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Prevalence of mastitis and identification of its bacterial causative agents in small holder dairy farms in and around Wukro of Tigray region, Ethiopia

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Abstract

A cross sectional study was conducted to determine prevalence of bovine mastitis to identify bacteria responsible for mastitis infection and to assess potential risk factors associated with the disease. A total of 360 lactating cows from small scale dairy farms in and around Wukro were included in the study. California Mastitis Test, clinical examination of udder and teats, and bacteriological examination were employed during the study period. The overall prevalence of mastitis was 36.1%, from which 9.4% and 26.7% were clinical and subclinical mastitis, respectively. The quarter level prevalence of the disease was also15.9%; from which, 13.3 % and 2.6% were found to be of subclinical and clinical form, respectively. Among the cause of bovine mastitis in the study area *Staphylococcus aureus* and *Escheria coli*were found to be the leading infectious causes with the relative percentage of 42.31 % and 35.38%, respectively. The potential risk factors considered in this study including age, parity, mastitis history and treatment history showed statistically significant effects on the prevalence of mastitis. However, risk factors such as lactation stage, breed, and management didn't have a statistically significant association with the occurrence of mastitis. Therefore, hygienic milking practice, culling of chronically infected cows, adequate sanitation of dairy environment, proper attention to health of mammary glands, regular screening tests and awareness of the people of the area about the disease should get emphases as control strategies.

Keywords: Bacteria: Dairy farm: Identification: Mastitis: Prevalence: Small scale: Wukro

1. Introduction

Ethiopia is one of the most populous countries in Africa, having an estimated population of more than 90 million. The country is very much dependent on agriculture. Livestock represent a major national resource and form an integral part of the agricultural production system. Ethiopia has the largest cattle population in Africa with an estimated population of 56.71 million. Out of this total cattle population, the female cattle constitute about 55.5% (CSA, 2015).

In contrast to huge livestock resources, the livestock productivity is found to be very low and milk production does not satisfy the country's requirement due to many factors (Addisalem & Mersha, 2012).

The major biological and socio-economic factors that contribute to the low productivity include the low genetic potential and performance of the local breed, traditional husbandry practices and different livestock diseases. Mastitis is among the various factors contributing reduced milk production (Biffa *et al.*, 2005).

Mastitis is a multi-etiological and complex disease (Sharma *et al.*, 2011). There are many factors acting simultaneously and the disease generally involves interplay between management practice and infectious agents but with other factors, such as genetics, udder shape or climate (Sori*e t al.*, 2005; Radostits *et al.*, 2007; Gera and Guha, 2011; Awale *et al.*, 2012).

Mastitis is a global problem as it adversely affects animal health, quality of milk and the economics of milk production, affecting every country, including developed ones and causes huge financial losses (Maiti and Sharma, 2007). There is an agreement among authors that mastitis is the most widespread infectious disease that leads to sever economic losses in dairy cattle (Mostert *et al.*, 2004; Halasa *et al.*, 2007; Elango *et al.*, 2010; Tiwari *et al.*, 2010; Sharma *et al.*, 2012).

Clinical mastitis results in alterations of milk appearance, composition and decreased milk production and the presence of the cardinal signs of inflammation. It is readily apparent and easily detected (Smith, 2002). In contrast, detection of mammary quarters with sub-clinical mastitis is more difficult because signs are not readily apparent (Quinn et al., 2002; Kivaria, 2006; Behiry et al., 2012) and because of the lack of any overt manifestation, its diagnosis is a challenge in dairy animal management and in veterinary practice. The sub-clinical form is 15 to 40 times more prevalent than the clinical form and usually precedes the clinical form and is of long duration (Seegers et al., 2003).

Mastitis is a complex disease, and thus there is no simple solution for its control, so understanding its occurrence, the related risk factors, and the mastitogenic pathogens involved are fundamental elements in developing a control program. A first step in mastitis control program is to quantify udder health by determining the prevalence and incidence of both clinical and sub-clinical mastitis, and assess bacteriological aspects of the disease (Karimuribo *et al.*, 2000).

Livestock kept or produced in small-scale farming systems are an important component of the agricultural economy in the developing world (McDermott *et al.*, 1999) and small-scale dairy development is a powerful tool for actively involving the poor in boosting rural economic growth, initiating a process of change and improving livelihoods (FAO, 2009). The small-scale dairy sector contributes significantly to alleviating poverty and reducing malnutrition, particularly in rural and peri-urban areas, in addition to providing regular income for the household and employment opportunities for women and animal attendants (Karimuribo *et al.*, 2006).

Several studies in the country have documented about the prevalence of both clinical and sub clinical mastitis with significant economic losses associated with the disease in various scales of dairy farming system (23% by Demelash et al. (2005) in Southern Ethiopia;52.8% by Hunderra et al. (2005) in Sebeta; 28.2% by Molalgn et al. (2010) in and around Bahirdar; 34.9% by Biffa et al. (2005) and 46.7 % by Aberaet al.(2010) in Adama;71.0% by Mekbib et al. (2010) in Holetta town; 59.1% by Bedane et al. (2012) in Borena Zone; 65.3% by Addisalem and Mersha (2012) in Addis Ababa; 46.9 % Sefinew et al. (2013) in and around Gonder; 52.9 % by Lidet et al.(2013) in Areka; 66.6% by Birhan et al.(2013) in Asella; 74.7% by Zeryehun et al (2013) in and around Addis Ababa). Studies conducted in Tigray region of Ethiopia (Enquabahir et al., 2008; Rgbe et al., 2012; Mekonnen et al., 2012; Zenebe et al., 2014) also indicated for higher prevalence of the disease and its importance in the production and productivity of the dairy sector. However, there is no study conducted in and around Wukro especially considering the small scale dairy production. Therefore the objective of the present study were to determine the prevalence of bovine mastitis in small scale dairy farms of Wukro and its surroundings, to isolate mastitis causing bacteria from CMT positive and clinically infected milk samples and to assess the association of some risk factors with the occurrence of mastitis in dairy cows.

2. Materials and Methods

2.1. Study Area

The study wasconducted in and around Wukro, Eastern zone of TigrayNational Regional State, Northern Ethiopia. Wukro is located at a distance of 826 km from Addis Ababa and 45 km from Mekelle, capital city of Tigray. Wukro town is found between $13^{0}34'0"$ and $13^{0}56'45"$ North latitude, and $39^{0}20'15"$ and $39^{0}42'45"$ East longitude with an average altitude of 2105 m above sea level (Fig. 1). The ranges of minimum and maximum temperature vary between

11.8°C and 29.9°C respectively, where the meant annual temperature is 20°C. The mean annual rainfall is 350mm. The cattle population of Eastern Tigray is 437,686 (224,852 males and 212,834 females) (CSA, 2015).

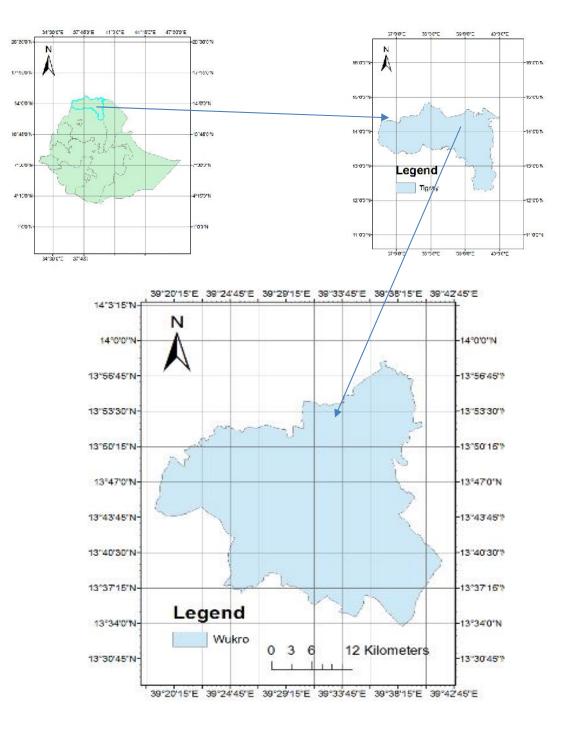


Figure 1: Map of the study area, Wukro and its surrounding

2.2 Study Animals and Study Design

The study animals were lactating local zebu, Jersey and Holstein Frisian cows in the study area. A cross sectional study was conducted from November 2015 to April 2016 in and around Wukro in lactating cows of small holder dairy farms. The sample size was calculated according to the formula given by Thrusfield (2005) by assuming 37.4% expected prevalence (Rgbe et al., 2012) and 95% confidence interval and 5% precision level. Accordingly, the sample size of lactating cows was determined to be 360. Simple random sampling method was considered to select individual small holder dairy farms. Farms were considered small scale if the number of animals are from 2 to 15 (Devendra, 2001). To determine the association of risk factors, data including age, breed, stage of lactation, parity, mastitis history, treatment history and management of the cows were recorded. Age of the study animals was determined by asking the owner and from dentition characteristics, and categorized as young adults (3 to 5 years), adults (6 to 9 years), and old (>9 years). Parity was also categorized as few (with one to three calves), moderate (four to six calves), and many (>6 calves). Lactation stage of the cow was categorized as early lactation (1–120 days), mid-lactation (121–240 days), and late lactation (above 240 days) (Rgbe et al., 2012).

2.3. Study Methodology

2.3.1 Clinical examination of the udder

Clinical mastitis was diagnosed at the quarter level based on visible and palpable signs like hard and swollen quarter, pain (kicking up on touching the udder) and heat (Kivaria *et al.*, 2007). In addition, milk from each quarter was 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.

2.3.2. California Mastitis Test (CMT)

The California Mastitis Test (CMT) was carried out as a screening test for sub clinical mastitis. It was carried out as per the procedure of Quinn *et al.* (2004). The teats were cleaned before conducting the test. A squirt of milk, about 2 ml from each quarter was placed in each of the shallow cups in the CMT paddle. An equal amount of the commercial CMT reagent was added to each cup. A gentle circular motion was applied to the mixtures in a horizontal plane for 15 seconds. Then depending on CMT results, cases were categorized as either positive based on degree of jell formation or negative which did not show jell formation. If at least one quarter was positive by the CMT, the cow was considered positive.

2.3.4. Milk sample collection

Milk samples were collected aseptically from quarter's diagnosed with CMT>+2 and clinical cases and were submitted for bacteriological examination. Briefly, the udder of the cow was thoroughly cleaned with water and dried with a clean towel. After disinfecting the teats with swabs with 70 % ethyl alcohol, milk was collected. The first three to four streams of milk were discarded, and then, 5–10 ml of milk was collected from each teat aseptically in separate universal bottles. Tubes were sealed properly and transported on ice to Veterinary Microbiology laboratory in Mekelle University, where samples were immediately cultured or kept in a refrigerator at 4 °C for a maximum of 24 h until cultured on standard bacteriological media.

2.3.5. Bacteriological examination of milk samples

The CMT positive and clinically infected milk samples were subjected to bacteriological examination according to the standard protocols with minor modifications Quinn et al. (2004) and NMC (2004). A loop full of milk sample (0.01 ml) was streaked on blood agar base (Oxoid,UK) enriched with 5-10% defibrinated sheep blood, and then sub-cultured on selective media for different bacteria using the quadrant streaking method; Mannitol Salt Agar for Staphylococcus, Eosine Methylene Blue Agar for E.coli, Salmonella Shigella Agar (SSA) for Salmonella and Shigella and Edwards medium for Streptococci. In addition, the samples were also cultured on MacConkey Agar for differentiation. All the plates were incubated aerobically at 37°C for 24 -48h. The plates were examined for gross colony morphology, pigmentation and hemolytic characteristics at 24-48h. For further identification of the organisms different biochemical tests (Gram reaction, the Catalase test, Haemolysis, O-F test, Indole test, Methyl red test, Citrate utilization test, TSI agar test and Mannitol fermentation test) were conducted (Quinn et al., 2004).

2.4. Data Entry and Analysis

Data obtained both from records prepared for the study, CMT results and bacteriological tests were stored in Microsoft Excel spreadsheet. Prevalence of mastitis related to specific risk factors was determined as the proportion of affected cows out of the total examined. These data were analyzed using R statistical package version 3.2.4. Descriptive statistics, chi-square test and principal component analysis were performed to see the contribution of the various risk factors for the occurrence of mastitis.

3. Results

3.1.1. Characterization of the Farms Visited

During the present study, a total of 105 small scale dairy farms were visited. Out of this, 94.7% of the farms exercise intensive management system(Table 1). The herd size of the farms ranges from 2-11, where the mean herd size was 5 (Fig. 2A). About 30 of the farms visited had 2 cows and about 18 of them had 10 cows (Fig. 2B). The age of the animals ranges from 3 to 10 years where the mean age is 4.96 years.

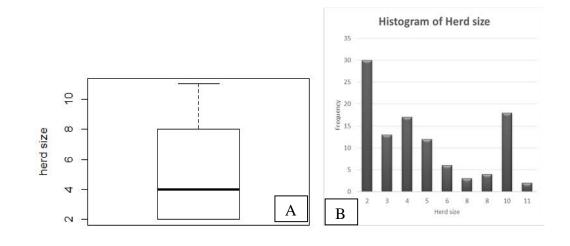


Figure 2: Average herd size (A) and number of farms with the herd size (B)

Table 1: Characteristics of small scale dairy farms visited

Farm characteristics	Frequency (percentage)	
Management system		
Intensive	341 (94.7%)	
Semi-intensive	19(5.3%)	
Herd size		
Minimum	2	
Mean	5	
Maximum	11	
Age of animals in years		
Minimum	2	
Mean	4.96	
Maximum	10	

3.1.2. Prevalence of Mastitis

Of the total 360 cows examined during the study period, 130 (36.1%) of the cows were positive for mastitis. Out of these,9.4% (34/360) and 26.7%(96/360) showed clinical and subclinical mastitis, respectively. The quarter level prevalence was found to be 15.9% (229/1440); from which, 13.3 % (192/1440) and 1.5 % (21/1440) were found to be of

subclinical form and blind teat, respectively. The remaining 1.1% (16/1440) was clinical form revealing active cases of mastitis with visible signs of inflammation on the udder and changes in milk quality. From the total of 130 cows found positive for mastitis,50.8%, 30.0%,11.5%, and7.7% were found positive for single, two, three, and four quarters, respectively (Fig. 3).

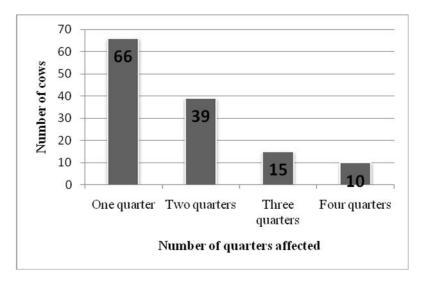


Figure 3: Mastitis positive cows with quarters affected

3.2. Risk factors

Association of the investigated risk factors and the occurrence of mastitis in farms of studied areas are shown in Table 2. Chi square analysis indicated that age, parity, mastitis history and treatment history (p = 0.000) showed a statistically significant association with the occurrence of mastitis. Cows older than 9 years were more affected (70 %) with mastitis compared to adult cows (65.6 %) and young adult

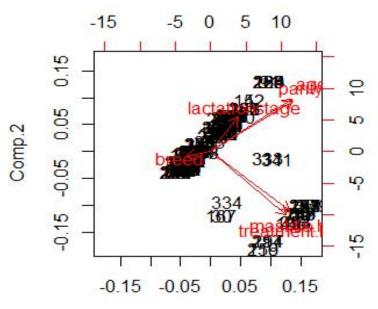
cows (17.6%). Similarly there was a higher occurrence of mastitis in cows with four to six calves (69.9%) than those cows with one to three calves (17.8%). In relation to lactation stages of the cow, a higher prevalence of mastitis was recorded in cows at late stage of lactation (42.4%) than cows in early (32.5%)and mid (30.3%)lactation stages. However, there was no statistically significant association within the stage of lactation.

Risk factors	Category level	Number	Prevalence, No. (%)	X^2	p-value
Age	2-5 years	222	39 (17.6%)		
	6-9 years	128	84 (65.6%)	86.394	0.0000
	>9 years	10	7 (70.0%)		
Breed	Holstein Friesian	302	112 (37.1%)		
	Jersey	18	9 (50.0%)	4.841	0.089
	Local	40	9 (22.5%)		
Lactation stage	Early	80	26(32.5%)		
	Mid	122	37 (30.3%)	4.934	0.085
	Late	158	67 (42.4%)		
Parity no.	1-3 birth	230	41 (17.8%)		
	4-6 birth	113	79(69.9%)	93.090	0.0000
	>6 birth	17	10 (58.8%)		
Mastitis history	Yes	29	27 (93.1%)		
	No	331	103 (31.1%)	44.406	0.0000
Treatment history	Yes	27	25 (92.6%)		
	No	333	105(31.5%)	40.362	0.0000
Management	Intensive	341	126(37.0%)		
	Semi intensive	19	4(21.1%)	1.972	0.160

Table 2: Logistic regression analyses (LR) of risk factors for the occurrence of mastitis in small scale dairy farms of Wukro town and its surrounding

To see the influence of these risk factors on the occurrence of mastitis, a principal component analysis (PCA) was also performed. According to the PCA, result age, parity, mastitis history and treatment history were found to be the most influential factors to

the occurrence of mastitis (indicated with long arrows) (Fig. 4). As it is indicated in figure 4, lactation stage and breed have no significant influence to the occurrence of mastitis.



Comp.1 Figure 4: Principal component analysis of potential risk factors to the occurrence of mastitis

3.3. Bacteriological examination result

Milk samples from 208quarters (96 CMT positive and 13 clinically infected cows) were cultured. A total of 179 bacteria were isolated from the milk samples processed. Out of these isolates, 154 (86.0%) were from subclinical mastitis, and the other 25 (14.0%) isolates were from clinical mastitis. The relative occurrence of S. *aureus* was highest compared to other bacteria isolated and it accounted for 30.7% (55/179) of the total isolates (7 and 48 isolates from clinical and

subclinical mastitis, respectively). The relative occurrence of *E. coli* in the present study was 25.7% (46/179) where 3 and 43 isolates were from clinical and subclinical mastitis, respectively. *S.aureas* and other staphylococcus *spp* were the most common isolate from clinical mastitis cases 7 (28%) followed by Klebsiella and shigella *spp* 4(16%) for each whereas *S.aureus* 48 (31.2%) was the predominant isolate from subclinical cases followed by *E.coli*43 (27.9%)(Table 3).

Table 3: Relative occurrences of bacteria isolated from clinical and subclinical mastitis

Bacterial spp	Clinical no. (%)	Subclinical no. (%)	Total no. (%)
E. coli	3(12.0)	43 (27.9)	46 (25.7)
Klebsiella spp	4 (16)	9 (5.8)	13 (7.3)
Shigella spp	4 (16)	9 (5.8)	13 (7.3)
Salmonella spp	-	16 (10.4)	16 (8.9)
S.aures	7(28)	48 (31.2)	55 (30.7)
Other staphylococcus spp	7 (28)	22 (14.3)	29 (16.2)
Streptococcus spp	-	7 (4.5)	7 (3.9)
Total	25 (100)	154 (100)	179 (100)

4. Discussion

The present study shows an overall mastitis prevalence of 36.1%, as determined by CMT and clinical examination of udder of cows from small scale dairy farms in Wukro and its surroundings. The finding is comparable with previous reports of 35.1% Nibret et al. (2012) in Hawassa, 35.5 by Moges et al. (2012) in Hawassa, 37.2 by Abera et al. (2012) in Shashemeneand 37.4 by Rgbe et al.(2012) in Northern Ethiopia. However, the overall prevalence is higher than the previous findings of other authors in different regions of Ethiopia like23% by Demelash et al. (2005) in Southern Ethiopia, 28.2% by Molalgn et al. (2010) in and around Bahirdar, and 34.9% by Biffa et al. (2005) in Southern Ethiopia. On the other hand, the findings of the present study were lower than the findings of other authors (46.7 %by Abera etal.(2010) in Adama;46.9 % Sefinew et al. (2013) in and around Gondar; 49.7% by Enkuabahir et al.(2008) in Tigray; 52.8% by Hunderra et al. (2005) aroundSebeta;52.9 % by Lidet et al.(2013) in Areka; 59.1% by Bedane et al. (2012) in Borenazone; 62.9% by Mekonnen et al.(2012) in and around Mekelle; 64.3% by Zenebe et al.(2014) inAdigrat;65.3% by Addisalem and Mersha (2012) in Addis Abeba; 66.6% by Birhan et al.(2013) in Asella;71.0% by Mekbib et al. (2010) in Holetta

town; 74.7% by Zeryehun *et al.* (2013) in and around Addis Ababa). The difference in prevalence reports of mastitis in the present study and other reports could probably be due to difference in breeds, management practices, geographic areas, level of production and differences in study methods.

The prevalence of subclinical mastitis was higher 96(26.7%) than clinical mastitis 34(9.4%). A similar observation of the dominance of subclinical mastitis was observed by several researchers: 68.0 and 24.6% by Mekonnen and Tesfave (2010), 28.6 and 8.6% by Abera et al.(2012),37.2 and 9.7% by Sefinew et al.(2013), 33.8 and 10.3% by Delelesse (2010), 30.6 and 4.9% by Moges et al. (2012):33.67 and 18.21% by Hameed et al., (2012), 63.8 and 3% by Jarassaeng et al.(2012), 16.3 and 4.8% by Katsande et al.(2013), 55.1and19.6 %, by Zeryehun et al. (2013), 38.0 and 21.1% by Bedane et al. (2012), 30.6 and 4.9% by Nibret et al. (2012) for sub clinical and clinical mastitis, respectively. The finding strengthened the claim made by Ojo et al. (2009) that subclinical mastitis remains the most economically damaging disease for dairy industry particularly to the small scale dairy producers.

The overall quarter prevalence of mastitis (15.9%) found in this study was in agreement with the report Rgbe *et al.* (2012) in Northern Ethiopia who reported 15.4% but slightly higher than the finding of Molalgne *et al.* (2010) in and around Bahir-Dar and who reported quarter prevalence rate of 14.2%. However, the present finding was lower than the report made by Mekonnen *et al.* (2012) from small holder dairy farmers in Mekelle, Zeryehun *et al.* (2013) in and around Addis Ababa, andZenebe *et al.*(2014) in Adigrat who reported 31.6%, 47.9% and 54% quarter level prevalence, respectively.

In the study, the predominant organisms isolated from mastitis were S. aureus(30.7%) followed by E. coli (25.7%). This is in agreement with the report of several investigators in the country (Hunderra et al., 2005; Abera et al., 2010; Rgbeet al., 2012; Zenebe et al., 2014). The predominance and primary role of S. aureus isolates in bovine mastitis has also been reported in other studies (Mekbib et al., 2010;Girma et al., 2012;Birhan et al., 2013;Zeryehun et al., 2013). High prevalence of S. aureus points to poor milking time hygiene as this pathogen is mainly spread during milking via milkers' hands and towels (Bradley, 2002;Khan and Muhammad, 2005; Sharma et al., 2007;Mdegela et al., 2009; Rahman et al., 2010; Ali et al., 2011). The organism is well adapted to survive in the udder and usually establishes sub clinical infection of long duration from which it shed in milk facilitating trans-mission to healthy animals mainly during milking (Radostits et al., 2007).

Higher prevalence of coliform mastitis is an indication of poor hygienic practices in dairy environment, as these organisms originate from the cow's environment and infect the udder through the teat canal (Mekonnen and Tesafaye, 2010). Contamination of end of the teat is a major predisposing factor in the development of environmental mastitis (Bradley, 2002). The most commonly used antibiotic for the treatment of mastitis in the study area is penicillin in combination with streptomycin, and this may explain for the lower rate of isolation of streptococcal organisms in the present study.

The study reveals the prevalence of mastitis to be affected significantly (p=0.0000) with age. The prevalence of mastitis was highest in old cows (70.0%) followed by young adults (65.6%) and young cows (17.6%). This is in agreement to the findings of previous works by Moges *et al.* (2011),Zeryehun *et al.*(2013) and Zenebe *et al.* (2014). The highest

prevalence in older cows is because of their largest teats and more relaxed sphincter muscles, which increase the accessibility of infectious agent in the cows' udder (Radostits *et al.*, 2007).

Jersey bred cows were affected (50.0%) at higher rate than Holstein Friesian (37.1%) and local breeds (22.5%), this result is in accordance with result of Moges *et al.*,(2012); Sudhan and Sharma, (2010); Joshi and Gokale, (2006); Dego and Tareke, (2003); Sori, Zerinhum and Abdicho, (2005); Lakew, Tolossa and Tigre, (2009)but the difference in this studyis statistically insignificant (p= 0.089). This agrees to the findings of Moges *et al.*, (2011).

Cows having 4-6 calves (69.9%) were at greater risk than those of cows having 1-3 (17%)and cows having more than 6 calves (58.8%). Similar finding was reported by Mekbib *et al.* (2010), Addisalem and Mersha(2012), Rgbe*et al.* (2012), Sefinew *et al.*(2013) and Zeryehun *et al.* (2013).

Late lactation stage had higher relative prevalence (42.8%) than early (32.5%) and mid (30.3%) lactation but the difference is statistically insignificant (p=0.085). This result aligns with reports made by Addisalem and Mersha(2012), Mekonnen *et al.* (2012), Rgbe *et al.* (2012) and Sefinew *et al.* (2013).

There was high prevalence of mastitis in those cows with previous mastitis history and treatment history (93.1 and 92.6%) respectively (p=0.000 for both).Absence of dry cow therapy regime could possibly be the major factor contributing to high prevalence of mastitis in previously treated animals and for the development of bacterial resistant for treatment due to lack of observance of the full course of antibiotic treatment or the habit of changing therapy, in an inappropriate manner.

There was relatively high prevalence of mastitis in intensively managed cows than semi intensively managed 126(37.0%) and 4(21.1%), respectively. These findings agree with the findings of Hameed and co-workers (2012) in Pakistan, who observed higher prevalence of mastitis in backyard housed animals than in animals kept on the street and open areas, possibly due to the highly contaminated environments in backyard areas. Intensively managed cows present a higher risk for the development of mastitis, followed by semi-intensive, with least risk among extensively managed animals (Hundurra *et al.*, 2005).

In all housing systems, high stocking density, dirty bedding orground, infected utensils, poor ventilation and high humidity are important risk factors. Housing increases the risk of mastitis because of the confinement of the animals, and the multiplication of pathogens in the litters elevates teat challenge, and consequently mastitis. Mastitis prevalence increases in herds housed under poor stable and drainage conditions and in herds where mastitic cows were not milked last. This is much more evident for coliform mastitis (Sudhan and Sharma, 2010).

5. Conclusion

The present study indicated considerable prevalence of mastitis in lactating cows of small scale dairy farmers in and around Wukro, Eastern Tigray with the isolation of major pathogenic microorganisms. A relatively high prevalence of mastitis in this study clearly indicates lack of strategic control measures against the disease, as well as poor surveillance measures. The bacteria isolated from cows' milk samples in the present study are types that cause both contagious and environmental mastitis. From stated risk factors, age, parity, mastitis history and treatment history were statistically significant. Therefore it is important to create awareness to the society to ensure strict personal hygiene and that of animals, and general sanitary condition of the farms. Correct and good milking techniques should be also be practiced in the farms to reduce the incidence of mastitis. In addition, regular screening test should have to be performed to detect subclinical mastitis and proper treatment of the clinical cases as well as appropriate treatment of cows during dry and lactation period should be practiced in the study area.

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