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#### Chapter

# Epidemiology of Bovine Mastitis and its Diagnosis, Prevention, and Control

S.D. Audarya, D. Chhabra, R. Sharda, R. Gangil, R. Sikrodia, J. Jogi and N. Shrivastava

#### **Abstract**

Mastitis is an inflammation of mammary glands that is prevalent in dairy bovines. It causes a significant proportion of economic losses to the dairy farmers in India. Cattle and buffalo farming contribute significantly to the economy of the state. Various infectious agents such as bacteria, fungi, and algae may cause mastitis. Hence, it is essential to understand the etiological agents and predisposing factors that lead to mastitis in susceptible bovine populations in Madhya Pradesh state so that appropriate prevention and control strategies can be implemented. In this chapter, epidemiology, diagnosis, prevention, and control measures of mastitis in general and in India, the state of Madhya Pradesh, in particular, will be presented.

**Keywords:** mastitis, cattle, buffalo, bovine, Madhya Pradesh, epidemiology, diagnosis, prevention, control

#### 1. Introduction

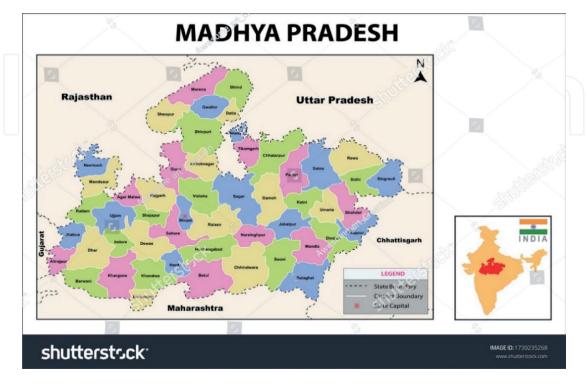
Mastitis, an inflammation of the mammary gland, is a very common disease in bovines. Among all the pathogens, bacteria are commonly implicated as the cause of mastitis [1]. Mastitis is characterized by inflammatory changes in milk and udder tissue. Mastitis is prevalent worldwide in dairy farming, causing heavy economic losses to the dairy industry. In those countries having well-developed dairy industry, morbidity of mastitis in dairy cows is 40% [2]. Affected bovines may lose their total milk production. Infectious agents like bacteria residing on the udder tissue or from the environment can enter through the teat canal. Milk from bovines with mastitis is unfit for human consumption because some mastitis-causing bacteria are zoonotic and can cause human infection [3]. Once the susceptible dairy bovine develops mastitis, it loses its milk production capacity significantly. Milk from a cow with mastitis is discarded due to inferior milk quality. Dairy animal owners have to bear extra costs for treating and maintaining such infected animals [4]. Clinically, there are two forms of mastitis: clinical form and subclinical form. The clinical form is characterized by local visible inflammatory changes in milk and udder tissue with or without systemic clinical signs, whereas subclinical form does not manifest clinical signs of mastitis but increased somatic cell counts with the presence of the causative agent. The diagnosis of the subclinical form of mastitis requires cow side test such as California mastitis test (CMT) or various laboratory tests including somatic cell

count (SCC) and milk bacteriological culture. Early diagnosis of subclinical form of mastitis is very much essential for successful treatment and control of infection [5]. Generally, high milk-producing cows are more suffered from mastitis than low milk producers [6]. This chapter highlights the epidemiology of mastitis and available diagnostic methods, prevention, and control measures with major focus on India in general and the state of Madhya Pradesh in particular.

## 1.1 The dairy sector and bovine population in the state of Madhya Pradesh, India

Madhya Pradesh is the second largest state in India with an area of 3,08,000 sq. km. The state is the part of the peninsular plateau of the country situated in the north-central part. It is one of the landlocked states in India (**Figure 1**).

It has three major seasons; summer, monsoon, and winter. The temperature of the entire state during summer (March–June) ranges above 29.4°C. Monsoon starts mid-June, and between June and September, the state gets the majority of its share of rainfall. In the winter season (November–February), the temperature remains low in the northern parts of the state compared to the southern parts. The state has over 70 million (7 crores) human population. The majority of the population of Madhya Pradesh (75%) resides in the villages, and most of them have income from agriculture. Tribal population accounts for 20% of the total population of Madhya Pradesh [7]. Livestock rearing provides them extra income and food security. Madhya Pradesh state Livestock and Poultry development corporation envision to increase the income levels of farmers involved in animal husbandry, particularly women by adopting a series of measures including a) increase in milk production, b) protect farmers from economic losses, and c) educate farmers about better management practices [8]. Madhya Pradesh state cooperative dairy federation limited has set up three-tier structures for dairy cooperatives. Primary village cooperatives (I tier), regional milk unions (II tier), and the apex federation (III tier) work in tandem for smooth functioning at field and plant operation levels and also for marketing the



**Figure 1.** *Madhya Pradesh state in India.* 

Animal _	Livestock population (in million)				Change in percent
	India (2019)		Madhya Pradesh		
	Total	Female	2012	2019	
Cattle	192.49	145.12	19.6	18.7	-4.42
Buffalo	109.85	100.57	8.2	10.3	25.88

**Table 1.**Bovine population in India and Madhya Pradesh.

branded products (Sanchi and alike). During 2018–2019, there were 517 milk routes and 4698 functional dairy cooperative societies. A total of U.S. \$ 148.71 million (Indian ₹ 1096.92 crores) has been paid to milk producers [9]. The total bovine population in 2019 increased by 1% to that of the total bovine population recorded in the previous census. According to 20th livestock census (provisional) statistics, the total bovine population in India stands at 302.79 (Cattle, Buffalo, Mithun, and Yak) million in 2019 [10]. However, in the state of Madhya Pradesh, only cattle and buffalo are reared. The cattle and buffalo population of the country and the state of Madhya Pradesh from the above-mentioned census is presented in **Table 1**.

Mastitis is an economically important infectious disease of cattle and buffalo in the state. It is essential to know its epidemiology, diagnosis, prevention, and control measures in India in general and in in the state of Madhya Pradesh in particular.

#### 1.2 Institutes working on mastitis in the state of Madhya Pradesh

Madhya Pradesh state at present is having three constituent veterinary colleges covering the western (Dr. Ambedkar Nagar-Mhow) and north-central and north-eastern region (Jabalpur and Rewa) of the state (**Figure 2**) under the auspices of Nanaji Deshmukh Veterinary Science University, Jabalpur. The laboratories there are well equipped for the diagnosis of cases of mastitis. Besides, the state also boasts of a vibrant veterinary service where disease diagnostic laboratories are the main-stay to avert any serious issues about infectious diseases.



**Figure 2.**Locations of institutes working for diagnosis and prevention and control of mastitis in the state of Madhya Pradesh (red dot—Dr. Ambedkar Nagar-Mhow, pink dot—Jabalpur, green dot—Rewa).

Work published on mastitis from the western and north-central regions from the state of Madhya Pradesh is available. However, there are very few published reports presently available from the north-eastern region of the state. Although recently a study was conducted for the assessment of subclinical mastitis in bovines in and around Rewa district [11], in this study, a 31.4% prevalence rate for subclinical mastitis in cattle was reported. It is essential to improve awareness of agriculture and livestock owners in general and dairy animal owners in particular about prevalent bovine diseases including mastitis in the state of Madhya Pradesh to minimize production losses. So, information and toolkits regarding mastitis, its diagnosis, prevention, and control measures are being distributed.

#### 2. Economic impact due to mastitis

Among the two forms of mastitis, the clinical mastitis is readily diagnosed by clinical manifestations (**Figure 3**) and affected animals can be treated but the subclinical mastitis is asymptomatic and often diagnosed late, after days or weeks of infection. In Asia, incidence rates of clinical and subclinical mastitis ranged from 1-8% and 55–60%, respectively. The dairy industry in India in general and in the state of Madhya Pradesh, in particular, contributes to the economy by uplifting people from poverty and also helps in earning a regular income. Mastitis is responsible for nearly 70% of milk loss. Economic losses due to mastitis in India were estimated to be around U.S. \$ 971.39 million (Indian ₹ 7165 crore) [12].



**Figure 3.**Cow affected with clinical form of mastitis showing visible udder swelling.

#### 3. Epidemiology of mastitis in India and Madhya Pradesh

#### 3.1 Clinical and subclinical mastitis

Clinical mastitis is diagnosed readily by visible clinical signs and changes in the milk. In one of the studies conducted in India, in buffaloes from rural and urban dairy farmers, there was 18.74% prevalence of clinical mastitis [13]. In one of the other studies, 4.77% incidence of clinical mastitis was reported in the state of Madhya Pradesh [14]. Out of 260 cases of bovines in the Jammu region of the

country, 30 cases (11.54%) were positive for clinical mastitis. Prevalence of clinical mastitis in bovines ranged from 4.77 to 18.74% [13–15].

A higher level of incidence of subclinical mastitis (47.79%) was reported in the state of Madhya Pradesh [14]. In subclinical mastitis, there are no visible changes in milk or udder appearances in the affected bovines. Incidence of subclinical mastitis ranged between 19.2 and 83% in cows of Punjab state in India [16]. Recently, 40% of the overall incidence of subclinical mastitis was recorded in buffaloes in the state of Madhya Pradesh [17]. However, in lactating cows, the overall occurrence of subclinical mastitis was 27.81% [18]. In a recent study carried out in 2020, the reported overall prevalence of subclinical mastitis in cows was 31.55%. The prevalence of subclinical mastitis was highest in 5–7 years of age group (38.50%). The prevalence percentages of subclinical mastitis in organized (scientifically reared animals with adequate floor space availability) and unorganized dairy farms (animals reared in open space by livestock owners) were 29.82 and 41.66%, respectively [19]. In one of the other studies conducted in India, in buffaloes, there was 32.9% prevalence of subclinical mastitis [13].

When compared to clinical mastitis, subclinical mastitis is 15 to 40 times more prevalent [13, 20]. The prevalence of subclinical mastitis in dairy herds varied from 5 to 75% [21].

Subclinical mastitis results in greater economic losses to the farmers rearing dairy cattle and buffalo. No visible clinical signs are noticed in subclinically affected animals. In India, about 70–80% of economic losses have been attributed to subclinical mastitis alone. It occurs worldwide and also has adverse impacts on animal health and the quality of milk produced. Besides culling chronically infected animals from herds, decreased fecundity in affected animals and cost of treatment for mastitis lead to major economic losses. Subclinically infected cattle and buffalo can be a source of infection to other susceptible populations in the herd [22].

The prevalence of subclinical mastitis in bovines in India ranged from 9.88 to 86.87% [23]. In Madhya Pradesh, the prevalence of subclinical mastitis was reported highest in Jersey Cross (86.87%) than Holstein Friesian (75%), Malvi (57.35%), Sahiwal (75%), and Gir (80%) [24]. Among the various bacterial causative agents, staphylococci and streptococci were reported to be the most common pathogens of mastitis in India [25].

#### 3.2 Etiology of mastitis

Mastitis is caused by physical, chemical, and biological agents. Generally, bacterial infections are the main causes of mastitis. Among many of the different microorganisms isolated from cases of bovine mastitis, the most common are Staphylococci (Staphylococcus aureus), Streptococci (Streptococcus agalactiae, Streptococcus dysgalactiae, Streptococcus uberis), and members of the family Enterobacteriaceae (Escherichia coli, Klebsiella pneumoniae) and other less common causative agents are Pseudomonas aeruginosa, Mycoplasma species, Mycobacterium species, Nocardia asteroides, Candida species, Cryptococcus species, and Aspergillus species. Rarely viruses are implicated in producing mastitis in bovines. The mastitis may be acute, per acute, subacute, chronic, and subclinical. Entry of the pathogen via teat canal into the mammary gland is characterized by increased leucocyte count in the milk [26].

Staphylococci, streptococci, *Escherichia coli, Pseudomonas* species, *Corynebacterium* species, *Mycoplasma* species, *Streptococcus agalactiae*, *Streptococcus dysgalactiae*, and *Mycobacterium tuberculosis* are isolated from buffalo suffering from mastitis. Among all the pathogens of bovine mastitis, staphylococci, streptococci, micrococci, *Corynebacterium* species, and *Escherichia coli* were isolated in Madhya

Pradesh. Listeria organisms were isolated from raw milk. Incidence of streptococci (32.53%), micrococci (5.74%), *Corynebacterium* (1.91%), and *Escherichia coli* (0.95%) was reported in cases of bovine mastitis. However, recently, 17.19% mastitis cases in buffalo were due to infection with *Escherichia coli* [27–31].

Depending on the mode of transmission of mastitis pathogens from their natural habitat to the mammary glands, there are contagious and environmental mastitis pathogens. Contagious mastitis pathogens exist on the udder or teat surface of infected cows. These are the primary source of infection from where they are transmitted during milking to uninfected cows. Major contagious bacterial mastitis pathogens are coagulase-positive *Staphylococcus aureus*, *Streptococcus agalactiae*, and *Mycoplasma bovis*. *Corynebacterium bovis* is the less common contagious mastitis pathogen [26]. *Staphylococcus aureus*, a bacterial species, is most commonly isolated from cases of bovine mastitis [32]. The reported prevalence of *Staphylococcus aureus* was 58.85% in Madhya Pradesh. *Staphylococcus aureus* is known to acquire antimicrobial resistance very quickly. Methicillin resistance *Staphylococcus aureus* (MRSA) is also a causative agent of mastitis in dairy cattle. Detection of MRSA has serious public health significance. In dairy cattle, the reported prevalence of MRSA mastitis was 16.47% in the Jabalpur region of Madhya Pradesh [33].

Environmental mastitis pathogens are in the environment of dairy cows and they can transfer to the mammary glands at any time. Major environmental mastitis pathogens include environmental streptococci (*Streptococcus uberis* and *Streptococcus dysgalactiae*) and coliform bacteria (*Escherichia coli*, *Klebsiella* species, *Enterobacter* species, and *Citrobacter* species). Some of them are opportunistic pathogens and infect mainly the immunocompromised host [26, 34]. Coliform bacteria (*Escherichia coli*) do not normally live on the skin or in the udder. These organisms can enter the teat canal when the animal comes in contact with a contaminated environment. Contaminated bedding materials, soil, manure, and organic matter in the environment can be a source of *Escherichia coli* that can lead to environmental mastitis. This kind of environmental mastitis was reported from several countries. *Escherichia coli* are one of the most frequently isolated bacteria from clinical infections. In severe, naturally occurring clinical cases of mastitis due to *Escherichia coli* there can be necrosis of the mammary epithelium [35].

#### 4. Mastitis risk factors

Mastitis risk factors include managemental factors and cow factors. In management, the most common measures that can be used to avoid mastitis at the farm level are regular floor cleaning, use of appropriate milking techniques, and udder washing before milking and pre- and post-milking teat dipping in antiseptic solutions. Culling is hardly practiced in the country and also in the state of Madhya Pradesh. Regular screening of bovines for mastitis is not practiced at the larger scale as it needs to be in the state. Generally, dairy owners approach to the testing facilities after the cases of mastitis in bovines are clinically visible. Cow factors include the stage of lactation, breed, history of mastitis, and parity. Even if udder defense mechanisms are there, microbial infection overpowers at times and causes mastitis. Additionally, at times due to inadequate livestock management and husbandry practices such as unhygienic maintenance of livestock, inadequate floor space available to the animals, improper ventilation, and faulty milking techniques used by milkman also contribute as predisposing factors for mastitis. Physical injury to teat skin, teat canal, and mammary cistern are also important predisposing factors for entry of microbial pathogens in the udder to cause mastitis [36].

#### 5. Diagnosis

Rapid cow side tests that are used to diagnose mastitis are highly required to implement prevention and control strategies. Some of the tests used in the diagnosis of clinical and subclinical mastitis are described below.

#### 5.1 Cow side test—strip cup test

The physical appearance of milk is checked by a test named strip cup test. Strip cup or strip plate-based visual examination is routinely used for detection of clinical mastitis in individual and herd animals. The quality of the foremilk is examined visually after squirting few stripes of milk on the strip cup for gross examination of blood, flakes, clots, wateriness.

#### 5.2 California mastitis test (CMT)

California mastitis test (CMT) is a simple rapid screening test, based on the estimation of the number of somatic cells in the milk sample. The somatic cell population consists of 75% leucocytes and 25% epithelial cells. The rise in the leucocytes indicates mastitis. The CMT reagent is mixed with the milk samples and the reagent causes lysis of somatic cells and release of DNA that form a gel. The CMT test result is qualitatively estimated (**Figure 4**). The average SCC of 2,00,000 cells/mL is considered as normal milk. For bulk tank milk >2,00,000 cells/mL of milk shows the presence of mastitis with a significant loss in milk production; for composite milk, from all the four quarters of a cow >2,00,000 cells/mL is considered mastitis whereas for milk from a single quarter of a cow >1,00,000 cells/mL is considered mastitis milk [37].

#### 5.3 Milk bacteriological culturing and identification

For isolation and identification of bacteria-causing mastitis, the milk samples for bacteriological examination are first centrifuged and the resulting sediment is streaked on ordinary, selective, or differential media and incubated aerobically. Attempts should be made to isolate mycotic and anaerobic organisms. Milk sample



**Figure 4.**California mastitis test paddle showing gel formation.

is streaked on sheep blood agar (0.05–1% aesculin), which supports the growth of most of the pathogenic bacteria-causing mastitis. The growth of bacteria can be further confirmed by primary and secondary biochemical tests and also by molecular detection methods.

After isolation of the bacterial pathogen, further identification is carried out using phenotypic and genotypic methods. Isolation and identification of the bacterial pathogen from cases of bovine mastitis are of paramount importance for testing effective antibacterial drugs against the bacterial isolate. Phenotypic characterization of bacterial pathogen includes: a) evaluation of bacterial morphology and growth characteristics, b) testing the ability of bacteria to metabolize substrates (biochemical tests), and c) testing of the antimicrobial sensitivity. Many of the commercial bacterial identification testing kits are developed using these phenotypic traits. These tests are easy to perform, readily available in the markets and cost effective. However, growing mastitis causing bacteria or fungi in the laboratory requires time and manpower. The major limitation for using tests based on phenotypic characterization is the variable expression of phenotypic characteristics by bacteria, even if the bacterial isolates are from the same species. This led to difficulty in identifying the bacteria correctly. However, culture systems at the farm site are increasingly being used for the diagnosis of mastitis [38].

Staphylococci—Colonies of *Staphylococcus aureus* (gram-positive cocci, generally appearing as irregularly arranged clusters) are round, shiny, golden yellow. These colonies are surrounded by a double zone of hemolysis on blood agar. Baired Parker agar and Mannitol salt agar are the selective growth media used to grow this species. Coagulase test (slide agglutination test and tube agglutination test) gives positive result in case there is a pathogenic strain of staphylococci. Streptococci (grampositive cocci, generally arranged irregularly in chains) produce small translucent colonies on blood gar (with alpha, beta, and gamma hemolysis) [26, 37].

Streptococci-Edward's medium is a selective as well as an indicator medium for haemolysis and aesculin hydrolysis. Darkening of colonies shows hydrolysis of aesculin. Streptococcus uberis and Enterococcus faecalis hydrolyze aesculin. Streptococcus agalactiae and Streptococcus dysgalactiae are hydrolysis negative. Only Enterococcus grows on MacConkey agar and produces red pinpoint colonies. Christie-Atkins-Munch-Peterson (CAMP) test is used to identify hemolytic streptococci [26, 37].

Coliform bacteria—Escherichia coli, Klebsiella pneumoniae, and Enterobacter aerogenes are the most common species of bacteria isolated from milk samples collected from bovines suffering due to mastitis. MacConkey agar is used to grow coliform bacteria. Escherichia coli gives a metallic sheen appearance on Eosin Methylene Blue (EMB) agar. Colonies of Escherichia coli are generally non-mucoid. Klebsiella pneumoniae (non-motile) and Enterobacter aerogenes colonies are generally mucoid [26, 37].

### 5.4 Species and strain determination for a bacterium isolated from a case of mastitis

Nowadays, species and strain determination for a bacterium are carried out using a technique based on matrix-assisted laser desorption ionization—time of flight mass spectrometry (MALDI-TOF MS). It is a reliable, easy to use, and cost-effective technique. This technique can have 100% sensitivity and specificity for identifying the infectious agent from the cases of mastitis [39–42]. The major limitation of MALDI-TOF MS is, it can only have existing bacterial protein profiles for any specific interpretation. The technology is not available to common laboratories that have mastitis diagnosis facilities in the state of Madhya Pradesh. Genotypic methods use nucleic acid for the identification of species as well as strain typing [43]. Polymerase chain reaction

Molecular method	Species level	Strain level	Reference
AFLP	✓		[44]
RFLP		✓	[45]
MLVA		✓	[46]
Ribotyping	✓		[47]
tDNAiSLP	✓	✓	[48, 49]
PFGE			[50]
DNA Sequencing		V	[51]

AFLP, Amplified fragment length polymorphism; RFLP, Restriction fragment length polymorphism; MLVA, Multiple locus variable-number tandem repeat analysis; tDNAiSLP, Transfer DNA intergenic spacer length polymorphism analysis; PFGE, Pulsed field gel electrophoresis.

**Table 2.**Molecular methods for identification of infectious agents of mastitis.

(PCR) is highly sensitive and the most commonly used molecular method to amplify target nucleic acid of a specific infectious agent causing mastitis. Virulent strains of organisms and those organisms that are not grown in culturing are also identified by using these molecular diagnostic methods (**Table 2**).

#### 5.5 Electrical conductivity test

An increase in conductivity of milk from bovines suffering from mastitis is due to an increase in the salt concentration, which can be measured by an electrical conductivity test. Mastitis led to changes in ion concentrations [52], which impacts on the electrical conductivity of milk. Electrical conductivity can be measured [53] and the electrical conductivity rises with the increase in the concentration of sodium chloride in milk. Therefore, the measurement of electrical conductivity is used as a simple physical method to diagnose mastitis. An electrical conductivity meter is used to determine the electrical conductivity (EC). The EC of milk is expressed in the unit of milliSiemens (mS). This test has the following advantages:

1) One-time marginal investment is enough, 2) no special training is needed, and 3) easy to do and results are readily available.

#### 5.6 Somatic cell count (SCC)

An increase in somatic cell count (SCC) in milk samples from bovines suffering from mastitis is measured by various tests such as CMT, white slide test, direct microscopic count, catalase test and anti-trypsin test, Brabant mastitis test (BMT), and Wisconsin mastitis test (WMT). Because of inflammation, the composition of milk in the animal suffering from mastitis is changed from normal to abnormal [54]. Somatic cell count is used to diagnose subclinical mastitis. The SCC includes direct microscopic somatic cell count (DMSCC), the bulk milk somatic cell count (BMSCC), and individual cow somatic cell count (ICSCC). The BMSCC is the universally accepted screening test for mastitis. In DMSCC, milk sample is smeared on a clean glass slide in the area of 1 cm² and stained with 1% methylene blue to examine 60 fields under the microscope for the count. The average number of cells in the fields is multiplied by the multiplication factor of the microscope to obtain the number of cells/ml of milk sample. Electronic somatic cell counters are used for DMSCC. A count of less than 1,00,000 signifies normal udder [1, 55, 56].

#### 5.7 Chlorine test

An increase in chloride concentration of milk is an indicator of mastitis and it can be detected chemically. The presence of an increased quantity of chlorides in mastitic milk forms the basis of the test whereas normal milk contains about 0.07% chlorides. In mastitis, there is decreased amount of lactose and an increased amount of sodium chloride to maintain the normal milk osmotic pressure; hence during inflammation, there is an increase in the chloride content (> 0.14 percent) [57].

#### 5.8 N-acetyl-B-D-glucosaminidase (NAGASE) test

This is a enzyme-based test that measure cell-associated enzyme N-acetyl-B-D-glucosaminidase in the milk. The highest level of the enzyme indicates a high cell count. It is a simple, effective, and most reliable test for the detection of subclinical mastitis. The readings for normal milk with  $0.5 \times 10^4$  cells/ml and for mastitis milk with  $1.5 \times 10^4$  cells/ml are 0.0053 and 0.034 moles/min/ml, respectively [58].

#### 5.9 Indicators

Indicators of inflammation used to diagnose mastitis are well documented [59]. Recently, acute phase proteins were compared for diagnosis of subclinical mastitis in cross-bred cows in India [60].

#### 5.10 Serological tests

Serological tests such as dot blot and enzyme-linked immunosorbent assays were used to detect antibodies to *Listeria* species antigens in the milk [61].

#### 6. Mastitis prevention and control strategies

Five-point core plan for mastitis prevention and control include the following—a) disinfection of teats, b) adherence to hygienic practices in the milking procedures, c) removal of cows with chronic mastitis, d) dry cow therapy with antibiotics, and e) treatment of clinical mastitis [62, 63]. Additionally, in cow herds the measures used in prevention and control of mastitis are as follows—a) reduce introduction of new infections, b) shorten the duration of existing infections, c) maintenance of the normal udder health [64], d) dry and clean storehouse for fodder, feeders, and mangers, e) supply of palatable water, f) disinfection of milking area, g) shoe dipping disinfecting solutions for visitors to check any entry of the pathogen, h) check spread of infection by sprinkling lime powder [65], and i) keeping dairy animals stress free that encourage the development of healthy immune system [66].

10 point mastitis control program in cow herds recommended by National Mastitis Council is described in brief as follows: 1) establishment of goals for udder health and review those to prioritize changes in management to achieve set goals, 2) maintenance of a clean and comfortable environment by bedding managment, keeping areas clean and dry, ensuring proper ventilation and provision of feed soon after milking so that animals remain in standing position, 3) adopting proper milking procedures by washing and drying teats, cleaning of udder with single-use clothes, examining foremilk and palpating glands to facilitate early detection of clinical cases, maintaining clean hands or wear gloves during the process of milking, 4) proper maintenance and use of milking equipment by routinely servicing it

and thoroughly washing and sanitizing, 5) good record keeping of incidence and prevalence of clinical and subclinical mastitis in cow herd, individual examinations, and treatments, 6) appropriate management of clinical mastitis during lactation by selecting appropriate therapeutic regimen and avoid treatment of cases suffering from resistant microbial and non-responsive agents, 7) effective dry cow management by drying cows off abruptly and careful administration of all quarters of all cows with suitable antibiotic, 8) maintenance of biosecurity for contagious pathogens by assessing test reports of cows (SCC, CMT) and also obtaining aseptically collected milk cultures from suspect cows before purchasing are must. Newly purchased cows must be kept and milked separately to ensure the absence of intramammary infection. Cows with a persistently high SCC (greater than 3,00,000) are segregated and observed for response to the treatment. Marketing of chronically infected cows with Staphylococcus aureus and antimicrobial-resistant microbial agents (Mycoplasma, Nocardia, Pseudomonas, Arcanobacterium pyogenes) that are unresponsive to treatment is carried out, and 9) regular monitoring of udder health status by enrolling cows for SCC program and monitoring rates and distributions of cows with high SCC. These cows with high SCC and cows with clinical infection are used for cultural examination. Inflammation is monitored by cow-side CMT. Reports from the regulatory agencies and marketing organizations are used for monitoring udder health for the herd, and 10) periodic review of the mastitis control program is evaluated by representatives from veterinary, industry and extension fields.

Bedding materials are the primary source of environmental mastitis. Hence, reducing the bacterial count in bedding generally decreases the risk of environmental mastitis. Teat cleaning (which includes wet cleaning followed by manual drying)/pre-milking teat dipping in antiseptic solution is important to reduce bacterial counts on the skin. In cows kept indoors, it reduces the incidence of new intramammary infection [67]. Teat dipping after milking dairy animals results in a reduction in the rate of new infection significantly. It is most effective against contagious mastitis pathogens such as *Staphylococcus aureus* and *Streptococcus agalactiae*. During the dry period, administration of, broad-spectrum antibiotics in each quarter of the udder at the last milking of lactation reduces the incidence of new intramammary infections. Dry cow therapy is the best way to cure chronic and subclinical mastitis. Effective and timely treatment of active cases is helpful to prevent new cases. Antibacterial therapy through the intramammary route is not always successful for treatment against mastitis. Particular antibiotics must be selected based on nature of the pathogen, results of antibiogram assay, and drug characteristics [68–70].

Farmer's awareness about a) what causes mastitis and b) how to prevent it, is essential to minimize economic losses not only in the state of Madhya Pradesh but also in India [71, 72]. Feeding micronutrients such as selenium and vitamin E are also very important to boost the immune response of the animals. Besides, recommending improvements in managemental practices, there must be proper attention made to provide better nutrition to animals for increasing immunity and reducing stress, and also encouraging farmers to participate in various awareness programs [73]. Knowledge of risk factors and characteristics of pathogens causing mastitis are essential to control the disease at farm level [74–77].

#### 7. Summary

Mastitis affects the bovine population in India, it is also of major health concern to the bovine population of the state of Madhya Pradesh. Madhya Pradesh ranks fifth in milk production in India. Activities related to milk production and sale fetch many livelihoods in the state. Bovine cases of clinical and subclinical mastitis caused by various pathogens are reported from the state and have economic implications for the dairy animal owners in terms of direct and indirect losses. There are various mastitis diagnostic tests routinely carried out by the institutes in the state for early and timely diagnosis of mastitis. It is aiding the authorities involved in livestock health for the better treatment of mastitis. Farmer awareness campaigns are undergoing but more impetus is needed to spread the word among them for better implementation of mastitis prevention and control strategies in bovines recommended by the National Mastitis Council.

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