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Risk factors and therapy for goat mastitis in a hospital-based case-control study in Bangladesh

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ABSTRACT

Bangladesh has a large population of goats, which contribute to the income, nutrition and welfare of the households of many families. Mastitis in goats has a low incidence, but is often very severe, making veterinary care necessary. The aim of this study was to identify seasonality and risk factors for goat mastitis in a hospital-based matched case-control study in a teaching veterinary hospital in Chittagong, Bangladesh and to describe the range of antimicrobial treatments applied in this situation. Cases of mastitis and controls were drawn from the hospital patient recording system, along with their risk factor status. Multiple imputation was applied to deal with the missing values in the data analysis. Mastitis occurred somewhat more in the rainy season, and comprised about 3% of all goats admitted to the hospital during January 2011–June 2014. Free-ranging farming system, poor body condition score and non-native goat breeds were significantly associated with case status. Treatment of clinical mastitis was variable and unsystematic, but the use of gentamicin was commonly recorded. The need for more prudent and evidence-based antimicrobial therapies is discussed.

1. Introduction

In Bangladesh, about 24 million goats are kept by mostly rural women to produce meat, milk and skin and thereby contribute to the family income, nutrition and welfare (Division of Livestock Statistics, 2011). Mastitis is an important goat disease in this country, because it reduces the profitability of goat farming, impairs animal welfare and poses public health risks (Razi et al., 2012). Clinical mastitis in goats has a low incidence, but is often very severe (Bergonier et al., 2003; Cable et al., 2004). Especially gangrenous mastitis, a common presentation of clinical mastitis in goats, is often lethal or may lead to loss of the affected udder half in case the goat survives (Ribeiro et al., 2007; Contreras et al., 2007; Sarker et al., 2015). Milder cases of clinical mastitis and subclinical mastitis reduce the milk yield (Koop et al., 2010b) and thus may result in reduced growth of the kids. The use of antibiotics for treating mastitis may lead to antimicrobial resistance and antimicrobial residues in milk (Sawant et al., 2005). Therefore, mastitis control is important in goat husbandry. As mastitis in goats is often caused by contagious pathogens, culling of infected and clinically diseased animals is an important strategy to reduce the incidence of clinical mastitis in large scale goat farming (Koop et al., 2009). However, in small scale household goat farming, the value of an individual

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Table 1
Overview of conditions diagnosed in female goats (≥1 year of age) at the Teaching Veterinary Hospital of the Chittagong Veterinary and Animal Sciences University, Bangladesh (2011–2014[a]).

Year		Mastitis					Other cases							All
		Acute	Subacute	Gangrenous	Chronic	Total	PPR[b]	Parasitic infestation	Reproductive cases	Acidosis	Anorexia	Dog bite	Other[c]	
2011	*N*	13	1	3	7	24	106	258	62	26	168	45	132	821
	%	1.6	0.1	0.4	0.9	2.9	12.9	31.4	7.6	3.2	20.5	5.5	16.1	100.0
2012	*N*	15	16	3	7	41	148	321	83	31	227	43	156	1050
	%	1.4	1.5	0.3	0.7	3.9	14.1	30.6	7.9	3.0	21.6	4.1	14.9	100.0
2013	*N*	15	4	0	3	22	94	258	48	23	159	20	118	742
	%	2.0	0.5	0.0	0.4	3.0	12.7	34.8	6.5	3.1	21.4	2.7	15.9	100.0
2014[a]	*N*	5	1	2	1	9	63	85	28	18	73	13	43	332
	%	1.5	0.3	0.6	0.3	2.7	19.0	25.6	8.4	5.4	22.0	3.9	13.0	100.0
Total	*N*	48	22	8	18	96	411	922	221	98	627	121	449	2945
	%	1.6	0.7	0.3	0.6	3.3	14.0	31.3	7.5	3.3	21.3	4.1	15.2	100.0

[a] Cases included up to July 2014.
[b] PPR:Peste des Petits Ruminants.
[c] Fracture, accidental injury, poisoning and undiagnosed cases.

animal is too high to make such measures economically justifiable. This means that other preventative mastitis control measures are needed. Identification of risk factors for mastitis is therefore crucial, as knowledge of risk factors can help target prevention strategies. Also, under these circumstances, optimizing treatment protocols is important, because survival of the animal is of direct economic relevance. The fact that incomplete bacteriological cure may lead to recurrence of mastitis or may present a risk for infection of other animals or lead to human health problems on ingestion of contaminated raw milk or through manual milking makes effective treatment even more relevant. Several studies have compared the effectiveness of different antibiotic treatment protocols for goat mastitis (Doğruer et al., 2010; Jeph et al., 2013), showing that substantial differences in efficacy between treatment protocols exist.

The overall aim of this study is to contribute to improved mastitis prevention and control in small scale household goat farming situations. Specifically, with this study, we aim to describe seasonal patterns in clinical mastitis in goats in Bangladesh and identify risk factors associated with clinical mastitis in these goats, using a hospital-based case-control study. Additionally, we describe the variation in antimicrobial treatments that have been applied to goats suffering from clinical mastitis in the Chittagong Veterinary and Animal Sciences University (CVASU) Teaching Veterinary Hospital (TVH) in Bangladesh, to explore if there is need for standardised protocols and evidence-based veterinary medicine.

2. Material and methods

A retrospective case-control study was performed using the paper-based patient recording system in place at the TVH, CVASU. All does (≥1 year age) with a diagnosis of mastitis, based on clinical examination by registered veterinarians or clinical faculty of CVASU were defined as cases. These animals were submitted to the hospital by their owner because of suspected udder problems.

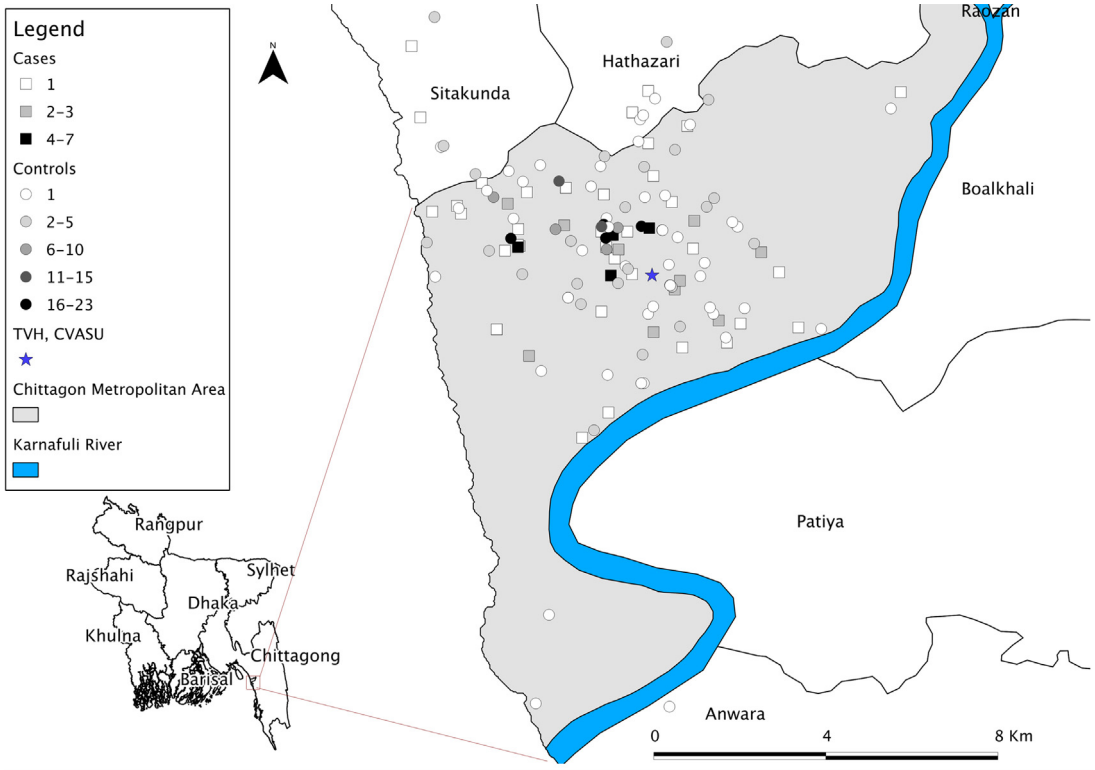

Fig. 1. Map of Bangladesh showing the number and location of case and control animals in this study.

All female goats of one year and older that were brought into the hospital between January 2011 and June 2014 were extracted from the recording system. Controls matched on month were randomly selected from all eligible controls in that month in an approximately 1:3 case to controls ratio, as a ratio of more than 3 controls per case adds little to the precision of the study. As matching 1:3 was not possible for all cases, we assigned 1, 2, or 4 controls to some cases.

For all cases and controls, date of admission to the hospital, rearing system, breed, weight, age and body condition score were extracted from the recording system. We used google earth (https://earth.google.com) to locate geocoordinates of cases and controls based on the address in the hospital recording system. For cases, also type of mastitis was recorded. Type of mastitis was classified by the clinic veterinarians as acute, subacute, gangrenous or chronic, but no systematic way of making this classification was used. Drugs prescribed to all clinical mastitis cases were also retrieved for the study and treatments of clinical mastitis were grouped according to the antimicrobial class or combination of classes used. Obvious associations between treatment regime and type of mastitis were assessed by descriptive and visual inspection of the results, as too different treatment regimes and not enough observations were present to statistically test for associations. For all other records in the dataset (i.e.,: all other goats brought into the hospital during the study period), only the diagnosis was recorded, to obtain an estimate of the fraction of cases brought in with mastitis relative to other diagnoses. The number and percentage of cases for each diagnosis were calculated over the years of the study and according to season of the year.

Univariate associations between potential risk factors (recoded into categorical variables, excluding missing values) and case or control status were investigated by means of univariate conditional logistic regression. Factors significant at $P<0.25$ were selected for the multivariable analysis. As the dataset had missing values in several variables, we used MICE 2.22 (van Buuren and Groothuis-Oudshoorn, 2011) to impute missing values. Ten imputed datasets were constructed with a maximum of 10 iterations. Convergence and mixing of the chains were assessed by plotting parameters against iteration number. Distributions of the imputed variables were inspected by plotting the density distributions for imputed and non-imputed data. Using the imputed datasets, conditional logistic regression models were manually constructed by backward-selection and the statistical significance of the contribution of individual predictors was determined using Wald's test. The presence of confounding factors and presence of collinearity was investigated by removing one of the variables and assessing changes in the beta-estimate and its standard error. A coefficient change of more than 10% was considered to indicate the presence of confounding; collinearity was suspected when the standard error of a variable in the model increased substantially upon entering a new variable in the model. Interactions between variables in the final model were tested for significance. The results were presented for each predictor variable as an odd ratio (OR) and 95% CI. For comparison, we also ran the final conditional logistic regression model on the non-imputed dataset.

ArcGIS-ArcMap version 10.2 (ESRI, USA) was used to prepare a map showing the spatial distribution of cases and controls in the study.

3. Results

3.1. Descriptive statistics

Descriptive statistics of diseases of goats brought in at the TVH over the 4 years of the study (January 2011 to June 2014) are displayed in Table 1, showing that mastitis was diagnosed in about 3% of all goats presented at the hospital. The most frequently diagnosed condition was parasitic infestation, of which about 1/3 was due to myiasis (data not shown). Table 1 also clearly demonstrates the importance of Peste des Petits Ruminants (PPR) in goats in Bangladesh as almost 15% of all goats presented at the hospital were diagnosed with this condition. Table 2 displays the distribution of mastitis cases over the 3 seasons. Mastitis was diagnosed most frequently in the rainy season but less in the winter season, whereas gangrenous mastitis was seen less in the rainy season and most in summer and winter. The spatial distribution of cases and controls was similar, as can be seen in Fig. 1.

Table 2
Proportion of cases of mastitis during summer (March–June), rainy season (July–October) and winter (November–February) in female goats (≥1 year of age) at the Teaching Veterinary Hospital of the Chittagong Veterinary and Animal Sciences University, Bangladesh (2011–2014[a]).

Mastitis type	Season							
	Summer		Rainy		Winter		Total	
	N	%	*N*	%	*N*	%	*N*	%
Acute	16	33.3	20	41.7	12	25.0	48	100
Subacute	8	36.4	8	36.4	6	27.3	22	100
Gangrenous	4	50.0	1	12.5	3	37.5	8	100
Chronic	6	33.3	8	44.4	4	22.2	18	100
Total	34	35.4	37	38.5	25	26.0	96	100

[a] Cases included up to July 2014.

3.2. Symptoms of clinical mastitis

In the TVH patient recording system clinical symptoms were recorded for most cases. These recordings were used to describe how the different classes of mastitis in the recording system (acute, subacute, gangrenous and chronic) were defined. Acute mastitis was the most common presentation ($n=48$) and was characterized by the udder showing multiple of the cardinal signs of inflammation (painfulness, heat, redness, swelling and loss of function (i.e., abnormal milk, such as watery or purulent)). In acute cases of mastitis, often the teat canal was blocked. The body temperature was mostly normal, but in about 2/5 of the animals, the body temperature was increased. In subacute ($n=22$) and chronic ($n=18$) cases of mastitis, the same symptoms could be seen, but generally the symptoms were less pronounced. The body temperature was almost always within the physiological range. In chronic mastitis, a firm udder consistency was often noted. Gangrenous mastitis ($n=8$) was characterized by a cold and hard udder, blood in the milk and abnormal skin color.

3.3. Risk factor study

The results of the univariate risk factor analyses are displayed in Table 3. As body weight and age were strongly correlated, suggesting collinearity, we decided to only use age in the multivariable model, because it is biologically easier to interpret than body weight. Multiple records had missing data for breed, rearing system, and body condition score. Table 4 shows that the final multivariable model contained rearing system, body condition score and breed as explanatory variables, with higher OR associated with free-ranging animals, poorer body condition scores, and non-native breeds. No interactions between these variables were significant. The model that used imputed data for the missing values gave similar results as the model without multiple imputation, except for the estimates for semi-intensive rearing systems (the estimate based on imputation was closer to the null-effect) and good body condition score (the estimate based on imputation was farther from the null-effect).

Table 3
Descriptive statistics and univariate analysis of 96 case goats with clinical mastitis and 278 control goats admitted to the Teaching Veterinary Hospital of the Chittagong Veterinary and Animal Sciences University, Bangladesh during 2011–2014.

Variable	Level	*N* Controls	*N* Cases	% Cases	*P*-value
Breed	Black Bengal	145	31	17.6	<0.001
	Cross breeds	44	18	29.0	
	Jamnapari	63	42	40.0	
	Missing	26	5	16.1	
Weight (kg)	<16	134	36	21.2	0.002
	16–24	85	21	19.8	
	>24	59	39	39.8	
Rearing System	Free-ranging	35	12	25.5	0.076
	Semi-intensive	170	70	29.2	
	Intensive	31	3	8.8	
	Missing	42	11	20.8	
Age (months)	<13	69	15	17.9	0.070
	13–24	138	46	25.0	
	>24	71	35	33.0	
Body condition score	Poor	31	27	46.6	0.162
	Fair	60	30	33.3	
	Good	32	24	42.9	
	Missing	155	15	8.8	

Table 4
Results of conditional logistic regression model of the association of rearing system, body condition score (BCS) and breed, with the presence or absence of mastitis in a case-control setting, imputing missing values in records for breed, BCS and rearing system and without multiple imputation (MI). Presented are odds ratios (OR) with a 95% confidence interval (95% CI).

Variable	Level	With MI			Without MI		
		OR	95% CI		OR	95% CI	
			Lower	Upper		Lower	Upper
Rearing system	Free-ranging	Ref.[a]			Ref.		
	Semi-intensive	0.97	0.42	2.23	0.54	0.15	1.92
	Intensive	0.15	0.03	0.71	0.13	0.02	0.85
BCS	Poor	Ref.			Ref.		
	Fair	0.30	0.13	0.66	0.36	0.13	0.98
	Good	0.31	0.12	0.79	0.56	0.17	1.85
Breed	Black Bengal	Ref.			Ref.		
	Cross breeds	1.82	0.82	4.04	1.69	0.54	5.29
	Jamnapari	4.24	2.23	8.08	4.66	1.71	12.70

[a] Ref.: Reference category.

3.4. Treatment of clinical mastitis

Various antimicrobial treatment regimens were used (Table 5). Gentamicin containing preparations being most often used and antibiotics from multiple classes were often combined. The distribution of mastitis types was similar over the different treatment regimes, so no obvious association between therapy and mastitis type was found.

4. Discussion

One aim of this study was to identify risk factors for clinical mastitis in goats brought to the TVH. Mastitis is an often clinically severe disease in goats, but has a low incidence (Bergonier et al., 2003; Contreras et al., 2007). Also in our study, we found that only about 3% of the goats brought into the hospital were diagnosed with mastitis (Table 1). This figure is obviously not necessarily representative of the occurrence of mastitis in the population, as it may be affected by the fact that owners sometimes have not enough time to bring their animal to the hospital, as severe gangrenous mastitis can lead to death within 24 h (Smith and Sherman, 2009). In addition, farmers may have chosen to bring their affected goats to other government veterinary hospitals, or rely on traditional treatments, thereby not contributing to the number of cases observed in our study. Furthermore, milder cases of mastitis may be treated by the owners or by informal health care providers (Sudhinaraset et al., 2013) without consulting a veterinarian. Most cases of mastitis in our dataset were classified by the clinic veterinarians as acute mastitis, but as could be seen from the recorded symptoms, the distinction between acute, subacute and chronic mastitis was not clear and the different classes seem to overlap, suggesting that the clinicians have variable interpretations of types of mastitis. The per-acute presentation of gangrenous mastitis, however, seems to be really distinct from the other classes, but this was seen at low frequency.

The number of mastitic goats was highest in the rainy season and lowest during winter. During the rainy season, food is scarce and many goats are kept inside, intensifying the contact between the animals and worsening the hygienic situation, which may lead to a higher incidence through increased transmission and a weakened immune system. Goats with gangrenous mastitis, however, were presented less frequently during the rainy season. We speculate that this is because during the rainy season, traveling is more difficult and takes more time, limiting people in their ability to bring their animal to the hospital before it dies from the disease.

Several risk factors for clinical mastitis were identified by comparing risk factor status between cases and hospital-based matched controls, using conditional logistic regression. The difficulty of such a study design lies in the fact that controls are not randomly selected from the population but are animals that are presented to the hospital for other reasons. Therefore, these animals may have other characteristics and may not be at the same risk of mastitis as the general population. For instance, these animals may be relatively well fed and cared for, as owners apparently have the means to bring their animals to the hospital, resulting in, for instance, higher BCS. Also, goats from more intensively kept systems may be submitted more or less frequently to the hospital than extensively kept animals. To what extent this has caused a bias is, however, impossible to determine, as we have no estimates on risk factor status and mastitis incidence in the general population. We tried to limit selection bias by not using all hospital patients as the base for drawing controls, but only using does of one year and older, so animals that could be in lactation and therefore be at risk for getting mastitis.

Goats from intensive farming systems were less likely to be cases than goats from free-ranging or semi-intensive farming systems. This is surprising, as more intensive goat farming has been associated with a higher prevalence of intramammary infection (Kyozaire et al., 2005). Similarly, in dairy cattle, more intensive farming has been associated with higher veterinary usage (Richert et al., 2013). In the urban Chittagong region, there are educated and econom-

Table 5
Frequency of antibiotics prescribed against different clinical mastitis cases in goat registered at Teaching Veterinary Hospital of the Chittagong Veterinary and Animal Sciences University, Bangladesh (2012–2014).

Classes of antibiotics	Antibiotic	Mastitis type					
		Acute	Subacute	Chronic	Gangrenous	All	%
Aminoglycosides	Gentamicin	14	9	6	2	31	37
Aminoglycosides + amino-penicillines	Gentamicin + amoxicillin	12	5	2	3	22	26
Aminoglycosides + sulphonamides + diaminopyrimidines	Gentamicin + sulphadimidine + trimethoprim	9	0	0	0	9	11
Aminoglycosides + penicillins + tetracyclines	Streptomycin + procainpenicillin + tetracycline	1	3	4	0	8	10
Amino-penicillines	Amoxicillin	3	0	1	0	4	5
Penicillins	Benzylpenicillin	1	0	0	2	3	4
Tetracyclines	Tetracycline	2	0	0	0	2	2
Tetracyclines + Sulphonamides	Tetracycline + sulphadimidine	1	0	1	0	2	2
Sulphonamides	Sulphadimidine	0	1	0	0	1	1
Cephalosporins	Ceftriaxone	1	0	0	0	1	1
Aminoglycosides + Amino-penicillines + Sulphonamides	Gentamicin + amoxicillin + sulphadimidine	1	0	0	0	1	1
Total		45	18	14	7	84	100
No treatment recorded		3	4	4	1	12	14

ically solvent people who rear small numbers of hobby-goats in intensive systems. These goats are better managed and get higher quality feed. This may explain the negative association between intensive rearing system and case status.

We found that the autochthonous goat breed (Black Bengal goats) had less mastitis than the Jamnapari breed or cross breeds, suggesting a better adaptation to the local environment of the native breed. The about 4 times higher odds of mastitis for Jamnapari compared to Black Bengal goats is likely also caused by the fact that Jamnapari goats are in part kept for milk production. Therefore these animals may have longer lactations and therefore are longer at risk of mastitis. Furthermore, the udder conformation of this breed may predispose it for mastitis (Megersa et al., 2010; Koop et al., 2013).

Poor body condition score was clearly associated with increased odds of mastitis. This is in line with previous work in goats (Megersa et al., 2010) and cows (Mungube et al., 2004; Sarker et al., 2013), but in contrast with findings by Tolosa et al. (2015), who showed increased odds of intramammary infection with higher body condition score. The exact mechanism leading from poor body condition score to increased mastitis incidence is unclear. A poor body condition score may negatively affects the goat's immune system, but in cattle, protein and energy deficient diets had limited effect on the immune responsiveness (Fiske and Adams, 1985; Doherty et al., 1996). Another possible mechanism may be that goats with a poor body condition score produce less milk for their kids. In sheep, it has been suggested that a relative milk shortage induced by multiple suckling lambs leads to more vigorous suckling and teat damage which predisposes the udder to mastitis (Waage and Vatn, 2008; Koop et al., 2010a). For a substantial number of cases, information on body condition score was missing. By imputing body condition score for these variables, we were able to take these cases into account, thereby preventing substantial loss of observations in the multivariable model. Moreover, it allowed us to assess to some extent the potential bias associated with these missing values. In the model using imputed data, the effect of body condition score was more pronounced than in the model without imputation and confidence intervals were smaller. As missingness was clearly associated with case or control status, we cannot assume that the missingness was completely at random (MCAR), but we assume that the missing data were missing at random (MAR), although this cannot be ascertained with the data at hand (Schafer and Graham, 2002). As we have no reason to assume that goats with a poor body condition score are more or less likely to have missing values than goats with a fair or good body condition score (which would be not missing at random (NMAR)), we believed that using multiple imputation was justified. Moreover, incorrectly assuming the missingness to be MAR was shown in many cases to only have a limited impact on the estimates (Collins et al., 2001).

In this observational study, we found that treatments used for clinical mastitis in goats were highly variable (Table 5). At least 11 different treatment regimens were employed, with no clear association between type of mastitis and treatment regimen. Apart from this, goats were treated with a considerable variation in supportive therapy, such as analgesics, vitamins and antihistamine preparations. This seems to suggest that no systematic approach for dealing with goat udder health problems exists. The most prevalent causes of mastitis in goats are gram-positive cocci, although also gram-negative bacteria have been found (Contreras et al., 2003; Islam et al., 2012). This may explain the generally broad spectrum therapies that are employed and the general preference for gentamicin containing products, which has activity against many aerobic gram-negative species as well as against gram-positive staphylococci. The world health organization regards gentamicin as a critically important antimicrobial for human medicine (WHO, 2012). Therefore, it may be better to treat animals with other antibiotics, which may be equally effective. This, however, requires a good understanding of the distribution of pathogens responsible for mastitis in goats in the Bangladesh specific situation, and their antimicrobial susceptibility. If that information was present, it would be possible to create evidence-based guidelines for treatment of mastitis, which would contribute to more effective and possibly cheaper therapies, and would contribute to prevention of antimicrobial resistance problems in humans.

The overarching aim of this study was contribute to improved prevention and control of mastitis in goats in a small scale farming situation. To reach this goal, we identified risk factors for mastitis and also described what therapies are currently being used for mastitis in goats in the setting we studied. With this, guidelines could be made for effective goat rearing in this specific setting and also guidelines for therapy could be established. However, in circumstances like the situation we described in Bangladesh, the major challenge will be in effectively implementing these guidelines. This will require a multidisciplinary approach and should be the subject of future studies.

5. Conclusions

Clinical mastitis in goats in Bangladesh is associated with a free-ranging rearing system, a poor body condition score and non-native breeds. Multiple imputation proved a valuable technique to model these effects in a matched case-control dataset with multiple missing records. Treatment of mastitis in this animal hospital was non-systematic and showed frequent use of antimicrobials that are

considered critically important for human medicine, showing the need for evidence-based veterinary medicine.

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