

Studies On the Prevalence of Bovine Mastitis and Its Associated Risk Factors in And Around Ambo Town Oromia Region Central Ethiopia

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Abstract

Mastitis is a major production disease of dairy animals of global importance. A cross sectional study was conducted in and around Ambo town from November 2014 to April 2015 on lactating dairy cows to determine the prevalence of mastitis and associated risk factors. The study was done on 384 lactating cows, of which 120 were local and 264 cross breeds from dairy farms and small stockholders by using questionnaire, clinical examination, and California mastitis test (CMT). Out of 5 dairy farms and 68 small stockholders surveyed, there was no use of machine milking rather they used manual hand milking. Of the total 384 lactating dairy cows examined, 161(42%) cows were infected mastitis with 36 (9.4%) clinical mastitis and 125 (32.6%) subclinical mastitis. Of the potential associated intrinsic cows risk factors considered in this study, breed, parity, age, body condition and milk yield showed significant effects on prevalence of mastitis ($p < 0.05$) in the present study. Floor type showed significant difference ($p < 0.05$) on the prevalence of mastitis among the extrinsic associated risk factors. High prevalence of mastitis was observed in cross breed than local; and cows with age of adult (6-8 years) and old (>9 years) were more susceptible to mastitis. The study also revealed that cows with high milk yield, poor body condition, intensively managed and those kept under bad floor type were more affected by mastitis. The present study revealed that mastitis is the major problem of lactating cows that managed in both dairy farms and small stockholders of different husbandry system, especially subclinical mastitis in the study area. Thus hygiene of the milkers, milking equipment' s and cows at milking and husbandry system, creation of awareness of importance and prevention of subclinical mastitis among dairy farms and small stock holders, milking infected animals and respective quarters at last is recommended.

Keywords: California mastitis test, Dairy farms, Prevalence, Risk factors, Subclinical mastitis

Introduction

Ethiopia is a country with a human population of 80 million with an annual growth rate of 2.2 % and a land size of 1.2 million Km² (CSA, 2007). Agriculture is the backbone of the country's economy, accounting for 45 percent of GDP, 90 percent of exports, and 85 percent of total employment. Livestock is an important part of the agricultural production system as well as a significant national resource. Livestock is a vital aspect of the agricultural production system and a substantial national resource. The population of livestock in the country is the highest of any African country (CSA, 2008). These livestock resources play a vital role in the agricultural and rural economies of the country. They do not only provide food, but they also give critical inputs to crop production. Most Ethiopian farms are in high demand to accommodate the adoption or use of the caterpillar, and animal selection may be due to animal capacity or human activity (ILRI, 1995).

Dairy production is an important part of the livestock production systems, especially in the highland areas of Ethiopia. According to FAO (2003), the total production capacity of national milk in Ethiopia ranges from 797,900 to 1,197,500 tons per square meter equivalent to raw milk. Cattle, camels, and goats are the main livestock species that supply both milk and dairy products. Milk from cows constitutes between 85 and 89% of the total annual milk output of the country. This amount, however, falls far short of the country's national demand for milk and milk products. As a result, the country is reliant on milk and milk products imported from other countries.

Despite many years of research, mastitis remains the most economically damaging disease for the dairy industry worldwide; and it reduces milk yield, profit, and quality of milk and milk products in all dairy-producing countries of the world. It is estimated that on average the affected quarter suffers 30% from production and the affected cow loses 15% of its lactation output (Radostits *et al.*, 2007). The huge financial losses are attributed due to mastitis globally (Pal, 2018). Apart from causing colossal economic losses, this disease also possesses the risk for the transmission of zoonotic diseases like tuberculosis, brucellosis, leptospirosis, and streptococcal sore throat to human beings. Another important public health concern is the problem of antibiotic residue in the milk following mastitis treatment, which can initiate severe allergic reactions in people (Radostits *et al.*, 2007).

Mastitis is a multifactorial disease involving various infectious agents as a causative agent with bacterial pathogens covering the greater share (Schalm *et al.*, 1971; Pal and Mehrotra, 1983; Pal, 2018). The most common causative organisms of udder disease include staphylococci (*Staphylococcus aureus* and *Staphylococcus epidermidis*), streptococci (*Streptococcus agalactiae*, *Streptococcus dysgalactiae*, *Streptococcus uberis*, and *Streptococcus bovis*) and coliforms (mainly *Escherichia coli* and *Klebsiella pneumoniae*). In addition, *Pseudomonas*, *Nocardia*, *Mycoplasma*, and fungi (*Candida albicans*, *Cryptococcus neoformans*, *Geotrichum candidum*, and others) are also implicated in the etiology of mastitis in dairy animals (McDonald, 1997; Pal, 2015; Pal, 2018). Among the microbes, *Staphylococcus aureus* is considered the leading cause of bovine mastitis in many countries of the world including India (Pal, 2018). A comprehensive review of the etiology, pathogenesis, risk factors, diagnosis, and management of bovine mastitis is described by Pal and co-workers (2019).

There are several ways of classifying mastitis. A simple classification recognizes mastitis as two major groups. Environmental mastitis is caused by bacteria, such as *Escherichia coli* and *Streptococcus uberis*, which do not normally live on the skin or in the udder but enter the teat canal when the cow comes into contact with a contaminated environment. The pathogens are normally found in feces, bedding materials, water supply, and feed, and transmitted by contact of the udder with those materials (Radostits *et al.*, 2007). The other form of mastitis is contagious mastitis that which the udder and teats serve as the reservoir of infection. Transmission occurs during the milking process or udder preparation by contaminated hands, udder cloths, and cleaners. The infection develops on the teat's surface and in the teat canal. Bacteria can then enter the mammary gland and cause infection. The majority of infections are subclinical, resulting in elevated cell counts. Contagious bacteria include *Staphylococcus aureus*, *Streptococcus agalactiae*, and *Streptococcus dysgalactiae* (Andrew *et al.*, 2004).

On the other hand, contagious mastitis is classified into three groups depending on the degree of inflammation (Philpot and Nickerson, 1991). The first one is clinical mastitis that characterized by the presence of gross inflammation signs (swelling, heat, redness, and pains). There are grossly visible changes in the udder and milk. Milk may be discolored, contain clots, flakes, and the udder becomes hot, painful and disturbance of function of the udder. The second form of mastitis is subclinical mastitis characterized by change in milk composition with no signs of gross inflammation of the mammary gland or its secretion with the presence of pathogenic microorganisms and

unusual number of somatic cells in the milk (Quinn *et al.*, 2004). Robertson (1985) suggested that the higher prevalence rate of sub-clinical mastitis as compared to clinical mastitis is due to the dissent mechanisms of the mammary gland acting to minimize the severity of the disease. Changes in milk composition during sub-clinical mastitis can be detected by special diagnostic tests including CMT. Chronic mastitis is the third type of infectious mastitis, which is an inflammatory disease that lasts for months and can last from one lactation to the next. Chronic mastitis is usually sub-clinical, although it can occasionally flare up into a sub-acute or acute version that lasts for a short time.

In Ethiopia, mastitis is among the various factors contributing to reduced milk production of dairy animals (Fekadu, 1995; Mekonnen *et al.*, 2006). Nevertheless, there are no nationwide studies on the loss of milk production due to mastitis and the overall economic importance of the disease to the dairy farms of Ethiopia. According to Lemma and co-workers (2001), mastitis was the second major disease next to reproductive diseases in dairy animals in Addis Ababa. Moreover, Bishi (1998) reported the economic losses from clinical and sub-clinical mastitis in Ethiopia. This situation has been worsened by the loss of milk production caused by a multitude of problems (Mungube, 2001).

In Ethiopia, the disease has received minimal attention, and data on its scope, distribution and risk factors is scarce. However, this information is necessary for developing effective management and preventative strategies to help reduce milk losses caused by mastitis. There were no more reports of bovine mastitis with its associated risk factors, magnitude, or distribution in lactating cows kept under various management systems in and around Ambo town, unlike Arga and co-investigators (2012), who only studied bacterial pathogens and udder infection dynamics in primiparous cows in the study area. Thus, systematic and well-designed research is important to understand the current situation of the disease and its associated risk factors to come up with appropriate and sound solutions to the problem.

Therefore, the objective of this study was to determine the prevalence of mastitis in lactating cows in and around Ambo town and also to assess major risk factors that are associated with bovine mastitis.

Materials and Methods

Study Area

The study was conducted in and around Ambo town, which is the capital of West Shoa Zone of Oromia National Regional State. It is found at a distance of 114 km from the capital city, Addis Ababa, in western direction. Ambo town is located at latitudes of 8°47'-9°20'N and 37°32'-38°3' longitude and at an elevation of 2101 meter above sea level (m.a.s.l.). The annual temperature ranges from 15°C to 29°C with an average of 22°C. The study area receives a mean annual rainfall from 800-1000 millimeters of which 70% is heavy rainfall which is from June to September and 30% is short rainfall from February to April (AWRDO, 2014).

Study Populations

The study was conducted on 384 lactating dairy animals found in 5 dairy farms, 68 of small stockholders in and around Ambo town that were kept under different management systems as intensive, semi-intensive and extensive. Two hundred sixty-four cross-breed of Holstein Frisian with indigenous zebu and 120 indigenous Zebu breeds of lactating cows were included in the study.

Study Design, Sampling Techniques, and Sample Size

Sample Size Determination

The number of animals required for the study was determined using the formula given by Thrusfield (2005) for simple random sampling.

$$N = \frac{1.96^2 p_{exp}(1-p_{exp})}{d^2}$$

Where N = required sample size; P_{exp} = expected prevalence; d = desired absolute precision (usually 0.05)

The number of the sample was determined using 95% level of confidence, 50% expected prevalence and 0.05 desired absolute precision. Therefore, 384 lactating cows were used for the study.

Study Design

From November 2014 to April 2015, a cross-sectional study was conducted to determine the prevalence and risk factors of bovine mastitis in nursing cows from 5 dairy farms in Ambo town and 68 small livestock owners in the town and rural areas surrounding Ambo town.

Sampling Strategy

Lactating dairy cows 384 in number were sampled during the study period from dairy farms and small stockholders by using simple random sampling. The sample size for each dairy farm and small stakeholders varies with the number of lactating cows during the study period. After having baseline data from each farm and the owners, the number of animals to be sampled was proportionally allocated.

Method of Data Collection

Questionnaire Survey

A structured questionnaire was prepared, information regarding farm attributes, and cow attributes were collected. The cow attributes included age, breed, parity, stage of lactation, udder tick infestation, and other relevant information. In addition, the other farm attributes, such as herd size, husbandry system, floor type, farm hygiene and hygienic practices before, during, and after milking, and other relevant data are included in the questionnaire.

Physical Examination of The Udder

The presence of obvious indications of inflammation and abnormal milk led to the diagnosis of clinical mastitis. A quarter of the udders were found to be warm, swollen, and painful upon probing, indicating acute clinical mastitis. The viscosity and appearance of each quarter's milk discharge were also checked for clots, flakes of blood, and watery secretion. In cases of acute mastitis, the rectal temperature was checked to see if the infection had spread throughout the body. Chronic mastitis, on the other hand, was defined as atrophied, misshaped, hard, and fibrotic quarters. Tick infestation and any wounds in the udder were checked because they are both possible risk factors for future mastitis development, even in otherwise healthy udders (Quinn *et al.*, 2004; Radostits *et al.*, 2007).

California Mastitis Test

California Mastitis Test (CMT) was carried out following the procedure described by Quinn and others (2004) for sub-clinical mastitis. In a horizontal plane, a drop of CMT reagent was put to 4 cups of CMT paddle, to which an equal amount of milk from the proper cow dung was added and carefully stirred around the plane. Then test result was interpreted based on the thickness of the gel formed by the CMT reagent and milk mixture and scored as negative (0), trace (T), + (weakly positive), ++ (distinctive positive), and +++ (strongly positive). Quarters with a CMT score of + or above were taken as positive. Cows were considered positive for CMT when at least one quarter is found to be positive for CMT. A herd was considered positive for CMT when at least one cow in a herd was tested positive for CMT. The prevalence of sub-clinical mastitis was calculated by subtracting the total number of blind teats and those with clinical infection from the total number of teats. The cows' age groups, parity, stage of lactation, milk yield, and body condition were grouped based on the classification used earlier by others (Biffa *et al.*, 2005; Nibret *et al.*, 2011).

Data Analysis

The data collected from the study area including intrinsic risk factors (breed, age, parity, stage of lactation, body condition, milk yield), and extrinsic risk factors (husbandry system, udder tick infestation, and floor type) was recorded in the format developed for this purpose. Later on, it was entered into Microsoft excel 2007, and finally, the data was analyzed using SPSS 16 version.

Results

The result from the questionnaire in the present study showed that manual hand milking was only practiced in all 5 dairy farms and 68 small stockholders owners conducted. Among the five farms, there was no udder teat washing practice except in one farm but pre-milking udder teat washing with separate towels and handwashing between milking of the two cows was performed in the remaining 4 farms. Of the 68 owners of small stockholders, only 21 (30.8%) practiced pre-milking udder teat washing without using towels and handwashing between the milking of the two cows. In all 5 farms, there was no use of pre and post milking calf suckling while 56 (82.4%) small stockholders used pre and post milking calf suckling. The total number of lactating cows in each dairy farm and small stockholders used for the prevalence study is shown in Table.1. Of 384 lactating cows, 161 (42%) were investigated for mastitis of which 36 (9.4%) had clinical and 125 (32.6%) subclinical in the study area.

Table 1: Study animals for the prevalence of mastitis in each farm and small stockholders

Farms	Total No. of examined	No. of mastitis positive			Prevalence (%)		
		SCM	CM	Total	SCM	CM	Total
Farm 1	28	11	2	13	39.3	7.1	46.4
Farm 2	22	8	2	10	36.4	9	45.4
Farm 3	31	11	4	15	35.5	12.9	48.4
Farm 4	43	16	3	19	37.2	7	44.2
Farm 5	35	13	2	15	37.1	5.7	42.8
Small stockholders	225	66	23	89	29.3	10	39.3
Total	384	125	36	161	32.6	9.4	42

SCM=Subclinical mastitis, CM=Clinical mastitis

Among the intrinsic and extrinsic cow's potential risk factors, breed, age, parity, and floor type were significantly associated with the occurrence of bovine mastitis in the study area. The association of mastitis with the risk factors was investigated using chi-square (χ^2) and odds ratio (OR) as shown in Table 2.

Table 2: Intrinsic risk factors for the occurrence of mastitis in cows

Risk factors	No. examined	CM	SCM	Total prevalence	χ^2	P-value	OR(95% CI)
Breed				13.58	0.001		
Local	120	16(13.3%)	24(20%)	40(33.3%)			
Cross	264	20(7.6%)	101(38.2%)	121(45.8%)			0.8(0.1-5.6)
Age Groups	15.022	0.02					
Young adult	143	7(5%)	37(25.9%)	44(30.8%)			
Adult	163	17(10.4%)	62(38%)	79(48.4%)			0.5(0.1-1.5)
Old	78	12(15.4%)	26(33.3%)	38(48.7%)			0.8(0.3-1.9)
Parity				13.364	0.001		
Few	190	14(7.4%)	49(25.8%)	63(33.2%)			
Moderate	131	14(10.7%)	55(42%)	69(52.7%)			1.6(0.5-5.4)
Many	63	8(12.7%)	21(33.3%)	29(46%)			2.5(0.9-6.4)
Stage of lactation				6.158	0.188		
Early	123	15(12.2%)	32(26%)	47(38.2%)			
Mid	151	10(6.6%)	51(33.7%)	61(40.3%)			0.6(0.3-1.1)
Late	110	11(10%)	42(38.2%)	53(48.2%)			0.8(0.4-1.4)
Milk yield	20.671	0.00					
Low	126	17(13.5%)	25(19.8%)	42(33.3%)			
Medium	118	9(7.6%)	37(31.4%)	46(38.9%)			0.3(0.04-1.7)
High	130	10(7.7%)	63(48.5%)	73(56.2%)			0.3(0.19-0.6)
Body condition				33.018	0.00		
Poor	69	13(18.8%)	37(53.6%)	50(72.4%)			
Good	315	23(7.3%)	88(27.9%)	111(27.9%)			7.3(3.7-14.2)

Debele (2010) (33.8%) but lower than the findings of Kerro and Tarekegn (2003) in Southern Ethiopia (63%), Mekibib *et al.* (2010) (48.6%), Machang and Muyungi (1998) in Tanzania (67%) and Kivaria *et al.* (2004). Because of the udder's defense mechanism, which minimizes the severity of the disease, subclinical mastitis has been reported to be higher than clinical mastitis (Erskine, 2001).

In the present study, an attempt was made to determine the prevalence of mastitis related to the association with various risk factors. The present finding showed that there was a highly significant difference in breed, age, parity, milk yield, and body condition susceptibility to mastitis. This could be attributed to the difference in certain physiological and anatomical characteristics of mammary glands, and the occurrence of mastitis may be influenced by some heritable characteristics, such as the capacity of milk production, teat structure, and udder conformation (Radostitis *et al.*, 2007).

The prevalence of mastitis in the present study showed statistically significant ($p < 0.05$) differences among breeds, age, parity, milk yield, and body condition that in agreement with Biffa and co-workers (2005) and Mungube and others (2004) who reported that mastitis prevalence was higher in older cows, those with many parities and high milk yielding cows. Accordingly, the likelihood of mastitis was higher in the crosses of indigenous zebu with Holstein Frisian (45.8%) than local indigenous Zebu breed (33.3%) that was also revealed in the findings of Nibret *et al.* (2011) and Biressaw and Tesfaye (2015). Older lactating cows (48.7%) and adult lactating cows (48.4%) were more affected than young adult lactating cows (30.8%), and the prevalence of mastitis was highest in cows with moderate calves (4-7) (52.7%) that followed by cows with many calves (>7 calves) (46%) and lower in cows with low parity (1-3 calves) (33.2%) that is in agreement with the work of Kerro and Tarekegn (2003), Mungube *et al.* (2004), Biffa *et al.* (2005), and Getahun *et al.* (2008). In this context, Quinn and co-workers (2004) have also stated that older cows, especially after four lactations, are more susceptible to mastitis.

The risk of mastitis was higher in lactating cows with high milk yield (≥ 9 L milk per day) (56.2%) than those cows with medium milk yield (4-8 L milk per day) (38.3%), and low milk yield (1-3 L milk per day) (33.3%) that was similarly justified in the finding of Nibret and co-workers (2012) in Hawassa, southern Ethiopia; and also stated by Radostits and others (2007) that high yielding cows are more susceptible to mastitis than low-yielding ones. This may be due to the ease with which injuries are sustained in large udders so that foci for the entrance of pathogens are created; and stress associated with a high milk yield may upset the defense system of the animal.

As increasing in age and parity and poor body condition, the risk of mastitis was increased as reported by Mungube and co-investigators (2004). Although current study showed that cows with poor body condition (72.4%) were more affected than those cows with good body condition (27.9%) and was statistically highly significant ($P < 0.001$). Even though statistically not significant, the prevalence of mastitis was higher in late lactation stage (48.2%) than the stage of mid (40.3%) and early (38.2%) lactation but the clinical mastitis was higher in lactation stage of early (12.2%) than mid (6.6%) and late (10%) lactation stages.

The current study indicated that udder tick infestation and housing system had statistically not significant ($P > 0.05$) but cows with udder tick infestation (59%) was more affected than cows without udder tick infestation (39%). This is due to the skin lesions of teats and/or udder that had a high prevalence of mastitis, possibly because of colonization of the lesion by pathogens (Nibret *et al.*, 2012). In addition, the observation of Mulei (1999) in Kiambu district of Kenya indicated that mammary gland quarters with teat lesions were 7.2 times more likely to have a positive CMT and 5.6 times more likely to have bacterial organisms isolated from them than those without any teat lesions.

Out of lactating cows of 168 intensively, 127 semi-intensive, and 89 extensively managed, the highest infection rate was seen in cows managed under intensive (45.8%), followed by semi-intensive (41.7%) and extensive (34.7%) husbandry systems. This is due to the confinement of cows in intensive management systems that facilitated the transmission of mastitis-causing agents from infected cow to others. The floor type was statistically significantly associated with mastitis ($P < 0.05$) that the prevalence was highest in bad concrete floor type (52.8%) than in good concrete floor type (40.5%) and in muddy soil floor type (32.3%) that managed under different husbandry system in the study area. Likewise, Getahun and co-investigators (2008) were also reported higher prevalence in cows living in the poor housing system.

Conclusions and Recommendations

The present study revealed that mastitis is the major problem of lactating cows that are managed in both dairy farms and small stockholders of different husbandry systems in the study area. From the 5 dairy farms and 68 small stockholders surveyed there was no use of machine milking rather they use manual hand milking. The subclinical form of mastitis was the most prevalent when compared to the clinical mastitis form. Mastitis prevalence in the present study was significantly associated with several risk factors such as breed, age, parity, milk yield, body condition and floor type that have high significance for the presence of mastitis. Crossbreeds, cows with high milk yielding, poor body condition, many parities (>7calves), adult and old cows, and cows kept under bad floor type were more affected by mastitis.

Based on the above conclusions, the following points are recommended.

- Hygiene of the milkers, milking equipment, and cows in the milking and husbandry system should be considered in attempt to reduce the occurrence of mastitis in the study area.
- Culling of chronically affected and old cows, screening the cows for clinical and subclinical mastitis should be practiced.
- Extension services and training programs focused at the creation of awareness at significance and prevention of subclinical mastitis in dairy farms and small stockholders, milking infected animals and respective quarters, at last, is recommended.
- Further study on the identification of causative agents and their antimicrobial drug susceptibility in the study area was suggested.

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Contribution of Authors

During the writing of the manuscript, all of the authors contributed equally. They read the final manuscript and gave it their approval for publishing.

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There is no conflict of interest.

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