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Bovine Mastitis: An Asian Perspective

[1]Neelesh Sharma, [6]Gyu Jin Rho, [4]Yeong Ho Hong, [3]Tae Young Kang, [5]Hak Kyo Lee, [7]Tai-Young Hur and [1,2]Dong Kee Jeong

[1]Department of Animal Biotechnology, College of Applied Sciences, Jeju National University, Jeju, South Korea
[2]Research Institute of Subtropical Agriculture and Biotechnology, Jeju National University, South Korea
[3]College of Veterinary Medicine, Jeju National University, Jeju, South Korea
[4]Department of Animal Science and Technology, College of Natural Science, Chung-Ang University, Anseong, South Korea
[5]Genomic Informatics Center, Hankyong National University, Ansung, South Korea
[6]College of Veterinary Medicine, Gyeongsang National University, Chinju, South Korea
[7]National Institute of Animal Science, Rural Development Administration, Cheonan, South Korea

Corresponding Author: Dong Kee Jeong, Department of Animal Biotechnology, College of Applied Sciences, Jeju National University, Ara-1 Dong, Jeju-city, Jeju-Do 690-756, South Korea Tel: 82-64-754-3331 Fax: 82-64-725-2403

ABSTRACT

Bovine mastitis is an inflammatory disease of cow and buffaloes mammary gland caused by various infectious or non-infectious etiological agents. Mastitis must have been one of the first observed disease of farm animals when cattle were domesticated over 5000 years ago. Since then it will have been an ever present problem for all those who kept and milked dairy cattle and buffaloes. The dairy industry in particular, plays a strong role for the livelihood of poor people because agriculture land is going to shrink as a results dependency of farmers is increasing towards dairy sector. The buffalo population in Asia has multiplied over the past half a century, by a factor of 2.5 rising by almost 2.2 billion in absolute numbers and at an average annual pace of over 1.8%. Over 3.9 billion, roughly 60% of the world population, reside in Asia. Bovine mastitis is one of the important production diseases of dairy animals which directly or indirectly affect the economy of the farmers and ultimately affect the economy of the country. However, mastitis is a global problem as it adversely affects animal health, quality of milk and economics of milk production and every country including developed ones suffer huge financial losses. In Asia, major mastitis causing organisms are *Staphylococcus aureus*, Streptococci, *E. coli*, *Corynebacterium* spp. and *Klebsiella* spp., recent reports indicating the changing trends from *Staphylococcus aureus* to Coagulase Negative Staphylococci (CNS) as major mastitis causing organism. The pattern of mastitis occurrence in Asia is also significantly increasing in both cattle and buffaloes which is a major challenge for policy makers, field veterinarians and researchers.

Key words: Bovine mastitis, etiology, prevalence, Asia

INTRODUCTION

Bovine mastitis, defined as inflammation of the mammary gland, can have an infectious or non-infectious etiology (Bradley, 2002). It is characterized by physical, chemical and usually bacteriological changes in milk and pathological changes in glandular tissues of the udder and affects the quality and quantity of milk (Radostits *et al*., 2000; Sharma *et al*., 2012). It is also defined as inflammation of mammary gland parenchyma which is caused by microorganisms, usually bacteria that invade the udder, multiply and produce toxins which are harmful to the mammary gland (Sharma *et al*., 2006). Mastitis must have been one of the first observed disease of farm animals when cattle were domesticated over 5000 years ago. Since then it will have been an ever present problem for all those who kept and milked dairy cattle and buffaloes. In first books published regarding mastitis problems (17th century) its importance was stressed and its association with poor cattle management and particularly with leaving cows half milked. From 19th century there has been constant stream of scientific and professional papers in veterinary and animal science literature on mastitis causes and control. Scientific research followed Pasteur's demonstration of the 'germ theory of disease' in 1860 and by 1900 it was established that most types of mastitis followed microbial infection.

Livestock and its inputs are a growing economic sector. The livelihood and income effects of the livestock economy are huge. More than a billion people keep livestock, 60% of rural households do so. It's a major income source of the poor and especially of women in developing countries. The dairy industry in particular, plays a strong role for the livelihood of poor people because agriculture land is going to shrink as a results dependency of farmers is increasing towards dairy sector. The livestock capital as part of the overall agricultural capital amounts to more than a quarter. Land valued at market prices is about half. Livestock capital is therefore a very important element of the overall capital stock of world agriculture (Braun, 2010).

The buffalo population in Asia has multiplied over the past half a century, by a factor of 2.5 rising by almost 2.2 billion in absolute numbers and at an average annual pace of over 1.8%. Over 3.9 billion, roughly 60% of the world population, reside in Asia (Cruz, 2010). The 2008 buffalo milk production in Asia represents 96.78% of the total volumes of world's buffalo milk of 89.2 Million tons. Production in South and Southwest Asia primarily from India and Pakistan contributed a hefty 93.17% (FAO, 2010). Buffaloes are significant sources of milk in this sub-region contributing as high as 68.35% of the total milk yield in Pakistan and 56.85% in total milk production in India. The trend of milk production in some Asian countries is given in Fig. 1. In India, the dairy sector's

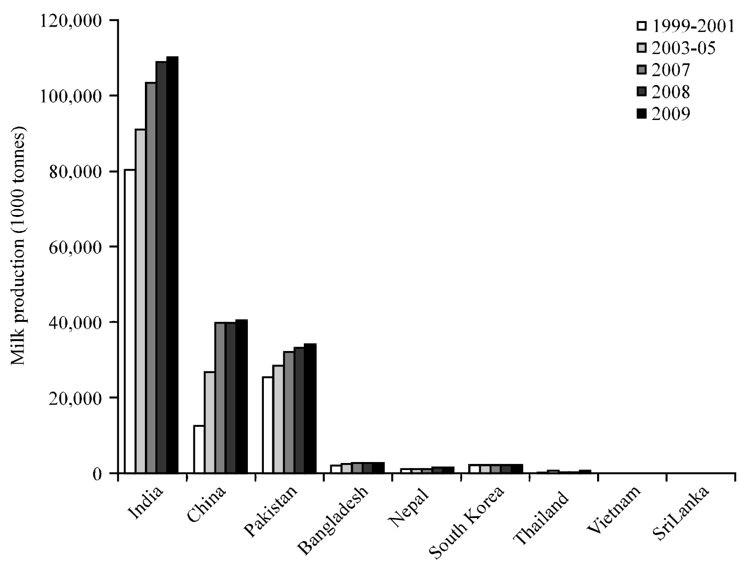

Fig. 1: The milk production trend in Asian coutnries

growth depended in large part on the use of buffalo which, unlike high-yielding dairy cattle are well adapted to tropical climes. Today, across India, more than half of all milk is produced from buffalo. Cross-bred cattle numbers are increasing but they still account for less than 14% of the total cattle population (FAO, 2009).

Bovine mastitis is one of the important production diseases of dairy animals which directly or indirectly affect the economy of the farmers and ultimately affect the economy of the country. In dairy cattle mastitis results in severe economic losses from reduced milk production, treatment cost, increased labor, milk withheld following treatment and premature culling (Miller *et al*., 1993). Mastitis continues as a problem in many dairy herds despite proper application of proven control methods of teat dipping and total dry cow therapy. In general, mastitis is a complex disease dealing with, the interaction of microorganisms and the cow's anatomy and physiology, dairy husbandry and management, milking equipment and procedures and environment (Woods, 1986). Because of udder, anatomical position are subject to outside influence and are prone to both inflammation and non-inflammatory conditions (Sharma, 2007). Infectious mastitis results from the introduction and multiplication of pathogenic microorganisms in the mammary gland and this leads to a reduced synthetic activity, changes in the milk composition and elevated milk Somatic Cell Count (SCC) (Harmon, 1994). The bacterial contamination of milk from the affected cows render it unfit for human consumption and provide a mechanism of spread of diseases like tuberculosis, sore-throat, Q-fever, brucellosis, leptospirosis etc. and has zoonotic importance (Sharif *et al*., 2009). It is well known from several previous studies that the incidence and the patterns of causative agents and disease occurrence markedly differ from place to place, herd to herd and time to time. The prevalence of bovine mastitis ranged from 29.34 to 78.54% (Sharma and Rai, 1977; Ebrahimi *et al*., 2007; Sharma and Maiti, 2010) in cows and 27.36 to 70.32% (Sharma *et al*., 2004, 2007; Beheshti *et al*., 2010) in buffaloes. Detailed analysis of previous studies conducted in Pakistan (Athar, 2007) revealed that highest prevalence of clinical and sub-clinical mastitis in cattle and buffaloes was due to *S. aureus* with a mean of 46.72%.

The vehement research on bovine mastitis is comporting since past 7 decades but unfortunately the problem is still challengeable for the bovine mastitis researchers and particularly for field veterinarians to treat and control it. Now there is a need to imply the strategic control measures worldover to control this deadly disease of dairy animals to prevent heavy economic losses of farmer's. To understand the detail mechanism of mastitis we need to compile the data on all aspects of mastitis from different parts of the world and, to aware the researcher and farmers with updates on mastitis. We need distribution and changing trend of etiological agents and prevalence of mastitis in different countries of the world to apply strategic plan for control of mastitis. In this study, we are compiling basic updated information and data particularly on distribution of etiological agents and prevalence, from some Asian countries on this important worldwide issue.

WHY BOVINE MASTITIS IS IMPORTANT?

Mastitis is the most economically important disease of dairy cattle, accounting for 38% of the total direct costs of the common production diseases (Kossaibati and Esslemont, 1997). Mastitis is a global problem as it adversely affects animal health, quality of milk and economics of milk production and every country including developed ones suffer huge financial losses (Sharma *et al*., 2007). It is the most important deadly disease of dairy animals is responsible for heavy economic losses due to reduced milk yield (up to 70%), milk discard after treatment (9%), cost of veterinary services (7%) and premature culling (14%). India is the highest milk producer in the world but the per capita availability of milk still remains half of the world average, demanding strategic intervention. One of the reasons for low productivity is poor animal health, particularly, mastitis which is single largest problem in dairy animal in terms of economic losses in India. It is proved by the reports that the annual economic losses due to bovine mastitis was increased 114 folds in about 4 decades from 1962 (INR 529 million/annum) (Dhanda and Sethi, 1962) to 2001 (INR 60532 million/annum) (Dua, 2001). The dramatic increase in the economic losses due to mastitis, divert the mind of researchers, policy makers and dairy farmers toward this costliest disease to control it. In addition to heavy losses in milk quality and quantity, it also causes irreversible damage to the udder tissue and less occasional fatalities (Radostits *et al*., 2000). Mastitis destroys the milk-secreting cells. Scar or connective tissue replaces the milk secreting tissue, resulting in a permanent loss of productive ability. Mastitis can lead to the reduction of offspring to a given production system due to the insufficient milk production resulting in starvation.

In South Korea, there are approximately 8,000 dairy farms and 472,000 cows, yielding an average of 177,770,000 kg of raw milk per year. The degree of self-sufficiency of milk produced in Korea is approximately 70%, so managing and preventing bovine mastitis is an inevitable task (Park *et al*., 2007). Dhakal and Thapa (2002) reported that about 68% of the total losses resulted from drop in milk production in buffaloes in Nepal. In Pakistan, losses due to clinical mastitis were estimated to INR 240 million per annum in Punjab only during 1978 (Chaudhry and Khan, 1978). This estimate might be much higher if losses due to sub-clinical mastitis (which is 15-40 times more prevalent than clinical form) had been included. The work on bovine mastitis economics is very scaring in Asian countries; hence, enough economic data on this disease is not available except few reports. Mastitis has significantly constrained the development of the dairy industry in Bangladesh (Islam *et al*., 2011). It has been estimated that annual economic losses due to mastitis in the US, $1.5 to 2.0 billion. Apart of its economic importance it also carries public health significance (Sharma *et al*., 2003).

RISK FACTORS OF MASTITIS

Bovine mastitis is a multi-factorial disease which is closely related to the production system and environment in which the cows and buffaloes are kept. Risk factors such as management practices (shed and udder hygiene, poor teat condition, poor environmental hygiene, sanitation, large herd size, use of hand wash cloth, improper teat dipping), host (breed, high yielder, udder immunity, teat lesions, genetic resistance) and diet (Cu, Co, Zn, Selenium and vitamin E deficiency) amongst others have been reported to be important in the prevalence and epidemiology of both clinical and sub-clinical mastitis. To simplify understanding of the mastitis complexity, it is useful to consider risk factors or disease determinants which are broadly classified into three groups- host (cow or buffalo), pathogen (micro-organisms) and environment (Fig. 2).

Breed: As demand of milk and milk products increased, the most of developing countries are focusing on the development of high producing new breeds of dairy cows especially Holstein to fulfill the need. Holstein breed of cattle is the most widespread dairy cattle breed, found in 128 countries worldwide (FAO, 2007).

The Boran breed which is recognised for its relatively high milk yield compared to other cross-breeds, has been shown to be 3.4 times more likely to be affected by clinical and sub-clinical mastitis than the Tanzanian Shorthorn Zebu breed (Karimuribo *et al*., 2006). Increased risk of

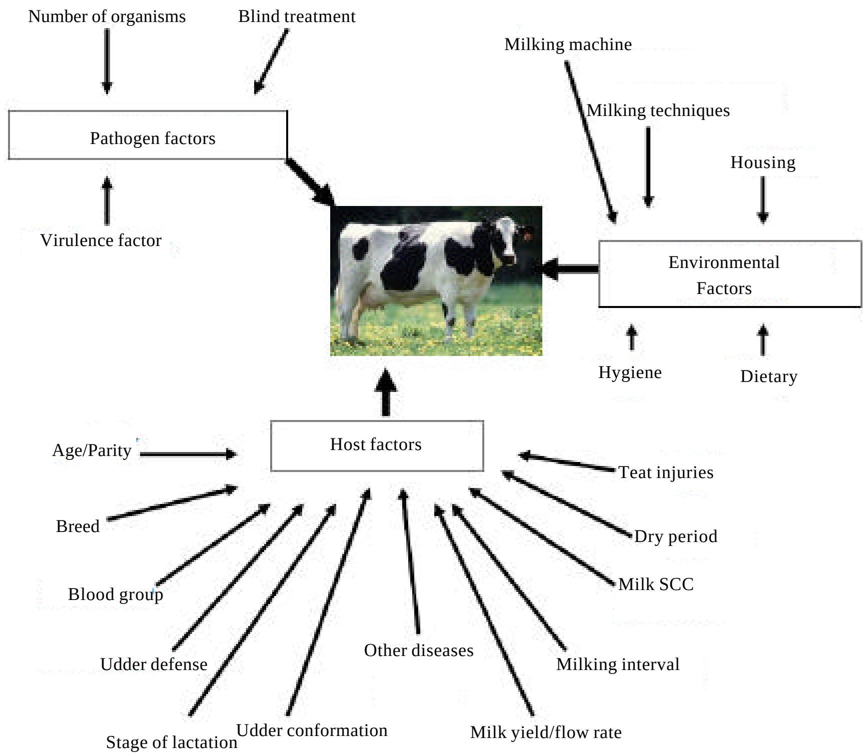

Fig. 2: Risk factors for bovine mastitis

clinical mastitis in Friesian compared with Jersey and Ayrshire heifers (Compton *et al.*, 2007; Myllys and Rautala, 1995). In Southern Ehiopia, Biffa *et al.* (2005) reported that Holstein-Fresian cows are affected at a higher rate (56.5%) compared with local zebu (30.9%) and Jersey cows (28.9%). Sharma and Maiti (2010) also found that Holstein-Jersey cross bred cows are at higher risk (94.54%) for mastitis than local zebu cows (31.25%) in India. These differences between breeds may be due to immune response to intramammary infection differs between breeds.

Species: The most of studies had been reported that cows are more vulnerable to mastitis than buffaloes (Hussain *et al.*, 1984; Sharma, 2003). The comparative high resistance of buffaloes to intramammary infections is may be possibly owing to following reasons- teat orifice is tightly closed due to presence of well developed circular muscles, stratified squamous keratinized epithelium of streak canal lining is thicker which provides an extra resistance against penetration of pathogens through epithelium and the stratum granulosum contained higher amount of keratohyalin granules in buffaloes than in cows. These keratohyaline granules may probably contribute in formation of large amount of keratin in lumen of streak canal.

Age and parity: Incidence of infected quarters increases with the age (Kumar and Sharma, 2002; Sharma and Prasad, 2002; Sharma *et al.*, 2007; Sharma and Maiti, 2010). Sharma *et al.* (2007) conducted a study on 500 lactating buffaloes of different age, parities and stage of lactation belonging to different organized or un-organized dairy farms in, Chhattisgarh State, India and found that the higher prevalence of Sub-Clinical Mastitis (SCM) in buffaloes was recorded in 5 to

9 years old animals and in 3rd and 4th parities. Older cows (>10 years) are at more risk (44.6%), particularly for sub-clinical mastitis (38.6%), than younger cows (23.6%) in which clinical mastitis was predominant (Biffa *et al*., 2005).

Stage of lactation: The incidence of mastitis is higher during just after parturition (first 2 months of lactation) and first 2-3 weeks of dry period and Corbett (2009) suggests that the highest number of clinical mastitis cases occurs during the first week of lactation and that the lactating cow is more likely to develop clinical mastitis during the first three months of lactation than the remainder of the lactating period. Fadlelmula *et al*. (2009) reported that the first month of lactation displayed the highest incidence of mastitis (62.7%), while the late stage of lactation showed the lowest incidence (11.2%). Sharma *et al*. (2011a) reported that dairy cows seemed to have more oxidative stress and low antioxidant defense during early lactation or just after parturition than advanced pregnant cows and this appears to be the reason for their increased susceptibility to production diseases (e.g., mastitis, metritis, retention of fetal membranes etc.) and other health problems.

Dry period: Dry period, a short term of approximately two months, takes place during involution of old tissue and remodeling of new tissue in mammary gland. A study suggests that a longer dry period (>40 days) increased the risk of clinical mastitis (Peeler *et al*., 2000). The rate of new infection is not constant across the dry period but is elevated during the two weeks following drying off and the two weeks prior to calving (Smith *et al*., 1985). Some studies said that the IMI rate is 2 to 12 times higher during the dry period than at any other time during the lactation cycle of the cow (Vliestra, 2003). Increased susceptibility is due, in part, to changes in the teat canal, the primary defense barrier against bacterial penetration.

Transition period: The transition period between late pregnancy and early lactation (also called the periparturient period) certainly is the most interesting stage of the lactation cycle of dairy animals. Although, the length of time classified as the transition period has been defined earlier by different authors but recently defined this period as last 4 weeks before parturition to first 4 weeks after parturition (Sharma *et al*., 2011b). Dairy cattle are more susceptible to a variety of metabolic and infectious diseases like mastitis, during the transition period compared with peak lactation (Sordillo *et al*., 2007). Dairy cows and buffaloes are more susceptible during this period because of compromised host defense mechanisms which may be directly owing to numerous physiological and environmental factors during the transition period. For example, physiological stresses associated with rapid differentiation of secretory parenchyma, intense mammary gland growth and the onset of copious milk synthesis and secretion are accompanied by a high energy demand and an increased oxygen requirement (Gitto *et al*., 2002). This increased oxygen demand enhances the production of oxygen-derived reactants, collectively termed Reactive Oxygen Species (ROS) which damage the cell membrane of phagocytic cells and compromise the immune system (Sharma *et al*., 2011b).

Milking interval: The influence of an irregular interval between morning and evening milking (<12 or >12 hours/day) on the prevalence of mastitis may have been the consequence of an enhanced chance for bacteria to colonize teat ends and streak canals during the longer milking intervals (Doherr *et al*., 2007).

Udder defense and milk factors: Udder has physical and anatomical barriers to prevent entry of infections organism in the teat canal. These barriers include teat skin, teat sphincter, keratin, furstenburg rosette etc. In addition, the teat canal produces antimicrobial substances, to act against any bacteria that enter the duct. Humoral and cellular factors inhibiting bacterial growth are in normal milk and in greater concentration in mastitic milk (Jain, 1979). Cell-free normal milk possesses a variable degree of growth inhibitory activity for Streptococci, Staphylococci and Coliform organisms (Schalm *et al.*, 1971). Milk contains various soluble factors (e.g., lactoferrin, immunoglobulins, tranferrin etc.) those play important role to prevent bacterial multiplication and establishment of infection. Lactoferrin is bacteriostatic *in vitro* for a variety of bacteria because of its iron-chelating ability which makes iron non-available for bacterial growth. Normal milk opsonizes bacteria for easy phagocytosis and mastitic milk is superior in this respect because of its content of serum opsonins. The main function of immunoglobulins in secretions of the bovine mammary gland is opsonization of microorganisms for phagocytosis by leukocytes but they also play a role as antitoxin (Nickerson, 1989). Immunoglobulins, mainly isotype IgG1, is selectively transferred into mammary secretions from blood; thus, it is the major isotype in mammary secretions for all stages of lactation.

Teat injuries: Teat canal is the main route of entry of mastitis causing organisms except tuberculous mastitis (hematogenous route), hence, teat injury is most important risk factor of intramammary infection. Changes to teat tissue, particularly the skin of the barrel, teat-end and teat canal may favor penetration of bacteria into the udder and increase the risk of new mastitis infections (Hamann *et al.*, 1994).

Blood groups: A recent study was conducted on association of the M blood group system with bovine mastitis by Larsen *et al.* (1985) in Denmark. Associations of the 11 bovine blood group systems with mastitis were examined in Red Danish dairy cattle. A significant effect of the M blood group system on mastitis incidence was observed in the first and second lactation periods and a lower frequency of mastitis is found among animals lacking the “M” factor as compared to those having the M blood group factor. Very less reports are available on this aspect of bovine mastitis.

Pathogenic risk factors: Important pathogenic risk factors include presence of number of organisms on teat skin and their virulence factors, presence of minor pathogens and blind treatment. The incidence of mastitis seems to be related to the number of organisms on the teat skin and teat end (McDonald, 1977). Streptococci and Staphylococci are in high numbers on teat skin; hence, they are the cause of most intramammary infections. The various reports indicated that the quarters that harbor minor pathogens are less susceptible to new infections by major pathogens than uninfected quarters, a phenomenon that is possibly related to the protective effect of the cell response triggered by the minor pathogens. Several studies have reported that infection by the minor pathogens *Corynebacterium bovis* and the coagulase negative Staphylococci can prevent subsequent infection with the major pathogens (Doane *et al.*, 1987). Awareness of protective tissues of the teat end becomes very important when administering intramammary therapy at drying off. Bacteria often colonize teat duct keratin, remaining there for months and may require some form of mechanical assistance to penetrate into cisternal areas. The blind intramammary treatment like full insertion of the teat canula could force keratin against the interior teat wall, creating a larger than normal opening and stretching the sphincter muscle, thereby enhancing bacterial penetration, resulting mastitis.

Environmental factors: There are various environmental factors those play crucial role in the occurrence of mastitis and development of new cases particularly environmental mastitis. These factors include housing system, climate, season, heat stress, milking hygiene, udder hygiene, milking machine, milking techniques etc. Housing has been previously identified as a risk factor for clinical mastitis and is thought to be related to an increase in exposure to environmental pathogens (Barkema *et al.*, 1999). Poorly designed facilities can contribute to increased incidence of environmental mastitis. Heat and humidity may increase the pathogen load in the environment (field or housing) (Godden *et al.*, 2003), resulting in a greater incidence of mastitis in warm weather. Shathele (2009) reported that, the incidence of mastitis decreased with increasing ambient temperature but increased with decreasing ambient temperature. Palanivel *et al.* (2008) reported that occurrence of mastitis was highest during summer season (53.80%) followed by winter (41.30%) and rainy season (37.50%) in Chennai, Tamilnadu, India. The occurrence of mastitis in different seasons depends on the agro-climatic conditions of particular region. As in Asia, different countries have different agro-climatic conditions. The results of Dhakal *et al.* (2007) showed that 37.3% of buffaloes had clinical mastitis during the summer season followed by the autumn season (31.7%) and minimum (7.83%) during spring season (February, March and April) in Nepal. Milking hygiene reduce the pathogenic organisms from inhabiting the immediate environment or skin of the animals and minimizing their spread during milking process. Udder hygiene significantly associated with the risk of environmental pathogen intramammary infection in cows (Compton *et al.*, 2007). The risk of new IMI by contagious as well as environmental pathogens such as *Streptococcus uberis* is increased by machine-induced changes such as greater degree of openness of the teat canal orifice after milking (Mein *et al.*, 2001). *Corynebacterium bovis* is a commensal of the teat canal. The milking procedure is one of the most important risk factors for both clinical mastitis and high SCC (Barkema *et al.*, 1999).

ETIOLOGY

Mastitis is a multietiological complex disease. The cow udder is an ideal environment for microbial growth and under optimum udder conditions, such as temperature, nutrition and freedom from outside influence, pathogenic organisms multiply astronomically and it is this factor that causes udder damage and triggers the response that is recognized as mastitis (Sharma and Vohra, 2011). Previously, the mastitis researchers associated the mastitis with physical factors like cold and mechanical injuries. Frank in the year 1876, transmitted the disease by inoculating inflammatory secretions from diseased quarters into the canals of healthy quarters. He thus, proved the infectious nature of this disease and put forward an entirely new concept in the investigation of mastitis etiology. An association between mastitis and pathogenic micro-organisms was established in 1887. Most major pathogens were identified by the 1940s (Sharma *et al.*, 2011b). More than 200 infectious causes of bovine mastitis are known to date and in large animals the commonest pathogens are *Staphylococcus aureus*, *Streptococcus agalactiae*, other *Streptococcus* and Coliforms in Asia (Kader *et al.*, 2002; Sudhan *et al.*, 2005; Chahar *et al.*, 2008; Yong *et al.*, 2009; Sharma, 2008; Sharma and Maiti, 2010). While in a recent report of Kumar *et al.* (2009) *Streptococcus dysgalactiae* was major (50.00%) organism isolated from the cases of subclinical mastitis in cows followed by *Staphylococcus aureus* and others. It may also be associated with many other organisms including *Actinomyces pyogenes*, *Pseudaomonas aeruginosa*, *Nocardia asteroides*, *Clostridium perfringens* and others like *Mycobacterium*, *Mycoplasma*, *Pastuerella* and *Prototheca* species and *yeasts* (Table 1). The majority of the cases are caused by only a few common bacterial

Table 1: Common mastitis causing microorganisms in the world including Asian countries

Bacteria	*Staphylococcus aureus, Streptococcus agalactiae, Escherichia coli, Streptococcus dysgalaciae, Streptococcus uberis, Streptococcus zooepidermidis, Streptococcus faecalis, Streptococcus pyogenes, Corynebacterium pyogenes, Pseudomonas aeruginosa, Mycoplasma bovis, M. Canadensis, M. californicum, M. bovihirnis, M. dispar, M. bovigenitalium, M. alkalescens, Campylobacter jejuni, Haemophilus somnus, Streptococcus pneumoniae, Corynebacterium ulcerans, Klebsiella pneumoniae, K. oxytocia, Enterobacter aerogenes, Mycobacterium bovis, M. tuberculosis, M. lacticola, M. fortuitum, Bacillus cereus, Pasteurella multocida, P. Haemolytica, Bacteroides funduliformis, Serratia marceseens, Acholeplasma laidlawii, Yersenia pseudotuberculosis, Mannheimia haemolytica, Mannheimia granulomatis.*
Facultative bacteria	*Peptococcus indolicus, Bacteroides melaninogenius, Eubacterium combesii, Clostridium sporogene, C. perfringens* type A and *Fusobacterium necrophorum, Citrobacter, Proteus*
Fungi / Yeast	*Aspergillus fumigatus, A. nidulans, Trichosporon cutaneum, Trichosporon beigelii, Pichia spp., Geotrichum candidum, Nocardia asteroids, N. brasiliensis, N. farcinica, N. neocaledoniensis, Candida krusei, C. tropicalis, C. paratropicalis, C. quillermondii, C. rugosa, Cryptococcus neoformans, Saccharomyces spp., Torulopsis* etc.
Algae	*Protothecal zopfii, P. trispora, P. wickerhamii, P. blaschkeae* etc.
Viruses	Adeno virus, Herpes virus, Rota virus, Reo virus, Mammilitis virus, Pseudocowpox virus, Parainfluenza virus, Apthovirus, R.P. virus, Bovine immunodeficiency virus etc.

pathogens, namely, *Staphylococcus* species, *Streptococcus* species, Coliforms and *Actinomyces pyogenes* (Sharma, 2010). Recently a new species of bacteria (*Mannheimia granulomatis*) had been isolated from the milk of subclinical bovine mastitis from Israel (Blum *et al*., 2010).

Fortunately, the vast majority of mastitis cases are caused by a relatively small number of microorganisms that can be grouped into three categories: (1) Contagious (*Staphylococcus aureus, Streptococcus agalactiae*, *Corynebacterium bovis, Mycoplasma* species), (2) Environmental (*Escherichia coli, Klebsiella pneumonia, Klebsiella oxytoca, Serratia* species, *Citrobacter* species, *Enterobacter aerogenes, Streptococcus uberis, Streptococcus bovis and Streptococcus dysgalactiae)* and (3) other Coagulase-Negative Staphylococci (CNS), *Serratia* spp., *Pseudomonas aeruginosa, Nocardia asteroids, Prototheca* spp., *Candida* spp., *Arcanobacterium pyogenes*) (Sudhan and Sharma, 2010).

Many studies from Asia countries have been reported that *Staphylococcus aureus* is the chief etiological agent of mastitis in cattle and buffaloes (Kang-Hee *et al*., 2001; Sharma *et al*., 2007; Abdel-Rady and Sayed, 2009; Rahman *et al*., 2010; Sharma and Maiti, 2010). *Staph. aureus* is ubiquitous and can colonize the skin as well as the udder. It is capable of causing peracute, acute, subacute, chronic, gangrenous and subclinical types of mastitis. The acute form of the disease usually occurs shortly after parturition and tends to produce gangrene of the affected quarters with high mortality. Grossly, the affected tissues are swollen, tense, hot, firm and painful. Milk secretion is reduced. Gangrenous tissues become blue and eventually black in color. *Streptococcus agalactiae* was a major cause of chronic mastitis in pre-antibiotic era and is still a serious cause of chronic mastitis in some herds, although it can be eradicated readily by proper antibiotic therapy and management. *S. agalactiae* multiplies in the milk and on the mammary epithelial surfaces, generally causing a subacute or chronic inflammatory reaction with periodic acute flareups. The affected tissue eventually is destroyed resulting in reduced milk production or agalactia.

CNS have traditionally been considered to be minor mastitis pathogens, especially in comparison with major pathogens such as *Staphylococcus aureus*, streptococci and coliforms. The main reason for this is that mastitis caused by CNS is very mild and usually remains subclinical (Taponen *et al*., 2006). The significance of CNS, however, needs to be reconsidered as in many countries they have become the most common mastitis-causing agents (Pittkala *et al*., 2004; Tenhagen *et al*., 2006). There is no doubt that some CNS species should be considered as mastitis pathogens but the large number of species included in the CNS group blurs our current understanding of their role in

Table 2: Distribution of common organisms in different countries of the Asia

	Prevalence (%) of common mastitis pathogens					
Country	*S. aureus*	CNS	*Strep.* Sp.	*E. coli*	Others	Reference
India	74.71	-	21.13	-	4.15	Singh *et al.* (1982)
	42.10	-	5.26	18.95	3.16[1], 4.74[2], 5.26[3], 2.10[7]	Misra *et al.* (1993)
	60.32	-	31.98	-	-	Shukla *et al.* (1998)
	74.04#	-	6.00	-	7.32[4], 2.93[5], 5.71[8],	Patel *et al.* (2000)
	39.01, 52.48*	-	-	-	8.51[6]	Ghose and Sharda (2003)
	18.99#	-	15.50	-	17.05[9]	Jha *et al.* (2004)
	27.90,	16.28	6.98[a]	17.44	5.81[1], 4.65[11], 5.81[12], 4.56[4], 3.49[2], 3.49[13], 3.49[7]	Das and Joseph (2005)
	-	-	15.45	12.73	10[5]	Sharma *et al.* (2007)
	34.38	25	9.38	21.87	6.25[2], 3.12[3]	Yathiraj *et al.* (2007)
	39.53#	-	20.93	9.30	16.27[6], 6.97[1], 6.97[2]	Vishwakarma (2008)
	16.66	40.47	33.33		9.52[6]	Ahire *et al.* (2008)
	59.37, 4.90*		16[a], 10.63[c], 1.1[b]	2.9	2.2[1], 1.8[4], 1.1[11], 1.1[12], 1.1[2]	Sahoo *et al.* (2009)
	27.86	72.13	-	-	-	Dutta (2009)
	27.27, 15.91*	-	50[b], 4.55[a]	-	2.27[12]	Kumar *et al.* (2009)
	57.27#	-	15.45	12.73	10[5]	Sharma and Maiti (2010)
	27.37	12.63	5.79	8.95	7.89[2], 1.35[14]	Ranjan et al. (2011)
	33.83	97.80	-	-	-	Krithiga *et al.* (2011)
Pakistan	12.06	-	7[d], 3[a]	10	3.5[5], 3[4]	Javed and Siddique (1999)
	33.99	-	-	27.09	35.46[12], 1.97[2], 1.48[5]	Rashid (2001)
	13.42#	-	9.39	46.98	14.77[2], 4.02[3], 2.01[15], 1.34[16]	Iqbal *et al.* (2004)
	45	-	23[a]	18	14[4]	Khan and Mohammad (2005)
	49.53, 6.54*	0.93	23.83[a], 8.88[e], 0.93[b]	1.4	3.74[4], 0.93[5], 0.47[14], 0.47[17]	Ali *et al.* (2008)
	28.32#	-	7.51	16.18	13.29[2], 12.42[4], 7.22[16], 6.64[5], 5.20[1], 3.17[18]	Ali *et al.* (2011)
South Korea	51.40, 2.7*	-	15.8[b], 2.1[a], 1[d]	16.50[ab]	5.1[4], 0.3[2], 0.3[15]	Kang-Hee *et al.* (2001)
	27.40	25.04	-	-	-	Moon *et al.* (2007)
	12.2	40.70	5.3[d],	4.5, 19.5[ab]	-	Nam *et al.* (2010)
Bangladesh	49.30#		14	6	8[5], 4.7[4],	Mahbub-E-Elahi *et al.* (1996)
	39.64#		2.47	11.11	3.7[4],	Kader *et al.* (2002)
	31	-	3.1	11.3	4.7[4]	Rahman *et al.*, 2010
China	23.81#	-	39.36	-	25.39[9]	Zhongwen *et al.* (2002)
	78.98	-	-	-	-	Liu *et al.* (2005)
	79.12, 6.59[$], 1.1*	-	2.2[b]	-	1.1[4], 1.1[1]	Liu *et al.* (2006)
	22.61#	-	25.22	-	15.65[18], 16.52[14], 6.96[5]	Wang and Niu (2009)
	41	-	53[d], 29[b], 27[a]	82	-	Cheng *et al.* (2010)
	29.5	4.7-19.7	7.6[a], 4.7[d], 2.9[b]	25.70	16.2[5]	Yang *et al.* (2011)
Iran	2.89	-	22.11[a], 11.43[b],	10.16	1.07[5], 1.76[4], 0.14[1], 0.21[2], 0.03[15], 0.03[19]	Atyabi *et al.* (2002)

Table 2: Contiuned

	Prevalence (%) of common mastitis pathogens					
Country	*S. aureus*	CNS	*Strep.* Sp.	*E. coli*	Others	Reference
	-	3.88	8.33	9.44	-	Ebrahimi *et al.* (2007)
	63.30, 12[#]	-	9.8, 0.4[a]	7.9[ab]	5.8[6],	Kheirabadi *et al.* (2008)
	14	36.18	-	-	8[5], 7[4]	Beheshti *et al.* (2010)
Israel						
	62.50	-	21.88[a]	12.50	-	Jemeljanovs *et al.* (2002)
Japan	-	-	-	75	13[1], 8[2], 4[18]	Nakajima *et al.* (1997)
Nepal	15.38, 57.69*	-	17.31[f], 7.69[b]	1.92	-	Dhakal (2006)

[#] *Staphylococci*, * *Staphylococcus epidermidis*, [$] *Staphylococcus hemolyticus* [a] *S. agalactiae*, [b] *S. dysgalactiae*, [c] *S. albus*, [d] *S. uberis*, [e] *S. hyicus*, [f] *S. bovis* [ab] Coliforms. [1] Others *Klebsiella* spp., [2] *Pseudomonas* spp., [3] Proteus spp., [4] Bacillus spp., [5] *Corynebacterium* spp., [6] *Micrococcus* spp., [7] *Candida* spp., [8] Gram-negative, [9] Gram positive Bacilli, [11] *C. bovis*, [12] *C. pyogenes*, [13] *Trichosporon* spp., [14] Yeast, [15] *Pasteurella* spp., [16] *Salmonella* spp., [17] *Prototheca*, [18] *Enterobacter*, [19] *Mycoplasma*

mastitis. Despite intensive aetiological research, still around 20-35% of clinical cases of bovine mastitis have an unknown etiology.

For understanding, treatment and effective control strategies it is important to know the distribution of etiological agent in that particular region, so here we summarizing the distribution of some important bovine mastitis causing organisms (Table 2).

Prevalence of major pathogens is decreasing, the relative importance of other organisms, particularly CNS, increasing. In India, the prevalence of CNS among bacterial isolates from milk samples increased from 9.91% in 2003 (Sharma and Prasad, 2003) to 72.13% in 2009 (Dutta, 2009). CNS are now among the most commonly isolated organisms from milk samples of cows with subclinical mastitis in many countries (Rajala-Schultz *et al.*, 2004; Osteras *et al.*, 2006; Ahire *et al.*, 2008).

Generally, the mastitis due to fungi and yeast is uncommon or rare. Kirk and Bartlett (1986) have been mentioned a prevalence of fungal mastitis of 2 to 7%. The prevalence of mycotic mastitis is usually very low (1-12% of all mastitis causes) but sometimes it can occur in enzootic proportions. The percentage of fungal isolation in surveys carried out in many countries varies considerably, with 6.1% rates reported in Egypt (Awad *et al.*, 1980), 1.3% in South Korea (Yeo and Choi, 1982) and 12.07% (Costa *et al.*, 1993) to 25.4% (De Casia dos Santos and Marin, 2005) in Brazil. In India, bacteria are the major mastitis etiological agents, prevalence of Mycotic mastitis is less but upto 60% of cases of Mycotic mastitis has been reported (Vimalraj *et al.*, 2006). Isolation of yeast like fungi from bovine milk was reported as early as 1901 (Beck, 1957). In Fungi, *Candida* spp., *Aspergillus* spp., *Trichosporon* spp. and *Saccharomyces* spp. are comparatively more prevalent but the involvement of *Geotrichum candidum* in bovine mastitis is very rare (Chahota *et al.*, 2001). *G. candidum* is an opportunistic, keratinophilic yeast like fungus that is widely distributed in nature i.e., soil, fodder etc.

Kheirabadi *et al.* (2008) from our studies concluded that subclinical mastitis is mainly caused by *Staphylococcus aureus*, in west central region of Iran. Ebrahimi and Nokookhan (2005) studied on fungal isolation from mastitis milk and found *Fusarium* (28.26%) were most frequent one in chronic cases and followed by *Trichosporon* (26.08%), *Candida* spp. (17.39%), *Aspergillus fumigatus* (13.04%), *Rhodotorulla* (10.87%) and *Geotrichum* (4.35%) while in cases of acute mastitis the most frequent ones were *Trichosporon* (66.04%) and followed by *Candida* spp.

(11.20%), *Geotrichum* (5.66%), *Aspergillus fumigatus* (3.77%), *Fusarium* (1.89%) and *Rhodotorulla* (1.89%) from Iran. Recent report from Iran indicating that CNS are major etiological agents for bovine mastitis in Iran (Beheshti *et al*., 2010). In West central Iran, *E. coli* and CNS are more frequent cause of bovine mastitis (Ebrahimi *et al*., 2007).

Jemeljanovs *et al*. (2008) analysed a total of 577 subclinical and clinical mastitis secretion samples from the different dairy farms in Lativa and found that *Staphylococcus aureus* and CNS prevail in subclinically (72.7%) and clinically (46.7%) diseased cow's udder secretion samples. *Staphylococcus aureus* were isolated from 41.1% of subclinically and 24.1% clinically diseased cow's secretion samples, but CNS from 27.0 and 20.4%, respectively. *Streptococcus* spp. were isolated from 5.9% of subclinically and 33.6% of clinically mastitic cows, while in the 1980s *Streptococcus* spp. were isolated from 65.5% of subclinical and 64.8% of clinical mastitis cases. Therefore, present findings revealed that replacement of predominating agents of mastitis from the genus *Streptococcus* to the genus *Staphylococcus* has been occurred in Lativa.

From the above studies in Nepal, it is concluded that pattern of bacterial distribution responsible for mastitis has been changed i.e., previously (1994-1998) *E. coli* was predominating organism, while recent reports showed that *Staphylococcus* spp. are more prevalent than *E. coli* in mastitis case (Dhakal, 2006; Dhakal *et al*., 2007).

STATUS OF MASTITIS DISTRIBUTION

The prevalence of mastitis is increasing in parallel with the development of new high milk producing breeds of cows and buffaloes. Some other factors may also be contributed in the increasing incidence of mastitis like lack of awareness, delay in the detection in absence of visible signs of abnormal milk, unhygienic milking practices and, delay and incomplete treatment of clinical and chronic mastitis. Figure 3 is clearly showing the increasing trend of bovine mastitis in Asia.

Studies conducted in different states of India reflecting the high prevalence of bovine mastitis all over India for the past seven decades when the first record of the mastitis was made by Land

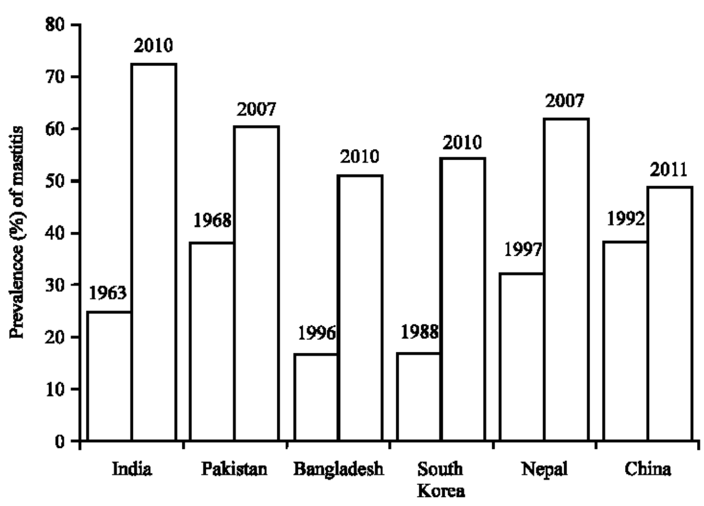

Fig. 3: The increasing trend of bovine mastitis prevalence

Table 3: Distribution of mastitis in different geographical regions of Asian countries.

Country	Species	Prevalence (%) of mastitis	Remarks	References
India	Cow	24.80		Anon (1963)
	Buffalo	21.20	-	
	Cow	29.34[#]		
	Buffalo	20.89[#]	FQ- 42.63%, HQ- 57.42%	Sharma and Rai (1977)
	Cow	23.08[1#]	-	Pal *et al.* (1989)
	Cow	20[#], 7.94*	FQ- 52.07%, HQ- 47.91%	Sharma *et al.* (1993)
	Cow	48.70[#]		
	Buffalo	11.32[#]	-	Bansal *et al.* (1995)
	Cow	74.56*, 74.10[#]	-	Nauriyal (1996)
	Buffalo	36.36[#2], 22.73[#1]	-	Sharma and Kapoor (2000)
	Cow	41.77*		
	Buffalo	50.00*	Winter- 51.47%; rainy- 30.88% and in summer- 17.64%	Bhikane *et al.* (2002)
	Buffalo	30.68*	-	Singh *et al.* (2003)
	Buffalo	70.32[2], 43.53[1]	-	Sharma *et al.* (2004)
	Cow	52.57	LF- 30.30 and RH- 38.46%	Samanta *et al.* (2006)
	Buffalo	68.60[2], 42[1]	-	Sharma *et al.* (2007)
	Cow	32.40[#]		
	Buffalo	22.22[#]	-	Munda *et al.* (2007)
	Buffalo	41.07[#], 7.90*	-	Vishwakarma (2008)
	Cow	58.76	-	Verma and Nauriyal (2009)
	Cow	60.00	-	Sharma *et al.* (2010)
	Cow	83.00	-	Kumar *et al.* 2010
	Cow	72.63	-	Ranjan *et al.* (2010)
	Cow	70.00[2], 55.25[1]	-	Sharma and Maiti (2010)
Pakistan	Buffalo	38.07[#]	-	Said and Abd-el-Malik (1968)
	Buffalo	44.90	-	Hashmi and Muneer (1981)
	Buffalo	47.50	-	Anwar and Chaudhry (1983)
	Buffalo	64[1], 30.50[2]	-	Rehman (1995)
	Buffalo	51.66-66.66[1], 76.00-82.61[2]	-	Bachaya *et al.* (2005)
	Cow	36		
	Buffalo	27	-	Khan and Muhammad (2005)
	Cow	47.58		
	Buffalo	60.27	-	Chrishty *et al.* (2007)
	Cow	27-48[#1], 30.40-60.00[#2]	-	Bachaya *et al.* (2011)
	Buffalo	44[#]	-	Ali *et al.* (2011)
Bangladesh	Cow	16.5[#]	-	Prodhan *et al.* (1996)
	Cow	21.20*	-	Nooruddin *et al.* (1997)
	Cow	46.60	-	Kader *et al.* (2002)
	Cow	44.80	Prevalence was high in wet season	Rahman *et al.* (2009)
	Cow	51.30[#]	-	Rahman *et al.* (2010)
South Korea	Cow	36.6[2], 16.7[1]	-	Lim *et al.* (1988)
	Cow	35.00-54.30	-	Nam *et al.* (2010)
China	Cow	16.58*[1], 38.4[#1]	Prevalence was high (40%) in summer season	Fang *et al.* (1993)
	Cow	36.5[#], 16.7*	-	Liu *et al.* (2005)

Table 3: Continued

Country	Species	Prevalence (%) of mastitis	Remarks	Reference
	Cow	$54.3^{\#2}$, $28^{\#1}$	-	Li *et al.* (2009)
	Cow	8.7^{*2}, 3.7^{*1} $48.8^{\#2}$, $19.00^{\#1}$	Blind quarter at cow and quarter level 11.5 and 3.7%, respectively	Yang *et al.* (2011)
Iran	Buffalo	$62^{\#1}$, 64^{*1}	-	Dhakal *et al.* (2007)
	Cow	48.75	-	Ghazaei (2006)
	Cow	29.03	-	Ebrahimi *et al.* (2007)
	Cow	$29.03^{\#}$	Highest prevalence of SCM was during spring months (34%) than summer (25%)	Kheirabadi *et al.* (2008)
	Buffalo	$27.36^{\#}$	-	Beheshti *et al.* (2010)

^{1} Quarter basis, ^{2} Animal basis, $^\#$ Subclinical, * Clinical, FQ = Fore quarter, HQ = Hind quarter, LF: Left fore, RH: Right hind

in 1926. The two decades ago an average incidence of clinical mastitis in India was 1 to 10% (Prabhakar, 1986; Kumar, 1990) and separately incidence of Sub-Clinical Mastitis (SCM) was 10 to 50% in cows and 5 to 20% in buffaloes (Kumar, 1988; Singh, 1991). A technical report of Indian Council of Agricultural Research 1960-1961 indicates the incidence of mastitis in 267 animals in Bangalore was 24.8 and 21.2% in cows and buffaloes, respectively and in 516 animals in Uttar Pradesh was 40.5 and 27% in cows and buffaloes, respectively (Anon, 1963). While recent studies have been reported the incidence of SCM ranged from 19.20 to 83% (Tuteja *et al.*, 1993; Sharma and Maiti, 2010; Kumar *et al.*, 2010) and 42% (Sharma *et al.*, 2007) in cows and Buffaloes, respectively. However, overall prevalence of bovine mastitis in India is 44.67% (ranged from 25.63 to 97.61%). This data is calculated as mean of more than 100 studies of 21 states of the India. This range (25.63 to 97.61%) of bovine mastitis occurrence clearly indicates the drastic increase in the prevalence of mastitis. This significant increase in the occurrence of bovine mastitis is an alarming phase for the dairy sector. The increasing trend is also in most of Asian countries like in Pakistan from 38.07 (Said and Abd-el-Malik, 1968) to 60.27% (Chrishty *et al.*, 2007), in Bangladesh from 16.50 (Prodhan *et al.*, 1996) to 51.30 (Rahman *et al.*, 2010) and so on (Fig. 3). The detail distribution of bovine mastitis in different major milk producing countries of Asia is given in Table 3.

It has been reported that sub-clinical mastitis is 3-40 times more common than the clinical mastitis and causes the greatest overall losses in most dairy herds (Schultz *et al.*, 1978). Only sub-clinical mastitis is responsible for 60-70% of total economic losses associated with all mastitic infections. It is assumed that the sub-clinical form frequently goes unnoticed and is associated with significant economic losses that include increased clinical diseases risks, impaired milk production and reduced reproductive performance and culling losses (Sharma, 2010).

The data of frequent changing trend of etiological agents and increasing prevalence rate of mastitis in cows and buffaloes in Asia is a matter of broad discussion and need more strategic research in this field to control the mastitis. Because as we know that the Asian countries are the major milk producing countries in the world. This review paper is providing the all basic data in different parts of Asia for policy makers and mastitis researchers to work in the direction of control of mastitis. Moreover, bovine mastitis is still a big challenge to the field veterinarians and mastitis researchers to fight against mastitis.

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