.4762
Ho: diff = 0                     degrees of freedom =       13

    Ha: diff < 0                 Ha: diff != 0                 Ha: diff > 0
 Pr(T < t) = 0.9861         Pr(|T| > |t|) = 0.0278          Pr(T > t) = 0.0139
```

```
. ttest Positive, by(Bed_mat)

Two-sample t test with equal variances
```

Group	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
0	8	1	.1889822	.5345225	.553128	1.446872
1	7	5.428571	2.080031	5.503246	.338918	10.51822
combined	15	3.066667	1.106116	4.283968	.6942843	5.439049
diff		-4.428571	1.94559		-8.631764	-.225379

```
    diff = mean(0) - mean(1)                                      t =  -2.2762
Ho: diff = 0                     degrees of freedom =       13

    Ha: diff < 0                 Ha: diff != 0                 Ha: diff > 0
 Pr(T < t) = 0.0202         Pr(|T| > |t|) = 0.0404          Pr(T > t) = 0.9798
```

```
. ttest Positive, by( Predip )

Two-sample t test with equal variances
```

Group	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
0	5	4.6	3.108054	6.94982	-4.029341	13.22934
1	10	2.3	.715697	2.263233	.6809809	3.919019
combined	15	3.066667	1.106116	4.283968	.6942843	5.439049
diff		2.3	2.349959		-2.776778	7.376778

```
    diff = mean(0) - mean(1)                                      t =   0.9787
Ho: diff = 0                     degrees of freedom =       13

    Ha: diff < 0                 Ha: diff != 0                 Ha: diff > 0
 Pr(T < t) = 0.8272         Pr(|T| > |t|) = 0.3456          Pr(T > t) = 0.1728
```

```
. ttest Positive, by( Floortype )

Two-sample t test with equal variances
```

Group	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
0	12	3.25	1.376893	4.769696	.2194797	6.28052
1	3	2.333333	.8819171	1.527525	-1.46125	6.127916
combined	15	3.066667	1.106116	4.283968	.6942843	5.439049
diff		.9166667	2.858392		-5.258514	7.091848

```
    diff = mean(0) - mean(1)                                      t =   0.3207
Ho: diff = 0                     degrees of freedom =       13

    Ha: diff < 0                 Ha: diff != 0                 Ha: diff > 0
 Pr(T < t) = 0.6232         Pr(|T| > |t|) = 0.7535          Pr(T > t) = 0.3768
. ttest Positive, by( Posrdip)

Two-sample t test with equal variances
```

Group	Obs	Mean	Std. Err.	Std. Dev.	[95% Conf.	Interval]
0	9	1.222222	.2222222	.6666667	.7097769	1.734668
1	6	5.833333	2.427848	5.946988	-.4076479	12.07431
combined	15	3.066667	1.106116	4.283968	.6942843	5.439049
diff		-4.611111	1.963279		-8.852517	-.3697052

```
    diff = mean(0) - mean(1)                                      t =  -2.3487
Ho: diff = 0                     degrees of freedom =       13

    Ha: diff < 0                 Ha: diff != 0                 Ha: diff > 0
 Pr(T < t) = 0.0177         Pr(|T| > |t|) = 0.0353          Pr(T > t) = 0.9823
```

Acknowledgement

I am humbled and deeply grateful to the Almighty for granting me the health, ideas and opportunity to accomplish this assignment.

I would like to express my deep appreciation and gratitude to my supervisor, Professor Dr. Mohammad Rashedul Alam, Department of Physiology, Biochemistry and Pharmacology, Chattrogram Veterinary and Animal Sciences University, for his guidance, collaboration, genuine assistance, valuable advice, constructive criticism and his active involvement in this study since its inception. I would also like to express my appreciation to all my friends, and senior brothers for their support, encouragement and inspiration throughout the study period and the preparation of this report.

Finally, I would like to thank my parents and family for their ongoing support, suggestions and cordial assistance throughout the duration of my study.

Biography

I am Hossen Murad Shaoun, son of Nazir Ahmed and Taslima Begum doing my graduation on Doctor of Veterinary Medicine (DVM) at Chattogram Veterinary and Animal Sciences University under the Faculty of Veterinary Medicine. I passed my Secondary School Certificate Examination (SSC) in 2014 from Nasirabad Government High School, Chattogram, and then Higher Secondary Certificate Examination (HSC) in 2014 from Chattogram City College, Chattogram. Currently, I am doing my year-long internship programe. In future, I would like to work in Pet Animal Medicine since I have huge interest in it.