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Bovine Mastitis: A Review of Causes and Epidemiological Point of View

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Abstract
Bovine mastitis is, inflammation of mammary gland parenchyma due to infection with a pathogen (intra-mammary infection, IMI), injury, allergy and neoplasm, considered to be the most costly disease of dairy animals worldwide. It is said to be a multi-etiological complex disease as it is the outcome of interaction of various associated factors: the host, pathogens and the environment. The disease is characterized by physical, chemical and bacteriological changes in the milk and pathological changes in the glandular tissue of the udder. Infectious agents, in particular, the various species of bacteria are accounted as the most important etiologic agents of mastitis. Disease dissemination within the herd is enabled with large number of possible contacts between dairy animals which especially enhanced by milkers' hand and milking equipment. Detection of the mastitis is often complicated due to the subclinical nature of the mammary infection. Mastitis is a disease warranting serious attention for its control and preventions as it is the most economically overwhelming disease, causing reduction in milk production & milk quality, veterinary expenses and/ or treatment costs with antibiotic withdrawal period besides to the possibility of transmission of zoonotic disease and antibiotics residue secreted by milk. Sound husbandry practices and sanitation, post-milking teat dipping, treatment of mastitis during non-lactating period of the dairy cow, and culling of chronically infected cow are the key elements to be recommended in the control and prevention of mastitis.

1. INTRODUCTION

Dairy cow has been called the foster mother of the human race with milk being a universal and nearly a very nutritional perfect food (Etgen, 1978), containing all the nutrients required by the neonate (I.e., it is rich in carbohydrate, proteins, fats, vitamins minerals and also a rich source of protective agents, enzymes and growth factors) and has thus long been recognized as perhaps nature's ultimate food (Chaplin and Lyster, 1988).

However, milk production often does not satisfy the country's milk requirements due to a multitude of associated factors and constraints such as suboptimal management and/ or inadequate management, low genetic potential, nutritional insufficiency, poor reproductive performance, and various diseases in particular mastitis, which is among the most important impediments confronting the economic milk production in dairy cow (Mukasa, 1998).

Mastitis, because of its high incidence, is one of the most common and a very significantly devastating potential dairy disease occurring in all dairy herds worldwide (Ojo *et al*., 2009), and has a consequence of transmissibility of such diseases as tuberculosis, brucellosis, leptospirosis etc., through milk to consumers, from zoonotic standpoint of view (Nibret *et al*., 2011).

The term is derived from Greek word *Mammae* or *Mastos* and *itis* meaning *breast* and *inflammation*, respectively. Mastitis is defined as an infection of the udder, caused by bacteria entering the quarter through the teat end ((Radostits, *et al.,* 2007)), and according to the National Mastitis Council's current concepts of bovine mastitis (1996), mastitis is an inflammation of the mammary gland characterized by physical, chemical and usually bacteriological changes in milk and pathological changes in glandular tissues, due in response to injury for the purpose of destroying and neutralizing the infectious agents and to prepare the way for healing and return to normal function. Inflammation can be caused by many types of injury including infectious agents and their toxins, physical trauma or chemical irritants (Jones and Bailey, 1998).

Mastitis is a multi-factorial disease and generally results from the interaction of various factors associated with the host, pathogen(s), environment and management. The etiological agents causing mastitis are very vast and complex; large numbers of micro-organisms are known to cause inflammation of udder. Among the infectious agents, bacterial pathogens are major threat to mammary gland. They are often contagious, widely distributed in the environment of dairy animals and thus increase prevalence rate of intra-mammary infections.

The infection originates either from the infected udder or the contaminated environments. The major sources of pathogens and means of transmission include infected quarters and soiled udder, contaminated milking machines, teat cups, milker's hands, washing clothes, flies and surgical instruments. Moreover, the stage of lactation, lactation number, trauma to udder, teat and teat canal, loose teat sphincters, lesions on teat skin, immunological status of each mammary gland, bulk of infection in the environment and manage-mental conditions are amongst the determinants which dictate the level of mastitis in dairy animals (Alemu *et al.*, 2013).

The incidence of mastitis is more common in crossbred cattle compared to indigenous cattle. Since considerable differences in the management of animals between organized and unorganized dairy farms, it is

possible that the milk quality parameter also varies between these systems. In Ethiopia, it is common and habituated practice to the use a calf for stimulating the let down of milk and in contrast, the National Mastitis Council's (NMC) report that calves are also a possible agency of mastitis organism's transmission.

This paper is, therefore, designed with the fulfillment of the following subsequent objectives, in view to:

- ❒ Review bovine mastitis
- ❒ Assess mastitis causing major microorganism and associated risk factors
- ❒ Finally recommend some possible solution on prevention and control of the disease

2. BOVINE MASTITIS

2.1. Types of Mastitis Depending upon the Etiological Agents

Depending on the causative agent, mastitis in cow can be categorized into three main types: Contagious, Environmental and Summer Mastitis (Heeschen, 2012) (Figure 1).

2.1.1. Contagious Mastitis

It is caused by bacteria living on the skin of the teat and inside the udder. Contagious mastitis can be transmitted from one cow to another during milking and can further be classified into as:

Clinical mastitis: It is characterized by the presence of gross inflammation signs (swelling, heat, redness, pain). That is by visual clots or discolorations of the milk, often in combination with tender and swollen udder, sometimes in combination with fever, loss of appetite *etc*. Clinical mastitis can again be divided into *Per-acute mastitis which* is characterized by gross inflammation, reduction in milk yield and changes in milk composition, Systemic signs like fever, depression, shivering and loss of appetite and loss of weight; *Acute mastitis* that is similar to per-acute mastitis but with lesser systemic signs like fever and mild depression and *Sub-acute mastitis,* the mammary gland inflammation signs are minimal and no visible systemic signs (Sears *et al*., 1993).

Subclinical mastitis: It is characterized by change in milk composition (SCC, leukocytes & epithelial cells, changes in milk pH and ion concentration) with no clinical signs of gross inflammation or milk abnormalities. In healthy lactating mammary gland, the milk SCC is often <100,000 cells/ml of milk while can increase to >1,000,000 cells/ml of milk during subclinical mastitis. The major factor affecting the SCC at the herd and individual level is the presence of intra mammary infections (IMM) (Guidry, 2007).

Chronic mastitis: It is an inflammatory process that exists for months, and may continue from lactation of one to the other lactation. It exists as subclinical but may exhibit periodical flare-ups sub-acute or acute form that last for a short period of time.

2.1.2. Environmental Mastitis

It is caused by organisms such as *Escherichia coli* which do not normally live on the skin or in the udder but which enter the teat canal when the cow comes in contact with a contaminated environment. The pathogens normally found in feces bedding materials, and feed (Heeschen, 2012).

2.1.3. Summer Mastitis

A third type of mastitis, referred to as summer mastitis, is an acute illness of dry cows and heifers which causes extensive and painful damage to the udder. The infected quarter is permanently damaged resulting in early culling of the cow. Infection is more likely to occur when cows are in environment where the teats can easily be exposed to damage and high fly populations. The clinical signs of summer mastitis are hot, hard and swollen quarter in association with a thick secretion characterized by a foul smell (http://www.Towe.Org uk).

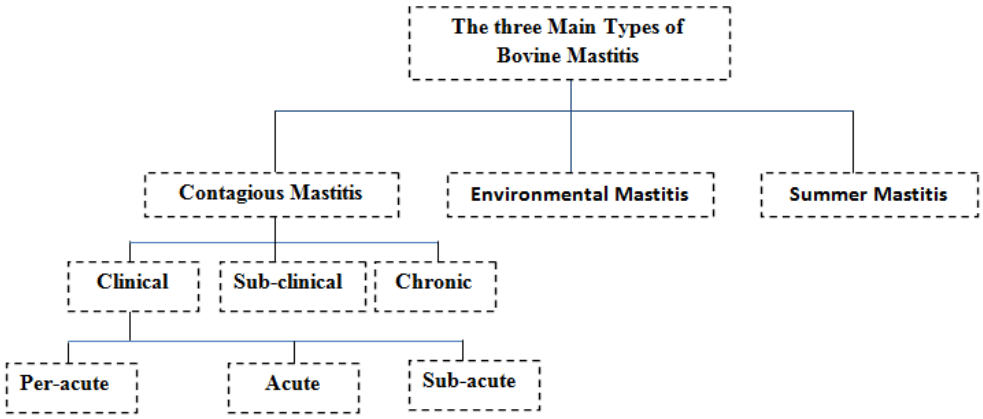

Figure 1: the three Main Types of Bovine Mastitis

2.2. Mastitis Causing Major Pathogen

According to International Dairy Federation (IDF) (1987), though mastitis may have a traumatic or toxic

etiology, it is generally a result of microbiological infection. And more than 150 different species of pathogenic microorganisms have been identified as causative agents of mastitis dairy cow. Bacteria, fungi and yeasts may all play a role; but of these, bacteria have by far the largest part (Quinn *et al.,* 2002).

Although about 20 to 35% of clinical mastitis cases are of unknown etiology (Wellenberg *et al*., 2002), it is widely accepted that bovine mastitis is mainly bacterial in origin. It can be classified as contagious or environmental. In the former case, it is caused by organisms such as *S. aureus*, *Strep. dysgalactiae* and *Strep. agalactiae*, which are all adapted to survive in the udder, causing subclinical infections. Environmental pathogens like *Strep. uberis* or Enterobacteriacae like *E. coli* are not well adapted to survive within the udder and, instead, they multiply rapidly following invasion, evoke a swift immune response and are eliminated (Bradley, 2002).

The main etiological agents responsible for mastitis infections in dairy cow can be divided into different groups of organisms depending on the source of the organism involved. These include *contagious pathogens, environmental bacteria, opportunistic bacteria* and/ or other organisms that less frequently cause mastitis (Philpot and Nickerson, 1999).

2.2.1. Contagious pathogen

They are usually found on the udder or teat surface of infected cows and are the primary sources of infection between uninfected and infected udder quarters, usually during milkings. The organisms that fit into this category include *Staphylococcus aureus* (Coagulase Positive Staphylococci, CPS), *Streptococcus agalactiae* and the less common sources of infection caused by *Corynebacterium bovis* and *Mycoplasma bovis* (Eriksson, 2005)**.** *Staphylococcus aureus is* accounted at the top and colonizes the nipple skin advancing through the mammary gland canal into the gland. The IMI with *Staphylococcus aureus* predominantly cause subclinical mastitis resulting in a chronic infection lingering lifelong (Shearer and Harris, 2008). During the infection's early stages, the mild damage may be reverted but *Staphylococcus aureus* infections; in its per-acute mastitis presentation generates gangrene and severe tissue damage. In comparison with *Streptococcus agalactiae*, *Staphylococcus aureus* is more difficult to be eradicated. And, a cow with infected mammary gland is difficult or even impossible to cure successfully due to the ability of the bacteria to produce different enzymes and toxins which cause damage in the udder tissue and enable the bacteria to penetrate the tissue; survive in the keratin layer of the teat canal which in normal circumstances acts inhibitory; avoid phagocytosis due to the presence of the protein A in some strains (Brown, 2006).

Streptococcus agalactiae is an obligated pathogen of the mammary gland transmitting directly among cows during milking (NMC, 1999). It infects the gland cistern and ducts of the mammary gland causing irritation, swelling and subclinical mastitis. The infected cow shows mere clinical signs without abnormalities drawn in milk. However, low production rates and high SCC are usually registered. *Streptococcus agalactiae* infections are related to Bulk-tank milk (BTM) figures around a 1,000, 000 cells/ml on SCC or higher (Hillerton and Berry, 2003).

Mycoplasma species are less common than *Streptococcus agalactiae* and *Staphylococcus aureus.* Nevertheless, *Mycoplasma* species damage the secretory tissue inducing the gland fibrosis, abscesses and the lymphatic nodules fibrosis. Animals from all ages are susceptible, as well as at any time during lactation. Those in early lactation are more susceptible to *Mycoplasma* infection and it can be isolated from high production animals without signology (Harmon, 1994). Mycoplasmosis is frequently related to the mastitis outbreak onsets, to the introduction of new animals to a herd, to previous respiratory or articular disease, and to herds with unresponsive mastitis to antibiotic treatment. When at least the recurrent mastitis, a non-signs illness and an unresponsive treatment are observed, a mycoplasma infection is suspected (Barbuddhe, 2002).

2.2.2. Environmental Pathogens

According to Quinn *et al.* (2002) a large number of Gram-positive and Gram-negative species are in a cow's environment and most often responsible for most of the clinical cases and fall into a descriptive category known as environmental pathogens such as *Streptococcus uberis, Streptococcus equinus, Enterococcus faecalis* and *Enterococcus faecium* are Gram-positive species. Gram-negative species include *Escherichia coli, Klebsiella* spp*., Enterobacter* spp., *Serratia* spp. and *Pseudomonas* spp.

Environmental pathogens require moisture, favorable pH and organic material for survival and they enter the gland through the teat canal. Environmental pathogens reside in soil, bedding materials, manure and other organic matter. Therefore, efforts at prevention or control of environmental mastitis should focus on cleanliness of a cow's workplace and cleanliness of a cow. Mastitis caused by environmental organisms is essentially opportunistic in nature and becomes established if the immune system of the host is compromised or if sanitation and hygiene is not adequately practiced (Jones *et al*., 1984).

Mekonnen and Tesfaye (2010) revealed that contagious bacteria like *Coagulase negative staphylococci* (CNS), *S. aureus, S. agalactiae, S. dysgalactiae* and environmental microorganisms like coliforms (*Escherichia coli, Enterococcus faecalis, and Streptococcus uberis* were found to be the major etiology of mastitis in market oriented smallholder dairy farms in Adama, Ethiopia. According to Watts (2002) bacterial isolates in the milk of

dairy herds in and around Gondar, Ethiopia were *Staphylococcus aureus*, S*treptcoccus agalactiae*, *Streptcoccus dysgalactiae*, *Streptcoccus uberis*, *Escherichia coli*, *CNS*, *Micrococcus* species, *Bacillus cereus*, *Corynebacterium bovis* and *Actinomyces pyogens.*

In general, among all the pathogens of bovine mastitis *Staphylococcus aureus* is the predominant organism. The etiological agents of mastitis in dairy cow have been reported to be *Staphylococcus aureus*, *Staphylococcus hyicus*, *Staphylococcus epidermidis*, *Staphylococcus capotus, Streptococcus dysaglactiae, Streptococcus agalactiae, Streptococcus pyogenes and Corynebacterium bovis* (Allore, 1993; Kapur *et al*., 1992).

2.3. Epidemiology

Mastitis is considered to be a typical example of complex diseases, known to be established as a result of the interactions of three bio-systems namely the causative agent (pathogen), the animal (host) and the environment in which the animal lives. Host factors include breed, physiological state of mammary gland, and anatomy of teat canal, sphincter tone and presence of teat lesion. Agent factor includes the ability to survive in the immediate environment of the animal, the ability to colonize the teat duct, the ability to adhere to the mammary epithelium and not to be flushed out with milk flow Environmental factor includes milking practice, housing system and bedding (Makovec and Ruegg, 2003) (Figure 1).

Sandholm and Korhonen (1995) reported that the primary and secondary body defense mechanisms prevent the pathogenic microbes from entering the mammary gland through the teat canal orifice. They also indicated that the concentrations of the antibacterial factors in the udder secretion are under genetic control and depend on the lactation stage and udder health. The environmental factors such as management, feeding, hygienic status, bedding, milking and the virulence of the organism contribute to the disease. Lesile (1996) reported that stress factors such as isolation of an individual and mixing groups of cows have been shown to increase somatic cells count in the absence of mastitis, moreover it has been reported that there was no increase in SCC.

2.3.1. Patterns of Infection

It is the mode of infection by different pathogens causing Mastitis in dairy cow varies in the mechanism and the resulting clinical subtypes. Three distinct modes of infection in the epidemiology of mastitis can be recognized as ***Contagious, Opportunistic/ Environmental and Vector infectious Patterns:***

Contagious disease pattern*:* it is the first and the most common mode where transfer of a microorganism from cow to cow is essential to propagate the disease. This pattern involves the transmission of disease from a carrier to a susceptible host. This involves mainly the spread of two major microorganisms in the dairy population. These microorganisms are *S. agalactiae* and *S. aureus*. Other epidemic contagious disease outbreaks have been reported, and involve Nocardia spp, Mycoplasma spp. and in some situations environmental streptococci. Contagious diseases only remain endemic when the mean number of susceptible individuals infected by an infected individual is appreciably larger than one (Mollalegn, *et al*., 2010). The reduction of the number of new infections is the major goal of prevention programs. New infections may be reduced by optimizing milking procedures and post milking teat disinfection. These practices will also reduce the number of shedders in the herd, separate the shedders from the uninfected cows, and optimize the immune function of the cow, which are key components of decreasing new infections. Eliminating existing infections reduces the exposure of susceptible quarters and may be obtained by treatment during lactation or at dry off, or by culling of the infected animals. Again, separation of the infected animals from the susceptible group may also be an effective method to limit the exposure of susceptible animals and reduce the risk of new infections (Dohoo and Leslie, 1991).

Opportunistic disease patterns: it is the second pattern is a pattern where infection is found mainly in housed or closely corralled cattle. Host factors and environmental factors put an animal at risk. A wide range of microorganism can then enter the mammary gland and cause disease. The causation of mastitis involves complex relationship of three major factors, and this involves an interaction of microorganism, host factors and the environment (Figure 1), and when solving herd problems, this epidemiologic triangle should always be kept in mind (Getachew and Gashaw, 2001). *A microorganism is* the most important microorganisms involved are the Coliforms (E.coli and Klebsiella spp.) and the environmental streptococci (Smith *et al*., 1985). Microorganism related factors (serum resistance, antigen determinants) are related to the severity of clinical mastitis. However, a problem solving approach only based on microorganism reduction is likely to fail. Most cow and environmental risk factors will remain. It is likely that another microorganism will fill the niche that is created by expelling one specific species. *Host factors* characteristic are important risk determinant in the pathogenesis of mastitis. These factors are associated with the development of specific immunity and with no specific herd defense mechanism (general resistance). This general resistance by the host relates to genetic predisposition, anatomic characteristics, nutritional status, and stage of lactation, parity and the use of management procedures to enhance resistance (Habbit *et al*., 1969). Several host factors are important in determining the outcome of an infection. Actually, most infection result in very little clinical signs, and host parameters like peripheral blood leukocyte activity,

blood leukocyte count, and presence of antibodies partially predict the outcome of infection (Treece *et al*., 1966). Other factors such as age of the animal, its metabolic status (noticeably ketosis), mineral nutrition (Selenium and vitamin E), peri-parturient stress and milk production level also affect the outcome of infection. Cows in early lactation appear to be specifically susceptible to clinical mastitis and have a relative high probability of becoming severely ill (Radostots *et al.,* 2007). Environmental factors are the several factors in the environment affect the exposure of a cow to microorganisms. Sources of environmental exposure are manure, bedding, feeds, dirt, mud and water. A good example of this is *E. coli,* which is present in the environment of the cow. Several studies have indeed linked the cleanliness of the barn, and the colony count in the bedding with the incidence of clinical mastitis (Bramley and Dodd, 1994). Of critical importance is hygiene in the dry period. Studies have shown that most infections with coliform and environmental streptococci take place in the last two weeks before calving, and often only show signs of clinical mastitis after calving.

Vector based patterns: The third type of infection pattern is found in non-lactating cows. This type of infection is common in the early part of the dry period particularly with Str. uberis and continues as clinical mastitis in the following lactation. This is 'summer' or 'heifer' mastitis and its etiology is the exposure to vector based pathogens carried by a species of fly (Myllys, 1 995).

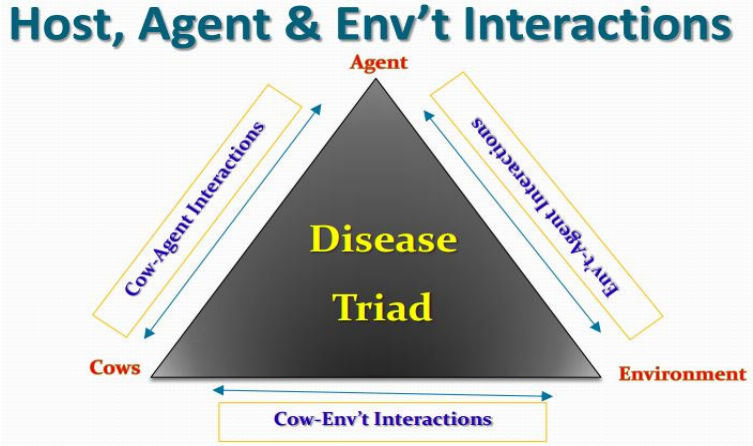

Figure 1: Epidemiologic disease triad (*host, agent & the environment interactions*).

2.3.2. Methods of Transmission

There are two main modes of transmission for bovine mastitis: contagious and environmental.

Contagious Transmission: Contagious transmission also called cow-to-cow transmission because; cows with mastitis are the main source of infection. Spread of the bacteria that cause the infection primarily happens during milking, e.g. via the milkers' hands, udder cloths, or the milking machine. Use of milking gloves and individual towels will help to prevent this (Zadoks *et al*., 2011). Milking infected cows last or with a separate unit will also prevent spread of infection, provided that the unit is disinfected before the next milking.

Environmental Transmission: Environmental mastitis originates in the environment, e.g. in bedding, manure or water. Occasionally, bedding contains high numbers of bacteria even before it is used. Routinely, bedding is contaminated with manure, which contains everything bacteria need moisture, warmth and nutrients. Water can be contaminated with bacteria from manure, or it may accumulate in a milking machine, particularly if hoses don't have the correct slope and if the temperature of the cleaning water is too low.

In addition to those common modes of transmission, there is also the possibility of transmission by people (Gurjar *et al*., 2012). Alternatively, people may be a direct source of infection for animals because many mastitis-causing bacteria occur in people as well as in cattle. The human-to-animal route of transmission has various names, none of which are particularly attractive humanosis, reverse zoonosis, or anthroponosis. An overview of transmission routes is shown in Figure 2 (Munoz *et al*., 2007).

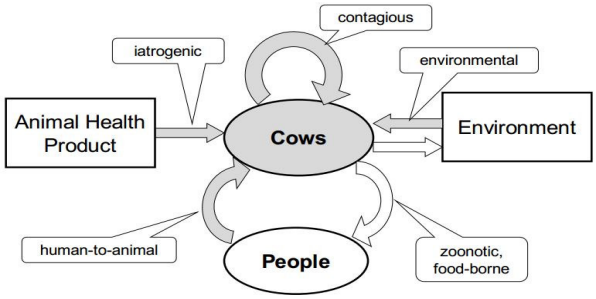

Figure 2: Possible sources and transmission routes for mastitis pathogens

Cows, the environment and contaminated products are not the only potential sources of mastitis pathogens. In rare cases, dogs or cats may be implicated, such as in the case of *Streptococcus canis* outbreaks (Tikofsky and Zadoks, 2005) but a more common occurrence is probably the introduction of mastitis pathogens by people. People and cattle share many bacteria *Strep. agalactiae*, *Staph. aureus*, *Staphylococcus epidermidis*, *E. coli* and *Klebsiella*. People are the natural host for two important staphylococcal species *Staph. aureus* and *Staphylococcus epidermidis*. Both species can be transmitted from people to cows with subsequent cow-to-cow transmission (Zadoks *et al*., 2011).

2.4. Associated Risk factors

Genetic aspects of mastitis*:* Susceptibility or resistance to mastitis in dairy cows has a genetic basis. Selection for increased milk yield has been shown to have a detrimental effect on the health condition of the mammary gland (Zadoks *et al*., 2011). The disease is thus a problem mainly in high-production herds raised in intensive farming conditions.

Type of purpose and breed of cows*:* Mastitis is a common problem and the most onerous one facing milk producers all over the world. It can appear at any time in any dairy herd, even in the most organized ones, so every herd is potentially at risk (Elbably *et al*., 2013). The mammary gland in beef cows serves to produce the milk necessary to feed their young. The udder of nursing cows is naturally emptied by the calves several times or even more than a dozen times a day and as a rule no mechanical damage to the teats or acute infections are observed. For this reason the occurrence of mastitis is not as carefully monitored in beef cow herds.

Udder structure: Udders with unequally developed quarters are most susceptible to infections. More frequent occurrence of inflammation is also associated with the presence of long teats, which are at greater risk of damage (Awale *et al*., 2012). Waller *et al*. (2014) report that cows with a pendulous udder after calving or an udder with long, funnel-shaped teats or inactive quarters are at a high risk of subclinical mastitis. Cows with mastitis have been observed to have smaller teats and shorter teat canals, with a larger teat base and diameter (Hussain *et al*., 2012). The disease is also linked to the cross-sectional area of the teat canal (Awale *et al*., 2012). The larger the cross-sectional area, the less elastic are the muscles closing it, so that it remains open longer after milking and the susceptibility to infection increases.

***Age of cows and stage of lactation*:** Both the age of cows and the stage of lactation are important factors associated with the occurrence of mastitis. It can be linked to the fact that the teat canal in older animals is wider due to frequent milking over several years, or it may permanently remain partly open. The age of cows may also affect the permeability of the mammary epithelium, which can be linked to damage caused by previous inflammatory states leading to increased permeability even after successful treatment (Król *et al*., 2013).

According to Parker *et al*. (2007), in heifers with an infected udder before parturition the risk of clinical mastitis after calving is as much as 4 times greater than in uninfected cows. The problem of clinical mastitis in pregnant heifers and primiparous cows during calving time has also been discussed and mastitis cases are particularly frequent immediately after parturition, in early lactation and during the first 2–3 weeks of the dry period (Borm *et al*., 2006; Malinowski and Smulski, 2007). This is probably due to increasing oxidative stress and lower efficiency of antioxidant defense mechanisms at the start of lactation.

Milking system and hygiene: Observing hygiene during milking is particularly important in counteracting and combating infectious mastitis in the herd. Shittu *et al*. (2012) noted that in cows kept in herds in which milking is conducted in a traditional manner and hand-washing before milking is a frequent practice, the risk of development of subclinical mastitis was lower than in cows in herds in which hand-washing was not observed. One of the causes they suggested was failure to recognize various types of soiling of the udder by the milking device.

Dry period*:* The dry period is considered to be one of the most important periods influencing the health condition of the udder in cows, and is also a time of increased risk of mastitis (Mulugeta and Wassie, 2013). During this period dynamic changes take place in the physiological, metabolic and hormonal status of the cow, primarily regeneration of the gland tissue of the udder. The optimal length of the dry period for a 305 day lactation period is considered to be 40–60 days. The elimination or shortening of the dry period reduces resistance of the udder to inflammatory states, leading to an increase in somatic cell count in the milk in the subsequent lactation.

Body condition score of animals: High-yielding dairy cows usually exhibit a negative energy balance after calving, which may influence the immune system and the metabolic system of the individual, chronic deficiencies of energy, protein, minerals, or vitamins have repeatedly been associated with increased disease susceptibility as a result of depressed immune function.

Because most udder infections occur in the peri-partum period, optimal feeding, both in the dry period and during early lactation, may be important in preventing mastitis. It seems difficult for the high-producing dairy cow to ingest enough feed shortly after calving to meet lactation demands for energy and protein. Cows with high BCS at calving lose more condition and achieve positive energy balance later than do cows with lower

BCS (Myllys *et al*., 1995). It is suggested that there might be differences in the length and severity of the immunosuppressive period after calving, which may influence the risk for mastitis.

Milk yield: A high 305-day previous-lactation milk yield was a significant risk factor for early lactation clinical mastitis and high yields increased the mastitis rate in low bulk milk somatic cell count herds (Tesfaye, 2009). A high milk protein content at the last milk-test day prior to drying-off was been found to be a risk factor for early lactation CM. This may reflect higher energy supplies to the udder and lead to delayed involution of the udder tissue.

Hygiene scoring: The environment in which dairy cows are kept has a decisive effect on their health and welfare. A clean and comfortable shelter represents the key to maintaining the dairy cows' health and longevity. The shelter's hygiene level can be evaluated through several assessment systems based on the quantification of the manure pollution in different body regions of the cows (Myllys *et al*., 1995).

For dairy cows the outcomes of low hygiene are the high risk of mastitis and the worsening of lameness. The relation between shelter hygiene, clean cows and low number of somatic cells in mixed milk were indicated in studies (Delelesse, 2010). At the herd level, choices in bedding type, slurry removal from alleys or housing type may be global hazards that affect the frequency of environmental mastitis.

2.5. Pathogenesis

A comprehensive understanding of the pathogenicity of mastitis is key for the development of appropriate detection techniques. The primary cause of mastitis is a wide spectrum of bacterial strains; however, incidences of viral, algal and fungal-related mastitis were also reported (Eriksson, 2005). Inflammation of the mammary gland predominantly occurs via the teat canal except in the case of tuberculosis, leptospirosis and brucellosis where the method of spread may be haematogenous.

The development of mastitis can be explained in terms of three stages as invasion, infection and inflammation. The invasive stage refers to the time in which pathogens move from the teat end to the milk through the teat canal. The infection stage is the stage in which the pathogens multiply rapidly and invade the mammary tissue. The stage of inflammation is the stage with varying degrees of clinical abnormalities of the udder and with systemic effects from mild to per acute as well as gross and subclinical abnormalities of the milk (Radostits *et al*., 2007).

The inflammatory response is initiated when bacteria enter the mammary gland and this is the body's second line of defense. These bacteria multiply and produce toxins, enzymes, and cell-wall components which stimulate the production of numerous mediators of inflammation by inflammatory cells. The magnitude of the inflammatory response may be influenced by the causative pathogen, stage of lactation, age, immune status of the cow, genetics, and nutritional status (Harmon, 1994).

Polymorphnuclear Neutrophil (PMN) leukocytes and phagocytes move from bone marrow towards the invading bacteria and are attracted in large numbers by chemical messengers (chemotactic agents) from damaged tissues. Masses of PMN may pass between milk producing cells into the lumen of the alveolus, thus increasing the somatic cell count (SCC) as well as damaging secretory cells. Somatic cells consist mainly of PMN (white blood cells). At the infection site, PMN surround the bacteria and release enzymes which can destroy the organisms. The leukocytes in milk may also release specific substances that attract more leukocytes to the area to fight the infection. Numbers of somatic cells remain in large concentrations after bacteria are eliminated until healing of the gland occurs. Clots formed by the aggregation of leukocytes and blood clotting factors may block small ducts and prevent complete milk removal. Damage to epithelial cells and blockage of small ducts can result in the formation of scar tissue in some cases, with a permanent loss of function of that portion of the gland. In other cases, inflammation may subside, tissue repair may occur, and function may return in that lactation or the subsequent one (Harmon, 1994).

2.6. Diagnosis

***Qualitative Milk examination*:** According to Quinn *et al.* (1994) changes in color of milk can be caused by the presence of blood (red or brownish) or pus (yellow). The consistency may be increased, resulting in thicker, "sticky" milk, or it may be more than usually watery. Flakes and clots are always abnormal. The smell of the secretion may also be altered as a result of mastitis. And, *udder Visualization and Palpation,* the primary step in preventing mastitis is regular examination (*visualization and palpation*) of the udder especially during milking. In clinical mastitis, visually the udder may turn red, hard and hot to touch. Udder may be painful to the cow at the time of palpation. These symptoms show the changes in vascularity and blood flow of the gland when inflamed (Mulugeta and Wassie, 2013). At the time of milking, Milk Visualization is also the necessary step to appreciate the presence of gross changes in the milk such as the flakes, clots or serous, blood and watery secretions. And, if these changes are present, it is the common means for clinical mastitis detection. Flakes or clots in the milk are detected by stripping the first few squirts of milk from each quarter into a strip cup at the beginning of milking. Milk turns very much watery during dry period and also a change in milk composition is

observed. In chronic case, the appearance of udder secretion is usually abnormal at intervals. The secretion becomes grossly altered in case of acute mastitis.

***Strip Cup or Plate Test*:** it is another cow side test and can be used for determining the presence of clinical mastitis through the detection of visible particles of milk. It is a practical and effective method of identifying cows with clinical mastitis. Any lay man can make use of this strip cup. In this test an enamel plate divided in four strip cups is used and the bottom of the plate is black in color so that the milk flakes are easily observed by tilting the cups at an angle. Abnormal milk is usually discolored, watery or contains flakes, shreds or clot (Radostits *et al*., 2007).

California Mastitis Test (CMT): The CMT is an easily everywhere applicable and reliable screening test for sub clinical mastitis. It has been developed to test milk from individual quarter and also bulk milk samples. Using the CMT, fresh, unrefrigerated milk can be tested for up to 12 hrs and refrigerated milk can be tested for up to 36 hrs to get reliable readings. The test helps to assess the level of infection in each quarter rather than to an overall udder result, and the result shows only whether the cell count is high or low. According to the National Mastitis Council (1990) described test procedure, squeezing about 2ml of milk from each quarter and adding an equal amount of CMT solution to each cup in the plastic CMT paddle. Thoroughly mixing the contents by rotating the paddle in circular motion and within 15 seconds of mixing the content, the CMT reaction must be scored but exceeding this time results in disappearance of weak reactions. The reagent used in the CMT is a detergent and bromoserol purple as an indicator of pH. The degree of reaction between the detergent and the DNA of cell nuclei determines the number of somatic cells in milk.

Flow Cytometry (FC): a method by which physical and chemical characteristics of cells or particles can be measured as they travel in suspension past a sensing point. This method has been developed recently to quantify Somatic Cell Counts in milk, and is particularly good for detecting subclinical mastitis (Tian *et al.*, 2005; Holm *et al.*, 2004).

***Culture Method*:** The surest way of diagnosing mastitis is by directly isolating and identifying any pathogenic microorganisms which may be present in the milk. This can be achieved by cultural methods and a number of additional determinative tests. To obtain correct results and avoid contamination and hence bias, it is important to work as securely and as accurately as possible under the circumstances (Quinn *et al*., 2002).

3. TREATMENT REGIMEN FOR MASTITIS

3.1. Treatment of clinical mastitis in practice

Treatment of mastitis should be targeted towards the causative bacteria whenever possible, but in acute situations, treatment is initiated based on herd data and personal experience. Rapid or on farm bacteriological diagnosis would facilitate the selection of the most appropriate antimicrobial. Treatment protocols and drug selection for each farm should be made by veterinarians familiar with the farm (Sawant *et al*., 2005; Wagner and Erskine, 2006). The use of on-farm written protocols for mastitis treatment can promote judicious use of antimicrobials (Raymond *et al*., 2006; Passantino, 2007). Therapeutic response of the cows can be monitored using individual somatic cell count data if available, or using the California Mastitis Test, and with bacteriological samples in herds with contagious mastitis. In general, the use of narrow-spectrum antimicrobials is preferable. Prudent use guidelines have been developed which also include antimicrobial treatment of mastitis (Anonymouse, 2003; Passantino, 2007). First choice antimicrobials for treating mastitis caused by streptococci and penicillin-susceptible staphylococci are β-lactam antimicrobials, particularly penicillin G. Broad-spectrum antimicrobials such as third or fourth generation cephalosporin should not be used as first alternatives for mastitis, as they may increase emergence of broad spectrum β-lactam resistance. Systemic treatment is recommended in clinical mastitis due to *S. aureus* and in severe cases of coliform mastitis, preferably in combination with IMM treatment (Barkema *et al*., 2006). Too short a duration of standard treatment is probably an important reason for poor cure rates in mastitis therapy. A longer treatment improves cure rates, and duration of treatment should generally be extended in mastitis caused by *S. aureus and Streptococcus uberis* (Oliver *et al*., 2004; Deluyker *et al*., 2005). Clinical mastitis should be treated for at least three days; this recommended treatment duration is longer than label treatments in many countries. All mastitis treatment should be evidence based i.e., the efficacy of each product and treatment length should be demonstrated by scientific studies (Cockcroft and Holmes, 2003).

3.2. Treatment of Subclinical mastitis

Treating subclinical mastitis with antimicrobials is generally not economical during lactation because of high treatment costs and poor efficacy. In a study with a large number of subclinical mastitis cases (Wilson *et al*., 1999) the overall bacteriological cure rate for antimicrobial treatment was 75% and that for no treatment 68%. The marginal benefit applied for streptococcal mastitis only; in mastitis due to *Staphylococcus aureus*, antimicrobials were equal to no treatment. Treatment of subclinical mastitis will not affect the incidence of mastitis in the herd unless other preventive measures are taken. Studies on treating cows based on high somatic cell counts have generally shown that no effect on milk production has been achieved (Shephard *et al*., 2000,

Hallen *et al*., 2008). In herd problems caused by very contagious bacteria such as *S. aureus* or *Streptococcus agalactiae* treatment of subclinical mastitis is advised (Wagner and Erskine, 2006).Antimicrobials have been used to treat mastitis for more than fifty years, but consensus about the most efficient, safe, and economical treatment is still lacking. The concept of evidence-based medicine has been introduced to veterinary medicine (Cockcroft and Holmes, 2003) and should apply also to treatment of mastitis. The impact on public health should be taken into account as dairy cows produce milk for consumption (OIE, 2008). Antimicrobial treatment of dairy cows creates residues into milk, and therefore residue avoidance is an important aspect of mastitis treatment (Wagner and Erskine, 2006). Selecting a substance with a low minimum inhibitory concentration value for the target pathogen is preferable, particularly when the antimicrobial is administered systemically. The antimicrobial should have bactericidal rather than bacteriostatic action, because phagocytosis is impaired in the mammary gland (Kehrli and Harp, 2001).Antimicrobial susceptibility determined *in vitro* has been considered as a prerequisite for treatment. However, activity *in vitro* does not guarantee efficacy *in vivo* when treating bovine mastitis. Antimicrobial resistance amongst mastitis pathogens has not yet emerged as a clinically relevant issue, but geographical regions may differ in this respect. The biggest problem is the widespread resistance of staphylococci, particularly *Staphylococcus aureus*, to penicillin G (Olsen *et al*., 2006; Hendriksen *et al*., 2008). Cure rates for mastitis caused by penicillin-resistant strains of *S. aureus* seem to be inferior to those of mastitis due to penicillin-susceptible strains (Sol *et al.*, 2000 and Taponen *et al*., 2003). It is not known if this is due to pharmacologic problems of the drugs used, or virulence factors possibly linked to β-lactamase gene of the resistant isolates (Haveri *et al*., 2005). Using an *in vitro* β-lactamase test for determining resistance to penicillin G of staphylococci before treatment is recommended (Olsen *et al*., 2006). *Coagulase negative staphylococci* tend to be more resistant th n *S. aureus* and easily develop multi resistance (Sawant *et al.,* 2009). Mastitis causing *streptococci* have remained susceptible to penicillin G, but emerging resistance to macrolides and lincosamides has been detected (Loch *et al*., 2005).

4. CONTROL AND PREVENTION

Speaking of control, it usually meant to refer to *contagious mastitis* and *environmental mastitis*. And, the fundamental principle of mastitis control is controlling the disease by either decreasing the exposure of the teat to potential pathogens or by increasing resistance of dairy animals to infection (Joren, 2009). It has been suggested that the mastitis control and treatment approach should be in line as in the same way as a surgeon approaches doing for surgery. In view of that, the key elements need to be considered the approach cuddles sound husbandry practices and sanitation, post-milking teat dipping, treatment of mastitis during non-lactating period, and culling of chronically infected animals.

To speak said approach precisely, washing hands with soap and water, washing teats and udder in sanitizing solution, thoroughly drying teats and udder with individual towels, dipping teats in an effective germicidal teat dip, allowing 30 seconds of contact time before wiping off teat dip with an individual towel, thoroughly scrubbing the teat end with a cotton swab soaked in alcohol. If all four quarters are being treated, start by cleaning the teat farthest from the nearest and work towards the closest teat. Treat teats nearest to the milkers first, then those farthest away to prevent contamination of the clean teat ends, and dipping teats in an effective germicidal teat dip after treatment (Smith and Hogan, 1993).

4.1. Contagious Mastitis Control

Staphylococcus aureus infections remain the largest mastitis problem of dairy animals. Cure rate with antibiotic therapy during lactation is very low. Many infected animals become chronic eases and have to be culled. *Streptococcus agalactiae* respond well to antibiotic therapy and can be eradicated from dairy herds with good mastitis control practices, including teat dipping and dry animal treatment. *Streptococcus dysgalactiae* may live almost anywhere in the udder, rumen, and feces, and in the barn. They can be controlled with proper sanitation and are moderately susceptible to antibiotics (Hogan *et al*., 1999).

In general, contagious mastitis is controlled in dairy herds by post milking teat dipping, total dry cow therapy, culling, therapy of clinical cases, and proper maintenance of milking equipment (Radostits *et al*., 2000).

4.2. Environmental Mastitis Control

Control of environmental mastitis is achieved by reducing exposure of teat ends to environmental pathogens and by maximizing the resistance of the cow to intra-mammary infection. Significant sources of environmental pathogens are organic bedding materials, manure, and wet or damp areas in barns, or pastures. Milking time hygiene can influence teat end exposure. In general, exposure is minimized when all areas of the environment are clean, cool and dry. The animal should have no access to manure, mud, or pools of stagnant water and calving area must be clean. Resistance is maximized by providing a stress free environment that minimizes teat injury, and by feeding balanced diets rich in vitamin E and selenium. Post milking teat dipping with a germicidal dip is recommended. Attempts to control environmental mastitis during dry period, using either germicidal or

barrier dips, have been unsuccessful, and antibiotic therapy during lactation or the dry period is of little value in the control of environmental mastitis, but rather at drying off for all quarters of all animals such that it helps to control environmental streptococci during the early dry period (Hettinga, 2008).

5. IMPORTANCE OF MASTITIS

5.1. Economic importance

Effect on Milk Composition: Mastitis resulting from major pathogens causes considerable compositional changes in milk including increases in SCC. The types of proteins present change dramatically. Casein, the major milk protein of high nutritional quality, declines and lower quality whey proteins increase which adversely impacts dairy product quality, such as cheese yield, flavor and quality. Serum albumin, immunoglobulin, transferrin, and other serum proteins pass into milk because vascular permeability changes. Lactoferrin, the major antibacterial iron-binding protein in mammary secretions, increases in concentration, which is likely because of an increased output by the mammary tissue and a minor contribution from PMN (Myllys and Rautala, 1995). Milk protein breakdown can occur in milk from cows with clinical or subclinical mastitis due to presence of proteolytic enzymes. Plasmin increases proteolytic activity by more than two fold during mastitis. Plasmin and enzymes derived from somatic cells can cause extensive damage to casein in the udder before milk removal. Deterioration of milk protein as a result of mastitis may continue during processing and storage. Mastitis increases the conductivity of milk and sodium and chloride concentrations are elevated. Potassium, normally the predominant mineral in milk, declines. Because most calcium in milk is associated with casein, the disruption of casein synthesis contributes to lowered calcium in milk (Harmon, 1994).

Effect on Cost: Mastitis is of great economic importance to milk producers, because the disease has negative impact on several important aspects of cow and herd performance. Incurred costs are of both direct and indirect nature (Makovec and Ruegg, 2003). Direct costs include veterinary costs, increased labour requirement, discarded milk (during the course of treatment), and reduced milk yield and quality. Indirect costs are those that are not always obvious to the milk producer, and are therefore referred to as hidden costs. They include increased risk of subsequent disorders, reduced fertility (extra services per conception and, as a result of this, an extended calving interval), increased risk of culling, and, occasionally, mortality (Radostits *et al.*, 2007).

5.2. Public health importance

With mastitis there is a danger that the bacterial contamination of milk from affected cows may render it unsuitable for human consumption by causing food poisoning and provides a mechanism of spread of disease to humans through consumption of raw milk. Many farm families simply consume raw milk because it is a traditional practice and it is less expensive to take milk from the bulk tank than buying pasteurized retail milk. Some believe that raw milk has a higher nutritional value than pasteurized milk (Hodges *et al*., 1984). Raw (unpasteurized) milk has been found to participate in spreading out of illnesses caused by *Mycobacterium bovis*, *Brucella abortus*, *Staphylococcus aureus*, *Listeria Monocytogenes, Campylobacter jejuni, Salmonella, Staphylococci species,* and *E. coli.* With severe clinical mastitis, abnormalities of milk are easily observed and milk is discarded by the producer. Such milk normally would not enter the food chain. But when milk of cows with subclinical mastitis, which is with no visible changes, is accidentally mixed into bulk milk, it enters into food chain and can be dangerous to humans. The principal bacterial infection associated with ingestion of milk and milk products are caused by different bacterial genera. The reservoir of these bacteria may be milk producing mammals, humans or contamination from environmental reservoirs. In diseases such as brucellosis, salmonellosis or tuberculosis milk may be contaminated with the causative bacteria before it leaves the infected udder. The selection of milk borne pathogen is difficult because of the different importance in different countries. However, in general Brucella species, Mycobacterium species, *Listeria monocytogenes*, *Campylobacter jejuni* and *Staphylococcus aureus* seem worth to be as milk borne zoonosis (Heeschen, 2012).

The presence of residue in milk following treatment of mastitis is a major public health concerns that adversely affects the dairy industry, the practicing veterinarian and the safety of milk for human consumption. Consumption of antibiotic contaminated milk results in allergic responses, changes in intestinal flora and development of antibiotic resistant pathogenic bacteria (Joren, 2009).

6. CONCLUSION AND RECOMMENDATION

Mastitis in dairy cows is a serious problem as it is an economically devastating disease causing immense economic losses in the dairy industry and is the worldwide costliest production disease in dairy herds. Subclinical mastitis is the most serious type as the infected animal shows no obvious symptoms and secrets apparently normal milk for a long time, during which causative organisms spread infection in herd, so it is an important feature of the epidemiology of mastitis.

Continuous monitoring of mastitis, and its careful management, is essential for the well-being of a dairy herd. This can be achieved through the detection of inflammation at its early stages and, subsequently, the

detection and treatment of the mastitis infection. Most bacterial species can spread via more than one route, including from cow to cow or from the environment to the animal. In some cases, an environmental point source such as a treatment product or even a dog, cat or person acts as the source of infection and in those situations, initial introduction from the environment may be followed by cow-to-cow transmission. Both removal of the original source and prevention of further within-herd spread are needed to control such outbreaks. Age groups, pendulous udder conformation, multiple parity, poor body condition score, bad hygiene score, high milk producers, early lactation stage, previous exposure to mastitis and blind teats are the associated potential risk factors.

Based on this, the following points are recommended

- Dairy producers must have to be willing to change old habits or ineffective/incorrect practices that may be causing or permitting new intramammary infections (IMIs) to occur.
- Milker's should be trained on proper hygienic milking methods,
- Regular investigation of mastitis especially sub clinical form should be practiced
- Mastitis treatments should be preceded with identification of the causative agent and susceptibility test profile of pathogens and
- Culling of old aged and repeatedly infected cows should be done on regular planned basis.
- Management, housing and environmental sanitation should be improved

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