Page 236 to 246

Volume 1 • Issue 6 • 2018

Research Article

Multidisciplinary Advances in Veterinary Science

ISSN: 2573-3435

Study on Udder Health Status and Antibacterial Susceptibility of Common Bacterial Isolates from Clinical and Subclinical Mastitis in North Gondar Zone, Ethiopia

Teshome Setegn[1]* and Takele Adugna[2]

[1]*College of Veterinary Medicine and Animal Science, University of Gondar, P.O.Box 196, Gondar, Ethiopia*

[2]*College of Veterinary Medicine and Animal Science, Department of Clinical Medicine University of Gondar, P.O.Box 196, Gondar, Ethiopia*

***Corresponding Author:** Teshome Setegn, College of Veterinary Medicine and Animal Science, University of Gondar, P.O.Box 196, Gondar, Ethiopia.

Received: December 23, 2017; **Published:** January 09, 2018

Abstract

A cross sectional study was conducted from October 2013 to April 2014 in selected districts of North Gondar Zone of the Amhara Regional State with the objective of assessing the prevalence of the most frequently encountered bacterial pathogens from mastitis cows, udder and evaluates their antimicrobial susceptibility profile. The study animals were lactating local zebu and crossbred (Holstein X zebu) cows'. Structured questionnaire, bacterial isolation and identification procedures and antimicrobial susceptibility testing were employed as a data collection tools. Of 100 farm and individual cow owners participated in this study, 90%, 80% and 50% of them reported that mastitis occurred due to dirty bedding, tick infestation and sleeping on hot and humid ground, respectively.

Analysis of risk factors indicated that climatic condition, breed, body condition, milk yielding potential, number of lactation, stage of lactation, tick infestation and physical injury were the main risk factors for the occurrence of mastitis in the study setting, ($p < 0.05$). The bacteriological investigation part revealed that staphylococcus species are the most frequently isolated bacteria (84.6%) followed by *E.coli* (6.7%) and bacillus species (3.4%). *S.aureus* and *S.epidemitis* were most commonly isolated species of *Staphylococcus* genus taking the leading and second position, respectively. The highest resistance pattern was observed in *Staphylococcus* species; particularly *S.aureus*, even to the level of 100% for some antimicrobials commonly used both in veterinary and human medicine practice. In the current study ,the worst antimicrobial resistance exhibited was against Penicillin G which was 100% for all of bacterial species exposed to it .As a conclusion ,mastitis is highly prevalent in the study area orchestrated by different risk factors and implicated by different drug resistance bacteria. Antimicrobial susceptibility tests should be carried out before any antimicrobial treatment regimen for any mastitis cases.

Keywords: *Antimicrobial susceptibility; Bacterial species; Identification; Mastitis; Risk factor*

Volume 1 Issue 6 January 2018

Citation: Teshome Setegn and Takele Adugna. "Study on Udder Health Status and Antibacterial Susceptibility of Common Bacterial Isolates from Clinical and Subclinical Mastitis in North Gondar Zone, Ethiopia". *Multidisciplinary Advances in Veterinary Science* 1.6 (2018): 236-246.

Introduction

Ethiopia has the largest cattle population in Africa with an estimated population of 49.3 million [1]. But the per capita consumption of milk is only 19 kg per year [2] due to the low production level coupled with nutrition poor husbandry system and diseases.

Mastitis is one of the most frequent diseases in dairy cattle that hinder milk production in dairy cattle. The different forms of infectious mastitis occur according to the host response and to the microorganisms which cause the infection. The occurrence and type of symptomatology are related to the pathogenicity of the micro-organism and to its ability to invade tissues, as well as to the resistance of the mammary gland. These factors determine the severity of the symptoms that can vary from increased cell counts with no macroscopic changes in milk to progressive fibrosis or the occurrence of severe toxemias [3].

For the treatment of mastitis different antibiotic therapies are quite commonly implemented Widespread antibiotic usage or misuse exert a selective pressure that acts as a driving force in the development of antibiotic resistance. Antibiotic resistance patterns may vary locally or regionally, so surveillance data needs to be collected. Patterns can change rapidly and they need to be monitored closely because of their implications for public health and as an indicator of appropriate or inappropriate antibiotic usage in a given area [4].

Antimicrobial resistance monitoring will help to review the current status of antimicrobial resistance and helpful in minimizing the consequence of drug resistance, limit the emergence and spread of drug resistant pathogens. Several studies have been conducted to estimate different forms of clinical mastitis in Europe [5-7], North America [8] and Africa [9,10].

A study conducted by Almaw., *et al*. [11] on selected small holder dairy farms in Gondar town revealed that the average incidence rate of clinical bovine mastitis of 21.26 per 100 cow-years at risk. The same authors also reported the prevalence rate of subclinical mastitis of 25.22%. Except these attempts to determine the prevalence of mastitis either clinical or subclinical mastitis, no study was conducted to assess the overall health status of cow's udder and the susceptibility of common bacterial isolates from mastitis in the zone.

General Objective of this study: to improve milk production through control and prevention of diseases and problems associated with udder and teat.

Specific Objectives: To assess the extent of diseases and problems associated with udder and teat and to assess antibacterial susceptibility of common bacterial isolates in North Gondar.

Methodology

The Study Area: the study was conducted from October 2013 to April 2014 in North Gondar Zone of the Amhara regional state. The zone is bordered by Lake Tana, West Gojjam zone and the Benishangul-Gumuz regional state in the South, Sudan in the West, Tigray regional state in the North, Wag Hemra zone in the East and South Gondar zone in the Southeast. The altitude ranges from 4620 ms in the Semiemountain in the North to 550 ms in the West. The rainfall varies from 880 mm to 1772 mm.

The minimum and the maximum temperatures are in the order of -10°c in the highland and 44.5°c in lowlands. The zone has an estimated total population of 3,083,347 of whom 1,562,977 were males and 1,520,370 were females. The zone has an estimated area of 48, 204.39 square kilometres. Many of the dwellers either rural or urban are involved in animal production and the zone has an estimated cattle population of 1,936,543. Many other non-food producing animals are also available in the zone (1). Mixed cereal-based agriculture and livestock farming are well practiced in the zone.

Study population and Data Collection Methods: The study animals were lactating local zebu and Holstein x Zebu crossbred cows in the study area. Three districts of the zone were selected from each agro-ecological zone (Dega, W/dega and Kolla). Structured and semi-structured questionnaires were prepared to collect the necessary information. Data were collected by interviewing (group and

individual). Farms in the selected district were selected randomly for follow up and data collection. Training was given in selected and voluntary farms so as to get informed any problem occurring on the teat and/or on the udder.

Data collection techniques

Diagnosis of clinical mastitis: Clinical cases were diagnosed physically at the quarter level based on visible and palpable signs (hard and swollen quarter, and heat) as previously described [12]. In addition, milk from each quarter were withdrawn and examined for any change (watery secretions, clots in milk, and blood-tinged secretions). The size and consistency of mammary quarters were inspected for the presence of any anatomical malformation, such as disproportional symmetry, swelling, firmness, and blindness.

Diagnosis of subclinical mastitis: Diagnosis of subclinical mastitis was carried out by using indicator paper test. It was carried out by adding a drop of milk sample to the test paper and observing the color change of the paper. If yellow color of the paper is not changed and remained as it is, such sample was considered as negative. A change of color from yellow to green or bluish green was recorded as positive.

Questionnaire survey: Information has been collected from farm owners and individual cow owners with structured and semi structured questionnaire. The contents of the questionnaire are listed in annex 2 of my paper.

Bacteriological examination of milk sample

Udder and teat preparation: The udder of the cow to be sampled were thoroughly cleaned with water and dried with a clean towel. Udder and teats were cleaned from the far side to the near side in order to avoid contamination during cleaning. After cleaning the teats with swabs .they were soaked with 70% ethyl alcohol. The teats are then allowed to be dried.

Milk Sample Collection and Transportation: Milk samples were collected from points considered to be associated with contamination (critical control points: CCPs). The CCPs were the teat during milking (CCP-1), milking personnel (CCP-2), and sampling bottles), transportation containers up on arrival at the processing plant (CCP-3), before taking sample, I have washed my hands and disinfected with 70% ethyl alcohol. The milk samples containers were properly washed and sterilized in autoclave at 121ºc for 15 minutes and are disinfected with 70% ethyl alcohol.

The first three to four streams of milk discarded, and then, 5-10 ml of milk was collected from each teat aseptically in separate universal bottles. The tubes were sealed properly and transported on ice to Veterinary Microbiology laboratory at Gondar University, where samples were cultured immediately or kept in a refrigerator at 4°C for a maximum of 24 hours until cultured.

Bacteriological isolation from milk sample: For bacteriological examination, a loop full of milk sample were streaked on tryptose blood agar base enriched with 7% defibrinated sheep blood and MacConkey agar plates using the quadrant streaking method. Both agar plates were incubated aerobically at 37°C for 24-48 hours and examined for characteristic bacterial colonies. The colonies were characterized by their color, their growth characteristics, and their hemolytic patterns.

Pure culture colonies were selected and sub-cultured on general purpose medium, nutrient agar, and incubated aerobically at 37°C for 24-48 hours for further biochemical identification. After this incubation on nutrient agar bacteria were identified according to their Gram reaction and morphology. Further identification of the organisms was done by implementing biochemical tests, catalase, oxidase, IMVIC tests. In addition, mannitol salt agar was used to differentiate *Staphylococcus aurous* from other *Staphylococcus* spp. Glucose fermentation test were used to identify those gram negative non lactose fermenter and oxidase positive bacteria. It is used to differentiate pseudomonas and *aeromonas* bacteria.

Antimicrobial susceptibility test: Antimicrobial susceptibility test was conducted on selected bacterial isolates; especial emphasis was given to those isolates which are frequently recovered during the study period. The isolates were tested for their susceptibility to antimicrobials using the Kirby–Bauer disk diffusion method [13]. The cutoff values for the evaluation of the susceptibility of isolates were based on the inhibition zone as described by Haftu., *et al*. [14].

As plating medium, Mueller–Hinton agar was used. Antibiotic-impregnated paper discs and plates were incubated at 37°C for 16-18 hours. The antimicrobial susceptibility was scored as resistant, intermediate, and sensitive based on diameter of pattern. The antibiotic discs that we used for the susceptibility test are cefoxitin, amikacin, clindamycin, doxicycline, naldixic acid, vancomycin and amoxaciline. Minimum inhibitory concentration of the discs were determinedfor the bacteria isolates using the broth microdilution method according to the approved standard of the clinical and laboratory standard institute. Susceptibility interpretation criteria were based on the clinical and laboratory institute guidelines.

Data Analysis: the data were recorded in Excel spreadsheet and analyzed using appropriate statistical software (SPSS)

Results

Questionnaire Survey result: According to the information gathered from farm owners and individual cow owners in the study area (Gondar , Dabat and Kolladiba towns ;the average herd size was 38, 4, and 5, respectively. Most farm owners clean the cows sleeping area once early in the morning but few farm owners in Gondar town clean twice a day. Most of the owners used to seek modern treatment for their cows acquired mastitis and all of the three study areas. But few peoples in Kolladiba were using herbal medicine to treat mastitis cows. From a total of 100 farm and individual cow owners participated in this study, about 50% they said mastitis was occurred when the cow sleeps in hot and humid ground.

The other factor that about 20% of the cow owners answered as problem in their dairy business was tick infestation. Participants responded that when tick infested the udder of the cow, it results in swelling of the udder, abscess formation and leads to blinds of teats and the udder quarters become atrophied and stop functioning. In Kolladiba town (Dembia district), 80% of cow owners said that teat blindness and udder atrophy is associated with tick infestation.

It was observed that dirty bedding area was also one of the factors that affect the udder health. This was a major problem in Gondar town. About 90% of farm owners said mastitis is occurred when the cows bedding area is dirty.

Physical injury associated with calf biting and milking personnel nail is also a factor for inflammation of udder and crackling of teats. About 13% of the questionnaire result shows calf biting during suckling and milkers nail during milking causes crackling of teats and injury of teats.

Identification of bacteria: The most common bacterial pathogens isolated from clinical and subclinical mastitis were staphylococci species followed by *Enterobacteriacea* species that are isolated from clinical and subclinical mastitis are summarized in table 1 below. From clinical mastitis cows *S.aureus* (52.4%), *S.epidemitis* (22%) and *E.coli* (6.1%) and in subclinical mastitis *S.epidemitis* (53.2%), *S.aureus* (33.3%) and *E.coli* (7.1%) are the leading causes.

Species isolated	Types of mastitis		Total
	Clinical	Subclinical	
S.aureus	43 (52.4%)	42 (33.3%)	85 (41%)
S.epidemitis	18 (22%)	67 (53.2%)	85 (41%)
Other staph spp	4 (4.9)	2 (1.6%)	6 (2.9%)
E.coli	5 (6.1%)	9 (7.1%)	14 (6.7%)
Klebsiella	2 (2.4%)	0 (0%)	2 (0.96%)
Streptococci	2 (2.4)	2 (1.6%)	4 (1.9%)
Bacilli	6 (7.3%)	1 (.8%)	7 (3.4%)
Citrobacter	1 (1.2%)	2 (1.6%)	3 (1.4%)
Aeromonas	1 (1.2%)	1 (.8%)	2 (0.96%)
Total	82 (39.4%)	126 (60.6%)	208 (100%)

Table 1: *Bacterial species isolated from mastitis affected udder.*

From clinically affected animals, the quarter affected and the pathogen isolated from it is summarized by the following table

Species isolated	Affected udder						
	Right rear	Right front	Left rear	Left front	Right rear and left rear	Left rear and left front	All quarter affected
S.aureus	7 (87.5%)	8 (47.1%)	15 (60%)	1 (16.7%)	4 (44.4%)	3	3
S.epidemitis	1 (12.5%)	6 (35.3%)	3 (12%)	3 (50%)	1 (11.1%)	2	2
Other Staph Spp	0	1 (5.9%)	1(4%)	0	1 (11.1%)	1	0
E.coli	0	0	2 (8%)	0	2 (22.2%)	0	1
Klebsiella	0	0	2 (8%)	0	0	0	0
Streptococci	0	1(5.9%)	0	1 (16.7%)	0	0	0
Bacilli	0	1 (5.9%)	1 (4%)	0	1 (11.1)	0	2
Citrobacter	0	0	1 (4%)	0	0	0	0
Aeromonas	0	0	0	1 (16.7%)	0	0	0
Total	8 (100%)	17 (100%)	25 (100%)	6 (100%)	9 (100%)	6 (100%	8 (100%)

Table 2: *Bacteria species isolated with their frequency and respective quarter.*

From animals that were clinically ill and previously treated and not treated, the pathogens isolated are summarized by the following table.

The current investigation revealed also that there was statistically significant difference (p = 0.002) Dega, Woyinadega and Kolla localities. From the species isolated from Kolla, *S.epidemitis* were relatively highly prevalent. In Dega and Woyina Dega climatic conditions S.aureus was more frequently isolated. In general mastitis were prevalent in cross-breeds than indigenous cows (p = 0.000) in this study. In addition, those high milk yielding cows were more prone to mastitis than low yielding cows (p = 003). Nevertheless, but udder cleanness was not found to be a clear risk factor for mastitis (p = 0.5). The result of this study indicated that there was statistically significant association (p = 0.000) among number of lactations.

Species isolated	Previously treated		Total
	Yes	No	
S. aurous	20 (58.8%)	23 (48%)	43
S.epidemitis	8 (23.5%)	10 (21%)	18
Other Staph Species	2 (5.9%)	2 (4.2%)	4
E.coli	1 (2.94%)	4 (8.3%)	5
Klebsiella	1 (2.94%)	1 (2.1%)	2
Streptococci.	0 (0%)	2 (4.2%)	2
Bacilli	2 (5.9%)	4 (8.3%)	6
Citrobacter	0 (0%)	1 (2.1%)	1
Aeromonas	0 (0%)	1 (2.1%)	1
Total	34 (100%)	48 (100%)	82

Table 3: *pathogens that were isolated from previously treated or not treated animals.*

	Isolated pathogens								
Factors	***S.aureus***	***S.epidemitis***	**other staphs**	***E.coli***	***Klebsiella***	***Strept.spp***	***Bacilli***	***Citrobacter***	***Aeromonas***
Climate (X^2 = 37.811, p = 0.002)									
Dega	14	4	0	5	0	1	0	0	0
W/dega	51	51	5	1	2	1	3	3	0
Kolla	20	30	1	8	0	2	4	0	2
Breed (X^2 = 31.44, P = 0.000)									
Local	12	19	0	10	0	1	4	0	1
Cross	73	66	6	4	2	3	3	3	1
Milk yield in liter (X^2 = 47.086, P = 0.003)									
1-5	18	21	0	10	0	1	4	0	2
6-10	28	22	5	3	1	2	1	0	0
11-15	19	29	1	1	1	1	1	2	0
16-20	20	13	0	0	0	0	1	1	0
Number of lactation (X^2 = 42.108, p = 0.000)									
1-4	75	66	4	6	1	3	5	1	2
5-8	7	18	2	4	1	1	2	2	0
>9	3	1	0	4	0	0	0	0	0
Stage of lactation (X^2 = 42.108, p = 0.000)									
1st	39	25	2	4	1	0	3	1	1
2nd	31	42	3	7	0	3	4	1	0
3rd	15	18	1	3	1	1	0	1	1
Udder cleanness (X^2 = 7.332, p = 0.501)									
Clean	38	37	4	7	2	1	4	1	2
Dirty	47	48	2	7	0	3	3	2	0

Body condition score, (X^2 = 71.123, P = 0.000)									
Poor	5	5	0	4	0	1	2	0	1
Medium	61	61	4	8	1	2	3	3	1
Good	19	19	2	2	1	1	2	0	0

Table 4: *Factors that affect mastitis.*

Antimicrobial susceptibility test: From 208 isolated bacterial species; antimicrobial susceptibility test was done for 200 species. Staphylococci bacteria have developed resistance almost too all antibiotics that are used in this study. They have developed 100% resistance to penicillin G. But they are susceptible to Doxycycline and Amikacine (71.4% and 62.22% respectively. The antibiotics and their sensitivity result for bacterial species is summarized by the following table.

Antimicrobial resistance pattern: The mastitis pathogens are developing resistance for most antimicrobials. The antimicrobial resistance pattern for clindamycinis 78.5%, vancomicine 74.5%, Amoxaciline 61%, and cefoxitine 60.5%. The antimicrobial resistance pattern for each antimicrobial is summarized by the following table.

Isolate	Fox			Ak			Da			Do		
	R N (%)	I N (%)	S N (%)	R N (%)	I N (%)	S N (%)	R N (%)	I N (%)	S N (%)	R N (%)	I N (%)	S N (%)
S. aureus	67 (68.3%)	4 (4.08%)	27 (27.54%)	31 (31.62%)	6 (6.12%)	61 (62.22%)	81 (82.62%)	8 (8.16%)	9 (9.18%)	20 (20.4%)	8 (8.16%)	70 (71.4%)
S. epidemitis	40 (58%)	1 (1.45%)	28 (40.6%)	15 (21.75%)	4 (5.8%)	50 (72.5%)	55 (79.75%)	5 (7.25%)	9 (13.05%)	13 (18.85%)	2 (2.9%)	54 (78.3%)
Other Staph	3 (60%)	0 (0%)	2 (40%)	1 (20%)	1 (20%)	3 (60%)	3 (60%)	1 (20%)	1 (20%)	1 (20%)	1 (20%)	3 (60%)
E.coli	2 (15.38%)	1 (7.69%)	10 (76.9%)	0 (0%)	0 (0%)	13 (99.97%)	5 (38.45%)	3 (23.0%)	5 (38.45%)	0 (0%)	0 (0%)	13 (99.97%)
Klebsiella	2 (50%)	0 (0%)	2 (50%)	1 (25%)	0 (0%)	3 (75%)	3 (75%)	0 (0%)	1 (25%)	0 (0%)	0 (0%)	4 (100%)
Streptococci	5 (71.45%)	0 (0%)	2 (28.58%)	1 (14.29%)	0 (0%)	6 (85.74%)	6 (85.74%)	0 (0%)	1 (14.29%)	1 (14.29%)	0 (0%)	6 (85.74%)
Bacilli	2 (66.6%)	0 (0%)	1 (33.3%)	0 (0%)	0 (0%)	1 (99.9%)	3 (99.9%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	3 (99.9%)
Citrobacter	0 (0%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)	1 (100%)	1 (100%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (100%)
Aeromonas												

Isolated spp	Na			P			Va			AML		
	R	I	S	R	I	S	R	I	S	R	I	S
S. aureus	64 (66.56%)	8 (8.16%)	26 (26.52%)	98 (99.96%)	0 (0%)	0 (0%)	69 (70.38%)	7 (7.14%)	22 (22.44%)	60 (61.2%)	0 (0%)	38 (38.76%)
S. epidemitis	32 (46.45%)	7 (10.15%)	30 (43.5%)	69 (100%)	0 (0%)	0 (0%)	54 (78.3%)	5 (7.25%)	10 (14.5%)	45 (65.25%)	0 (0%)	24 (34.5%)
Other staph spp	2 (40%)	1 (20%)	2 (40%)	5 (100%)	0 (0%)	0 (0%)	4 (80%)	1 (20%)	0 (0%)	4 (80%)	0 (0%)	1 (20%)

E. coli	0 (0%)	2 (15.38%)	11 (84.59%)	13 (99.97%)	0 (0%)	0 (0%)	11 (84.59%)	1 (7.6%)	1 (7.69%)	6 (46.14%)	0 (0%)	5 (3.83%)
Klebsiella	3 (75%)	0 (0%)	1 (25%)	4 (100%)	0 (0%)	0 (0%)	2 (50%)	0 (0%)	2 (50%)	1 (25%)	0 (0%)	3 (75%)
Streptococci	2 (28.58%)	1 (14.29%)	4 (28.58%)	7 (100.00%)	0 (0%)	0 (0 %)	6 (85.74%)	0 (0%)	1 (14.29%)	3 (42.87%0	0 (0%)	5 (7.16%)
Bacilli	1 (33.3%)	1 (33.3%)	1 (33.3%)	3 (99.9%)	0 (0%)	0 (0%)	2 (66.6%)	0 (0%)	1 (33.3%)	2 (66.6%)	0 (0%)	1 (33.3%)
Citrobacter	0 (0%)	0(0%)	1 (100%)	1 (100%)	0 (0%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)	1 (100%)	0 (0%)	
Aeromonas												

Antimicrobial	Resistant	Intermediate	Susceptible	Total
Fox	121 (60.5%)	6 (3%)	73 (36.5%)	200 (100%)
Ak	49 (24.5%)	11 (5.5%)	140 (70%)	200 (100%)
Da	157 (78.5%)	17 (8.5%)	26 (13%)	200 (100%)
Do	35 (17.5%)	11 (5.5%)	154 (77%)	200 (100%)
Na	104 (52%)	20 (10%)	76 (38%)	200 (100%)
P	200 (100%)	0 (0%)	0 (0%)	200 (100%)
Va	149 (74.5%)	14 (7%)	3 7(18.5%)	200 (100%)
Aml	122 (61%)	0 (0%)	78 (39%)	200 (100%)

Discussion

This study has been conducted to investigate the status of udder health (normal and abnormal, to know the factors that affect the udder health, to isolate the most common bacterial species from clinical and subclinical mastitis and to make antimicrobial susceptibility test on the most bacterial isolates. During this study period, from a total of 208 isolated bacterial species, 82 (39.4%) and 126 (60.6%) were isolated from clinical and subclinical mastitis cases respectively.

The main pathogen that isolated during the study was staphylococcus species (94% of the isolated pathogens), of which *staphylococcus aurous* is the main pathogen for clinical and subclinical mastitis. This study agrees with studies conducted by Mullenberger and Kirk at university of California Davis [15]; at Egypt and Middlenton who studied *S.aureus* mastitis at university of Missouri in [16]. Mekibib., *et al.* at Hawassa University, Ethiopia has studied the prevalence ,risk factors and major pathogens of bovine mastitis .In his study he had found that S.aureus as the most prevalent mastitis pathogen (47.1%) [17].This study nearly agrees with my study (40.5%).

When this study result compare the isolation rate *S.aureus* from clinical and subclinical mastitis with research done on dairy cattle mastitis in and around Sebeta, Ethiopia by Hundra., *et al.* [18], from clinical mastitis is higher (52.4%) but in subclinical mastitis is lower (33.3%). the isolation rate by hundra., *et al.* for S.aureus from clinical and subclinical mastitis was 34.04% and 49.3% respectively.

From clinically affected udder quarter, the most prevalently affected udder quarters are the rear quarters, especially the left rear quarter. The reasons for higher hind quarter involvement might be due to more frequent exposure to dung and urine, larger capacity and mass, greater vulnerability to direct trauma and relatively more closeness to the floor as compared to four quarters. When quarters are individually compared the left rear and right front quarters are most affected.

This may be due to contamination from the operators hands which are assumed as less hygienic (without proper washing and disinfection [19]. It might be due to greater production capacity of hind quarter and ease of first grasping by milkers hand in case of right front quarter [18]. Climate was a risk factor for mastitis pathogen isolation with (p = 0.002). This because the climate, humidity, temperature and rain fall affects mastitis pathogens (3).

Breed found as highly significant factor with (p = 0.000) for mastitis. Cross breeds are much more vulnerable to mastitis this is similar with Hundra., *et al*. (18) and Doherr., *et al*. [20] finding. Body condition score is also found to be a highly significant factor for mastitis (p = 0.000). In this study, the bacterial isolation is much more prevalent in lactating cow with medium body condition score.

Milk yield was a significant risk factor for mastitis with (p = 0.003) this is because higher milk yielding cows has been found more susceptible owning to position of teat and udder and anatomy of teat canal, making them more prone to injury and due to less efficient phagocytic cells in higher yielding cows associated with dilution (18). Stage of lactation is not a significant factor for mastitis (p = 0.602). But the 2nd stage of lactation is more prevalently affected.

During questionnaire survey respondents suggested hot and humid environment is major facilitating factor for mastitis. This may be due to heat and humidity increase the pathogen load in the environment (field or housing) resulting in a greater incidence of mastitis in warm weather [21]. Tick infestation is also factor for mastitis this because physical injury of the teat and udder is a factor for mastitis since bacteria can easily enter and multiply and cause inflammation of udder and this result also similar with Sefinew's., *et al*. [22] study.

Staphylococci bacteria have developed resistance for much of antimicrobials used by this study. *S.aureus* has developed resistance for cefoxitin (67%), Naldixic acid (66%), penicillin (99.9%), vancomycine (70.78%) and amoxaciline (61.2%) but it had shown better susceptibility to Doxicycline and Amikacine. *S.epidemit* is had developed resistance for cefoxitin (58%), clindamycin (79.75%), penicillin (100%), vancomicine (54.3%) and amoxyline (65.25%).

Generally most antimicrobials used in this study had developed resistance. Their resistance pattern is increasing (cefoxitine 60.5%, clindamicin 78.5%, vancomycine 74.5%. The reason for development of antimicrobial resistance of staphylococci is production of Ɓ lactamases, enzymes that inactivate the drugs by hydrolyzing the Ɓ lactam ring [23]. Inappropriate use of drugs is main factor for resistance development. Doxycycline has been found a better antimicrobial to treat most mastitis pathogens. This is due to the property of Doxycycline in that it is much more lipophilic, resulting in higher tissue penetration, larger volume of distribution, and better overall antimicrobial properties (23).

Conclusion and Recommendations

Mastitis is one the most frequent diseases in dairy cattle that hinder milk production. It affects dairy cows and remains the most economically important disease of dairy industries around the world. It is characterized by physical, chemical and microbiological changes in the milk and pathological changes in the glandular tissues of the udder. It results teat blindness and culling of cows without the appropriate age of the cow. Contagious type of mastitis is the major problem that is mostly associated with the cow's dirty environment that creates a conducive environment for multiplication. These contagious mastitis pathogens have developed resistance for most antimicrobials. Using antibiotics is not an ideal solution rather than the problem they cause within the milk (withdrawal for the treatment dose, contamination from antibiotic residue. Using antibiotics cannot reduce the incidence of mastitis. Problems associated with resistance or even ineffectiveness is quite real in in the case of mastitis caused by S.aureus. Holstein Friesian cattle's are much more prone to mastitis than the local zebu breeds.

Based on the above conclusion, the following recommendations are forwarded:-

- Preventing new infection by focusing on managemental efforts on milking technique and hygiene Proper milking hygiene, using disinfectants:
- Follow the proper milking order of milk cows in the herd.
- Since local zebu breeds are more resistance to mastitis than Holstein Friesian cows the hybrid of local zebu and Holstein Friesian should be used for dairy industry to reduce the susceptibility of exotic breeds.

References

1. CSA. "Federal Democratic Republic of Ethiopia". Central Statistics Agency, Agricultural sample survey report on livestock and livestock characteristics. Addis Ababa, Ethiopia (2008).
2. Redda T., *et al*. "Small-scale milk marketing and processing in Ethiopia". Antibacterial Activity of Ultra Bath (2012).
3. Radostits OM., *et al*. "A textbook of the diseases of cattle, horses, sheep, pigs and goats". *Veterinary Medicine*, New York, USA (2007).
4. Benites NR., *et al*. "Etiology and Histopathology of Bovine Mastitis of spontaneous occurrence". *Journal of Veterinary Medicine* 49.8 (2002): 366-370.
5. Barkema HW., *et al*. "Incidence of clinical mastitis in dairy herds grouped in three categories by bulk milk somatic cell counts". *Journal of Dairy Science* 81.2 (1998): 411-419.
6. Peeler EJ., *et al*. "Risk factors associated with clinical mastitis in low somatic cell count British dairy herds". *Journal of Dairy Science* 83.11 (2000): 2464-2472.
7. Barnouin J., *et al*. "Dairy management practices associated with incidence rate of clinical mastitis in low somatic cell score herds in France". *Journal of Dairy Science* 88.10 (2005): 3700-3709.
8. Sargeant JM., *et al*. "Clinical mastitis in dairy cattle in Ontario: frequency of occurrence and bacteriological isolates". *The Canadian Veterinary Journal* 39.1 (1998): 33-38.
9. Karimuribo ED., *et al*. "Basic Facts of Mastitis in dairy animals: A Review". *Pakistan Veterinary Journal* 26.4 (2006): 204-208.
10. Kivaria FM., *et al*. "Risk factors associated with incidence rate of clinical mastitis in smallholder dairy cows in Dares Salaam Region, Tanzania". *The Veterinary Journal* 173.3 (2007): 623-629.
11. Almaw G., *et al*. "Incidence rate of clinical bovine mastitis in selected smallholder dairy farms in Gondar town, Ethiopia". *Ethiopian Veterinary Journal* 16.1 (2012).
12. Cappucino JG and Sherman N. "Microbiology: a laboratory manual, 7th edn. Pearson crossbred dairy herds in urban and periurban production systems in the central highlands of Ethiopia". *Tropical Animal Health and Production* 33 (2005): 85-89.
13. Haftu R., et al. "Prevalence, bacterial causes, and antimicrobial susceptibility profile of mastitis isolates from cows in large-scale dairy farms of Northern Ethiopia". *Tropical Animal Health and Production* 44.7 (2012): 1765-1771.
14. Mullenberger R and Kirk J. "Mastitis control programfor s.aureus infected dairy cows". University of California davis (2001).
15. Middlenton JR. S.aureus mastitis: Have we learned anything in the last 50 years, NMC Regional Meeting Proceeding, University of Missouri, and Columbia, Missouri, USA (2013).
16. Mekibib B., *et al*. "Bovine Mastitis Prevalence, Risk Factors, and Major Pathogens in dairy farms of Holeta town, Central Ethiopia". *Veterinary world* 3.9 (2010): 397-403.
17. Hundra, S., *et al*. "Dairy cattle mastitis in and around Sabetha, Ethiopia". *The International Journal of Applied Research in Veterinary Medicine* 3.4 (2005): 332-338.
18. Shittu A., *et al*. "Subclinical mastitis and associated risk factors on lactating cows in Savannah region of Nigeria". *Biomed central veterinarian research* (2012).
19. Doherr MG., *et al*. "Risk factors associated with subclinical mastitis in dairy cows on Swiss organic and conventional production system farms". *Veterinary medicine* 52.11 (2007): 487-495.
20. Duval j. Treating mastitis without antibiotics, ecological agriculture projects. AGROBIO. Education, South Asia (1995).

Citation: Teshome Setegn and Takele Adugna*.* "Study on Udder Health Status and Antibacterial Susceptibility of Common Bacterial Isolates from Clinical and Subclinical Mastitis in North Gondar Zone, Ethiopia". *Multidisciplinary Advances in Veterinary Science* 1.6 (2018): 236-246.

21. Sefinew A., *et al*. "Study on bovine mastitis and its effect on chemical composition of milk in and around Gondar town Ethiopia". *Journal of veterinary medicine and animal health* 5.8 (2013): 215-221.
22. Adams HR. Veterinary pharmacology and therapeutics, 8th edition, Blackwell publishing, IOWA State University press (2001).
23. Wattiaux M. "Mastitis the diseases and its transmission". Babcock Institute for International Dairy Research and Development (2001).