

Prevalence and Associated Risk Factors of Bovine Mastitis in Lactating Cows of Small Holder Dairy Farms in Ethiopia

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Abstract

Mastitis is inflammation of the mammary gland and the most common and expensive disease of dairy cattle throughout most of the world. Infection by invading bacteria or other microorganisms and physical injuries on the gland can cause mastitis. Mastitis is classified as sub clinical and clinical and milk quality is affected by the level of both forms. Risk factors that play significant role in causing mammary incompetence are host, environmental and pathogen risk factors. Mastitis not only affects animal health and wellbeing it can have major implications on the profitability of dairy, financial loss and public health significance. Loss of milk production, replacement of culled cows, extra labor, discarded milk from cows with treatment and cost of control measures are the major economic losses to bovine mastitis. Smallholder farmers are not well informed about the serious invisible loss from sub clinical mastitis. Different tests like California Mastitis Test (CMT), clinical examination, somatic cell count (SCC), measurement of pH and specific laboratory have been developed for diagnosis and detecting the presence of microorganisms in the mammary gland. Antimicrobial susceptibility test is used to identify the most effective drugs for mastitis treatment. Intra mammary antimicrobial therapy, parenteral antimicrobial therapy, supportive and dry cow therapies are the major options of treatment. Mastitis control strategies include creating awareness of people on the management practices like milking and housing hygiene by eliminating existing infection, prevent new infection, and monitor udder health are the basic principles of mastitis control program.

Keywords: Mastitis; Mammary gland; Risk factor; Economic loss

Introduction

Ethiopian cattle population is estimated to be about 60 million. Out of this total cattle population, the female cattle constitute about 55.5 percent (CSA, 2019). However, milk production often does not satisfy the country's requirements

due to different factors. Mastitis is one of the factors contributing to reduced milk production [1].

Mastitis is an inflammation of the udder, a common disease among dairy cows worldwide caused mainly by a bacterial infection [2]. It is divided into Clinical Mastitis (CM) and Subclinical mastitis (SCM). Clinical mastitis is with visible changes in milk and udder while Sub Clinical Mastitis is an increased number of inflammatory cells in the milk without an abnormal appearance of the milk or the udder.

Application of hygienic measures during milk collection by washing dirty teats and udders aseptically and dried thoroughly before proceeding to sample collection, using milking machines, lactation and dry cow therapy, teat sealers, dietary supplements and culling are used to reduce the incidence of mastitis [3].

Bovine mastitis is an economically important disease due to its impact in the quantity and quality of milk production. The cost of CM is direct cost discarded milk, cost of medicines and labor cost and indirect costs loss of future production and increased culling [4-6]. Prevalence of subclinical mastitis in Ethiopia and other African regions may impose substantial costs due to indirect losses. In Ethiopian highland crossbred dairy cows have economic loss due to Subclinical Mastitis (SCM). However, most dairy farmers in the country normally do not recognize subclinical mastitis, which incidentally occurs at a much higher frequency than clinical mastitis, while quite few ignore the disease [6].

Mastitis is a complex disease that interacts with microorganisms, host and the environmental factors. Methods commonly employed for diagnosis of mastitis are screening tests, bacteriological examination and physical examination. Mastitis treatment can be administered by different routes by intramammary antimicrobials infused into the udder through the teat canal and Parenteral treatment given by injection [7]. The disease has been reported by several authors in different parts of Ethiopia. But still there is a gap the disease is insufficiently investigated and information relating to its magnitude and risk factors is scanty. The objective of this review is to generate general information on the status of Bovine mastitis in small holder lactating dairy cows in Ethiopia [8].

Literature Review

Definition of bovine mastitis

Mastitis is defined as an inflammation of the mammary gland and most commonly caused by a bacterial infection, but other origins, such as yeasts, fungi, algae and trauma may also result in mastitis. Mastitis is characterized by physical, chemical and bacteriological changes in the milk and pathological changes in the glandular tissue of the udder and affects the quality and quantity of milk. Consequences of mastitis reduces milk yield, increases culling rates, bring treatment costs and occasional death from severe infections. In addition, some udder pathogens affect food safety because they produce toxins that cause food poisoning, as in the case of *Staphylococcus aureus* [9-11].

Class of bovine mastitis

Mastitis can be classified into two main categories, subclinical and clinical. Subclinical mastitis is defined by an increased number of inflammatory cells in the milk without an abnormal appearance of either the milk or the udder. Clinical mastitis is palpable or visible changes in milk and udder and can be mild only abnormalities in the milk, moderate clinical inflammatory signs of the udder tissue or severe additional systemic symptoms [12].

Etiologic agents and source of infection

To date, more than 140 potentially pathogenic species that cause bovine mastitis. Based on the pathogen involved the disease is broadly classified into four types such as bacterial, mycotic/Fungal/algae, and Mycoplasma and Nocardial mastitis. The viruses have least clinical significance [13-15].

Bacterial mastitis is broadly two type: Gram-positive and Gram-negative, of which the major agents of mastitis are the Gram-positive bacteria including *Streptococcus agalactiae*, *Staphylococcus aureus* and *Mycoplasma bovis*. The chief agents of gram negative mastitis include *E. coli*, *Proteus* and *Klebsiella* species [14].

Fungal infection of bovine mammary tissue caused by contamination of teat dips, intramammary infusions and moldy surroundings play significant role. The important mycotic mastitis causes are *Aspergillus fumigatus* and *Candida albicans* [15].

Algal agent like *Prototheca zopfii* is also incriminated in bovine mastitis, as a result of algal contamination of feed and fodder, drinking water, and cattle premises by house hold sewage, discarded food items including bread, rotten vegetables, and fruits. The disease is more prevalent in the regions where cattle are often grazed in the vicinity of public parks, lakes and tourist places [16].

Among several species *Mycoplasma bovis* and to some extent *Mycoplasma bovirhinis* and possibly to a lesser extent *Mycoplasma canadense* are causal agents of contagious bovine mastitis. *Nocardial mastitis* is saprophytic bacteria in origin; the

causal agents, *Nocardia asteroides*, *Nocardia braziliensis* and *Nocardia farcinicus* are involved in several chronic and granulomatous forms of mastitis. Bovine mastitis due to nocardia occurs as a result of poor environmental hygienic conditions, soil contamination of udders, teat dips and intramammary infusions. Mastitis is epidemiologically categorized in to contagious and environmental mastitis. Both of which severely damage the udder tissue of affected cows [17].

The viruses have least clinical significance. The main viral affections of the bovine udder are ulcerative bovine mammillitis (mammary pustular dermatitis) due to bovine herpes virus, pseudo cowpox (milker's nodule) and cow pox viruses, which are truly the infection of epidermis/dermis of udder.

Contagious mastitis pathogens: Contagious mastitis is caused by pathogens live and multiply on and in the cow's mammary gland and are spread from cow to cow, primarily during milking. Contagious pathogens include *Staphylococcus aureus*, *Streptococcus agalactiae*, *Mycoplasma spp.* and *Corynebacterium bovis* [18].

Environmental mastitis pathogen: Environmental mastitis is caused by pathogens found in the habitat of the cow, such as soil, plant material, manure, bedding, or a contaminated water source. Frequently, isolated causative pathogens that contribute to environmental bovine mastitis include members of *streptococci* and gram-negative bacteria, such as *Escherichia coli* and *Klebsiella*

Spread of infection

Infection enters by way of the teat and can spread from cow to cow by milkers' hands or the cups of the machine, in a heavily infected herd the skin of the cows' bodies, milkers' clothes, floor, partitions and less easily by towels. In an infected herd, a large proportion of organisms hiding and these may be a source of infection of the udder itself in the same cow or in another. The skin of the teats and the milkers' hands may remain infected from one milking to another [19,20].

Factors that affect occurrence of bovine mastitis

Bovine mastitis is predisposed by several epidemiological risk factors that play significant role in causing mammary incompetence to protect it from the invasion of infectious agents. The risk factors include the host factors, environmental factors and the pathogen factors [20].

Host risk factor: A great number of cow-specific risk factors for CM have been identified, including breed, parity, period of lactation, udder and teat morphology, age, milk production and number of milk somatic cells increase. The levels of SCC are elevated in early lactation and gradually increase towards the end of lactation. Early stage and late stage of the mammary glands were the most susceptible stages. This is possibly due to absence of dry cow therapy that is considered major factor contributing to high prevalence at early lactation [21].

Prevalence of mastitis is highest in pure breeds followed by crosses; and indigenous zebu being less frequently affected than others. The increase in prevalence in exotic breeds as opposed

to local indigenous zebu could be the indigenous zebu is low in milk production and higher yielding cows are more susceptible to mastitis [22].

Age of cows has effects in occurring of mastitis. It has been shown that manifestation of mastitis in infected quarter's increases with advancement of age in cows. This may be due to more dilated teat canals in older age, permanent udder tissue damage resulting from the primary infection or due to an increased cellular response to intra mammary infection after parturition, early lactation and during the dry period and the incidence of mastitis is reported to be higher during these times [23].

The prevalence of SCM increases with increasing lactation number and parities Cows with the most pendulous quarters appear to be the most susceptible to mammary infections, the pendulous udder exposes the teat and udder to injury and pathogens easily adhere to the teat and gain access to the gland tissue [18-22].

Environmental and pathogen risk factors: The cows environment influences the number and types of bacteria exposed to their ability to resist those organisms. The design of housing system, hygiene, and size of milking cow herd, milking

practice and the climate interact to influence the degree of exposure of a cow to mastitis pathogens. Moisture, mud and manure present in the environment of the animals are primary sources of exposure for environmental mastitis pathogens. In fact in many studies in Ethiopia such as those conducted by a higher prevalence is recorded in cows with poor hygiene in the milking process. Intensively managed cows present a higher risk for the development of mastitis, followed by semi-intensive, with least risk among extensively managed animals [24].

The occurrence of mastitis varies from season to season, because growth and multiplication of organisms depends on specific temperature and humidity. Incorrect ventilation, with high temperature and relative humidity, encourages the multiplication of various bacteria. Exposure of animals to high temperature can increase the stress of the animal and alter immune functions. In Ethiopia, it was noticed by 2003 that the prevalence was higher in the rainy season than in the dry season. Different types of milking methods (stripping, knuckling, full hand method, machine milking) are practiced by dairy farmers. Faulty milking practices, especially knuckling, cause great harm to tissue and they become prone to infection. Summarize different risk factors in the following **Table 1**.

Table 1: Main factors identified as a risk for the occurrence of the bovine Mastitis.

Risk factors	Occurrence of Mastitis	Source
Production level	Higher in high yielding bovines Holstein Friesian (HF), Jersey or HF and Jersey crossbred dairy cows	Moges et al., 2012; Sudhan and Sharma, 2010; Sori et al., 2005; Lakew et al., 2009
Quarters appearance	Cows with pendulous quarters appear to be the most susceptible to mammary infections	Almaw, 2004; Sori et al., 2005
Teat size	Long teats increase the risk of accidental trauma	Almaw, 2004
Breed	Prevalence is highest in pure breeds followed by crosses and indigenous zebu.	Radostits et al., 2006
Age, lactation number and parities	Prevalence of SCM increases with age, increasing lactation number and parities	Deogo and Tareke, 2003; Awale et al., 2012; Moges et al., 2012; Lakew et al., 2009
	Higher prevalence of mastitis in older animals	Girma et al., 2012
Seasonality	Prevalence was higher in the rainy season than in the dry season	Deogo and Tareke, 2003; Tilahunand Aylate, 2015
Milking methods	Faulty milking practices, especially knuckling, cause great harm to tissue and they become prone to infection	Sudhan and Sharma, 2010
	Higher prevalence of the disease in animals milked by folded thumb	Awal et al., 2012
Calf suckling	Highest prevalence of mastitis in animals with calf suckling	Hameed et al., 2012
Moisture, mud and manure in the environment	Moisture, mud and manure present in the environment of the animals are sources of exposure for mastitis pathogens	Sudhan and Sharma, 2010
Poor hygiene in the milking process.	Higher prevalence is recorded in cows with poor hygiene in the milking process.	Lakew et al., 2009; Deogo and Tareke, 2003
Management system	Intensively managed cows present a higher risk followed by semi-intensive with least risk	Sori et al., 2005
Housing systems	Mastitis prevalence increases in herds housed under poor stable and drainage conditions	Sudhan and Sharma, 2010
Weather and climate	A higher incidence of mastitis during summer rainy months	Akyuz et al., 2010 and Godden et al., 2003

Economic impact of mastitis

Mastitis, both clinical and subclinical is known for resulting in a substantial economic loss. Milk yield loss, loss from discarded milk, veterinary service, medicine, increased sanitation, additional labor and equipment major economic loss. Subclinical mastitis in cattle is estimated to result in a loss of 1592.87 Indian Rupees (INR). The largest loss was due to milk yield loss and medicine direct losses due to clinical mastitis in cows to be 2086.96 INR per clinical case also mention a poorer product quality and culling of diseased animals as factors that affect the economy. Subclinical mastitis (SCM) is of great economic importance to dairy farmers because it results in reductions in milk yield and undesirable changes in the milk's composition [24].

Very limited published data are available to quantify production losses and expenditures related to mastitis in developing countries, and thus to assess the economic impact of the disease. Because production systems, environment, management and breeds are different, it is not possible to compare data from developed and developing countries. So there is the need to assess the extent of financial losses due to mastitis on the basis of studies conducted in the developing countries. It is important to bear in mind that mastitis cows are a constant source of contagion due to shedding of bacteria. What farmers may not notice and may not be aware of is the indirect cost stemming from reduced reproductive performance. Studies confirm that mastitis has detrimental effects on reproductive efficiency of dairy cows and thus negatively affects the profitability of dairy herds.

Ethiopia produces approximately 3.2 billion liters from 10 million milking cows and average of 1.54 liters per cow per day over a lactation period of 180 days. In Ethiopia total milk production losses accounted for 78% caused by mastitis. The economic loss from mastitis in the urban and peri urban area of Addis Ababa is US\$ 58 and 78.65 per cow and per lactation, respectively. Losses were highest in large-scale (13%) farms and lowest (3.7%) in small-scale and overall financial loss for each cow per lactation was 984.64 Eth Birr (US\$78.65) and losses in large farms 1,882.40 Eth Birr or US\$150.35.

Public health significance

Milk is a well-known medium that favors the growth of several microorganisms. Milk from a sub clinically mastitis cow commonly contains the etiological agents, while milk from non-mastitis cows is known to be often contaminated from extraneous dirt or unclean processing water. The health hazards posed by milk-borne zoonotic diseases such as brucellosis, tuberculosis and mastitis-related enterotoxaemia are well-documented [25,26].

Besides mastitis render milk unsuitable for human consumption, it provides a mechanism for the spread of many diseases to humans. Most important human disease causing organisms that can be found in milk are *Mycobacterium bovis*

and *tuberculosis*, *Brucella species*, *Salmonella species*, *E. coli*, *Staphylococcus aureus*, *Streptococcus progeny* and *Corynbacterium haemolyticum* (Table 2). Milk and milk products have, therefore, pose a risk to consumers if it is contaminated by any pathogens and subjected to temperature abuse, where these organisms can multiply to high counts and may produce toxins.

Table 2: Important human disease causing organisms that can be found in milk.

No.	Pathogenic micro organisms	Their effect and disease condition in humans
1	<i>Mycobacterium bovis</i> and <i>tuberculosis</i>	Tuberculosis
2	<i>Brucella species</i>	Undulant or Malta or Mediterranean fever
3	<i>Salmonella species</i>	Salmonellosis
4	<i>E. coli</i>	Toxigenic micro organisms
5	<i>Staphylococcus aureus</i>	Intoxication
6	<i>Streptococcus progeny</i>	Otitis media, septicemia
7	<i>Corynbacteriumhaemolyticum</i>	Pharyngitis, cervical adenitis

Milk contains an unacceptable high level of antibiotic residues so causes problems to consumers of such milk and its products. Drug residues in milk apart from other hazardous effects it also affects negatively the health of the consumer of milk with high level of antibiotic residues. These effects include allergic reactions and bacterial resistance in the body of humans.

Status and significance of mastitis in Ethiopia

Ethiopia is believed to have the largest livestock population in Africa. An estimate indicates that the country is a home for 59.5 million cattle with the largest member of cows. Ethiopia is best for dairy development due to its cattle population and favorable climate conditions. The contributions of dairy sector for smallholder poverty alleviation are considerable to be high. However, many factors are constrained by disease like mastitis especially subclinical one. Smallholders' farmers in Ethiopia are not well informed about the invisible loss from sub clinical mastitis. This is for the reason that dairying is handled as a sideline business among farmers [27].

Prevalence of subclinical mastitis in Ethiopia and other African regions may impose substantial costs due to indirect losses. Over the last several years, a number of studies are available that describe the prevalence of bovine Mastitis in different parts of the country. But a number of epidemiological studies carried out in Ethiopia showed that mastitis is a serious problem. To this effect, cumulative data from Ethiopia can serve to develop a large-scale disease control program for other African countries will be explained in Table 3.

Table 3: Prevalence of Mastitis in different area of Ethiopia.

Study area	Sample size	Diagnostic Method	Positive animal%			Source
			clinical	Subclinical	Prevalence	
In and Around Wolaita Soddo SNNPR, Ethiopia	349	CMT and milk culture	2.60%	26.90%	29.50%	Yohannis and Molla 2013
Holleta Agricultural Research Center, Ethiopia	90	CMT	7.80%	73.30%	81.10%	Duguma et al., 2014
Hawassa and Wendo Genet Small Holder Dairy Farms SNNPR, Ethiopia	122	CMTmilk and k culture			63.10%	Kassa et al., 2014
Sidama Zone SNNPR, Ethiopia	96	CMT and milk culture	2.08%	42.70%	42.71%	Tekle and Berihe, 2016.
Dire dawa City, Eastern Ethiopia	334	CMT and milk culture	15.27%	84.73%	39.20%	Kedir et al., 2016
Addis Ababa, Ethiopia	444	CMT	21.20%	46.80%	68.00%	Tilahun and Aylate, 2015
Horoguduru Wollega zone, western Ethiopia	154	CMT	10.39%	36.36%	46.75%	Beyene and Tolosa, 2017
Addis Abeba	499	CMT	19.60%	55.10%	74.70%	Zerhun et al., 2013
Around Sebeta	180	CMT	16.11%	36.67%	52.78%	Sori et al., 2005
Adama town, Eastern shoa	300	CMT	10.00%	36.70%	46.70%	Abera et al., 2010
Eastern Shoa Zone	250	CMT, Milk culture	4%	38.80%	43.20%	Hajje and Teka, 2017

The economic loss from mastitis in the urban and peri urban area of Addis Ababa is U\$ 58 and 78.65 per cow and per lactation, respectively. Losses due to mastitis are commonly derived from sub-clinical and clinical mastitis and their effects are reflected on milk production, composition and quality. The magnitude of these changes in individual cows varies with severity and duration of infection and the causative microorganism that cause mastitis.

Reported a substantial economic loss in Ethiopian highland crossbred dairy cows is due to subclinical mastitis. However, still there is a gap in Ethiopia, the disease is insufficiently investigated and information relating to its magnitude, distribution and risk factors is scant. Such information is important to envisage when designing appropriate strategies that would help to reduce its prevalence and effects [27].

Diagnosis of bovine mastitis

Diagnosis of clinical mastitis is by physical examination including swollen quarters/udder and poor milk quality, can be detected by farmers. The most frequently used diagnostic methods for sub clinical mastitis detection are California mastitis test, somatic cell counting (SCC) and bacteriological culturing of milk.

Physical examination: This involves close clinical examination of the mammary gland for any signs of inflammation, milk for its color, viscosity. This can be done through visual examination of the milk and mammary gland and or palpation of the mammary gland.

California Mastitis Test (CMT): The California Mastitis Test (CMT) is useful technique for detecting subclinical mastitis on-farm, providing an immediate result and for selection of the samples for the bacterial culturing from the cows under. It

conducts in each quarter milk sample immediately after collection. A drop of milk, nearly 2 ml from each quarter placed in each of the four wells of the CMT paddle and an equal amount of the CMT reagent applied to each cup. A gentle circular movement applies to the mixture, in a horizontal plane for seconds. The obtained reaction result classify as Negative, Trace, 1, 2 and 3.

Somatic Cell Count (SCC): Somatic Cell is normal Constituent of milk consists of different cell types, including neutrophils, macrophages, lymphocytes and some epithelial cells. The somatic cell count (SCC) is the number of cells present in milk. Determination of SCC is widely used to monitor udder health. The increase in SCC during mastitis is part of the immune defense system of the cow. SCC of milk in healthy mammary gland is lower than 1×10^5 cells/ml, while bacterial infection can cause it to increase to above 1×10^6 cells/ml [28].

Measurement of pH: Normal milk has pH between 6.5 and 6.7. When infection is present that it tends toward alkalinity with the use of reagent sodium hydroxide.

Bacteriological diagnosis: The laboratory procedure of inoculating standard volume of hygienically collected milk on agar culture medium has been the standard diagnostic method for bovine mastitis. The resulting bacterial growth is observed, quantified & tested.

Bacterial isolates was identified based on colony morphology, pigmentation, Gram stain and conventional biochemical tests. For Gram positive cocci, catalase tests with hydrogen peroxide (3%) used to differentiate between catalase-positive *staphylococci* and catalase-negative cocci. Morphology, haemolysis patterns, coagulase and polymyxin susceptibility test was used to distinguish *Staphylococcus aureus* from *non-aureus Staphylococci*. Gram-negative bacteria identified by using colony

morphology, oxidase test and lactose fermentation on MacConkey agar [29].

Treatment of bovine mastitis

The specific Anti-microbial therapy during dry period is the best method to eliminate existing infection. Mastitis treatments can be administered at two different lactation cycle stages in lactating cow therapy administered to cows while they are in milk and dry cow therapy administered when the cow is dried off. Mastitis treatment can be administered by different routes by intramammary treatment infused into the udder through the teat canal and parenteral treatment given by injection [30].

Conclusion

Control of mastitis requires understanding of its causes and management techniques which limit the spread of infection. The principle of mastitis control is that the disease is controlled by either decreasing the exposure of the teat to potential pathogens or by increasing resistance of dairy animals to infection. The key elements in the control of mastitis include: sound husbandry practices fly control, long-acting intramammary antibiotics and sanitation, post-milking teat dipping, treatment of mastitis during non-lactating period, and culling of chronically infected animals.

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