Chapter I

INTRODUCTION
“Mastitis” describes an inflammatory reaction in the mammary gland. The term comes from the Greek derived word elements masto- referring to the mammary gland and -its meaning- “inflammation”. Mastitis could be technically used to describe udder injury that may result in inflammatory reaction, it is generally accepted that the causative agents for inflammations are microorganisms that gained entry into the teat canal and mammary tissue (Eckersall et al., 2001). Joshi and Gokhale (2006) reported prevalence of mastitis in Indian cattle and buffaloes, which shows significant decrease in the production of milk, as well as alters its composition. The microorganisms invade the mammary gland, multiply there and produce harmful substances that result in inflammatory responses. Mastitis is difficult to control as several different species of microorganisms have the ability to infect the udder. Inflammation is characterized by swelling, heat, redness and pain. Persisting inflammation leads to tissue damage and replacement of secretory tissues of udder with non productive connective tissues ultimately leading to change in the milk quality.

There are several ways of classifying mastitis. A simple classification recognizes mastitis as two major groups:

A- **Contagious Mastitis:** Caused by bacteria live on the skin of the teat and inside the udder. Contagious mastitis can be transmitted from one cow to another during milking.

B- **Environmental mastitis:** Describes mastitis 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. Cases of environmental mastitis rarely exceed 10% of the total mastitis cases in the herd.

Contagious mastitis can be divided into three groups:

1- Clinical mastitis
2- Sub-clinical mastitis
3- Chronic mastitis

**Clinical Mastitis:** Visible signs of mastitis which include-

- Mild signs flakes or clots in the milk, may have slight swelling of infected quarter.
• Severe signs secretion abnormal, hot, swollen quarter or udder; cow may have a fever, rapid pulse, loss of appetite, dehydration and depression; death may occur.

**Subclinical Mastitis:** No visible signs of the disease-

• Somatic cell count (SCC) of the milk will be elevated.
• Bacteriological culturing of milk will detect bacteria in the milk.
• Causes the greatest financial loss to dairy farmers through lowered milk production.
• For every clinical case of mastitis, there will be 15 to 40 sub-clinical cases.

**Chronic mastitis:** An inflammatory process that exists for months, and may continue from one lactation to another. Chronic mastitis for the most part exist as sub-clinical but may exhibit periodical flare-ups sub-acute or acute form, which last for a short period of time.

Mastitis is regarded as the costliest disease of dairy industry worldwide; it is associated with economic losses of $35 billion annually (Annapoorani et al., 2007). Of the two forms of mastitis, subclinical mastitis is subtle, causes huge economic losses, and is difficult to detect as the cow appears healthy, the udder does not show any signs of inflammation and the milk appears normal. However, microorganisms and somatic cells are found in elevated numbers in the milk. The cows that have subclinical mastitis serve as reservoir of organisms and may result in infection to healthy cows. Most clinical cases start as subclinical and therefore, control of subclinical mastitis is extremely important to check it’s development to clinical cases and curtailing heavy economic losses. A variety of pathogens can establish chronic infection that only occasionally manifests clinical signs of mastitis. The primary focus of most subclinical mastitis programmes is to reduce the prevalence of contagious pathogens viz. *Staphylococcus aureus, Streptococcus agalactiae* and other Gram-positive cocci, most notably *Streptococcus dysgalactiae* (which may be contagious or be environmentally acquired) and environmental pathogens including *Escherichia coli, Klebsiella pneumoniae, Streptococcus uberis, Enterococcus* and numerous other coagulase-negative *Staphylococci*, including *Staphylococcus hyicus, Staphylococcus epidermidis, Staphylococcus xylosus* and *Staphylococcus intermedius*. This list is not complete and organisms that cannot be cultured by standard culture conditions may escape
notification. It is widely accepted that up to 99% of the microbes in the environment cannot be readily cultivated (Sekiguchi, 2006). To overcome these difficulties and limitations associated with cultivation techniques, different DNA-based molecular methods have been developed for characterizing microbial species and assemblages, and these have significantly influenced our understanding of microbial diversity and ecology (Delong, 2005). Though much of metagenomics focuses on bacteria (especially the 16S rRNA gene), the field is expanding rapidly to encompass the entire spectrum of organisms in an environmental sample that includes bacteria, archaea, viruses, small eukaryotes, plasmids, and short RNAs (William, 2009). In general, methods based on 16S rRNA gene analysis provide extensive information about the taxa and species present in an environment, however, this data usually provides only little, if any, information about the functional role of different microbes within the community and the genetic information they contain about microbial niches (Streit and Schmitz, 2004). Thus, Metagenomics is a rapidly growing field of research that aims to study uncultured organisms to understand the true diversity of microbes, their functions, cooperation and evolution in environments such as soil, water, ancient remains of animals or the digestive system of animals and humans (Ghazanfar and Azim, 2009), and therefore is capable of overcoming these difficulties.

The ongoing revolution in metagenomic sequencing technology has led to the production of sequencing machines with dramatically lower costs and higher throughput. Next-generation platforms are helping to open entirely new areas of biological inquiry, including investigation of ancient genomes, characterization of ecological diversity and identification of unknown etiologic agents.

In recent years, the role of cytokines in the patho-physiology of bovine mastitis has been the subject of many studies. Cytokines are immunoregulatory mediators that play a central role in the regulation of immune responses against different infections. Several studies have indicated that variations in cytokine expression are associated with disease activity in immune-mediated or inflammatory disorders (Hansen et al., 2004). Understanding the regulation of the immune response during infections of the mammary gland is important for designing prophylactic vaccines and for optimization of therapeutic protocols. Certain aspects of immune mechanisms in cattle and their
regulation by cytokines and growth factors have already been addressed (Sordillo et al., 1997). However, the interplay of immune compartments involved in the generation of protective immunity against specific bacterial infections is yet to be fully understood.

The conventional antimicrobial agents have been the mainstay of mastitis therapy over the last many decades and have potential high cure rate when the treatment is well targeted. However, use of antibiotics is associated with cost, the possibility of development of acquired drug resistance, drug residues in the milk, and disruption of symbiotic gut flora of the host when systemic administration is used. Given the seriousness of such problems, researchers and clinicians have been trying to find effective therapeutic agents using alternative medicine. Traditional healthcare of animals (ethno-veterinary medicine) includes the use of medicinal plants/herbs, surgical techniques and management practices to prevent and treat a range of diseases and problems encountered by livestock keepers. Research has shown that many of the plants used to prepare indigenous medicines do contain valuable active ingredients, however, much research remains to be done in this area. Modulation of cytokine secretion may offer novel approaches in the treatment of a variety of diseases. One strategy in the modulation of cytokine expression may be through the use of herbal medicines. A class of herbal medicines, known as immunomodulators, alters the activity of immune function through the dynamic regulation of inflammatory molecules such as cytokines.