

ASSESSMENT OF EFFICACY OF CEFTIZOXIME AND MARBOFLOXACIN PARENTRAL
IN TREATMENT OF CLINICAL MASTITIS IN BUFFALOES

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ABSTRACT

The study aimed to assess the effectiveness of Ceftizoxime and marbofloxacin antibiotic on bacteriological cure, clinical cure and pathogen cure in lactation clinical mastitic (CM) buffaloes. The research was carried out between September 2021 and March 2022 on 48 lactating buffaloes with clinical mastitis (Treatment group, T1 = 24 and Treatment group T2 = 24). Most of the buffaloes were from farmers' doorstep and some from buffalo herds of Bidar in Karnataka. Follow-ups were done on day 7 and day 14. Buffaloes (24) in different lactation with infected quarters received ceftizoxime treatment on day 0 and repeated after day 3. while treatment (T2) buffaloes (24) in different lactation with infected quarters received marbofloxacin treatment on day 0 and repeated after day 3. All the animals received supportive therapy for five days. In Treatment group 1 (T1), 91.66% (22/24) of milk samples tested positive in culture on day 0, while 29.16% (7/24) and 20.83% (5/24) of culture-positive samples were found on day 7 and day 14 after treatment. In Treatment group 2 (T2), 95.83% (23/24) of milk samples tested positive in culture on day 0, while 20.83% (5/24) and 8.33% (2/24) of culture-positive samples were found on day 7 and day 14 after treatment.

The most common causative agent was *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus* spp., *Pseudomonas* and mixed infection. Although the bacterial cure rate for IMI was 70.84% at day 7 after treatment in Treatment group 1, it increased to 79.17% at day 14 in the same group. A bacteriological cure rate of 87.50% was observed 7 days after treatment in Treatment group 2, while a rate of 91.66% was observed 14 days post-treatment. The Treatment group T2 exhibited slightly superior clinical and pathogen cure rates to the Treatment group T1.

Keywords: *Bubalus bubalis*, buffaloes, lactation, clinical mastitis, Ceftiozoxime, marbofloxacin, therapy

INTRODUCTION

Mastitis involves inflammation of the mammary gland with physical and chemical alterations in the milk and pathological changes in the glandular tissue. Bovine mastitis, a highly important disease for the Indian dairy sector, economically results in losses of 7165.51 crores. Decreased milk production, lower fertility rates, costs of replacing dairy animals and higher

treatment expenses are some of the economic losses associated with mastitis. In India, half of dairy animals suffer from mastitis, with clinical mastitis making up 10% of cases (Dua, 2001; Ali *et al.*, 2021). Several factors, including age, parity, stage of lactation, milk yield, history of previous mastitis, udder and teat morphology, udder hygiene, floor type, season and milking practices can affect the occurrence of clinical or subclinical mastitis (Halasa *et al.*, 2007). *Staphylococci*, *E. Coli* and *Streptococci* groups are the most common bacteria linked to bovine mastitis worldwide. The gold standard for diagnosing of intramammary infection (IMI) is the bacteriological culture (BC) of mastitis pathogens in milk samples. Somatic cell count (SCC) is often used as a screening test to track udder health status in dairy herds.

Monitoring of antibacterial susceptibility should be done carefully and consistently due to the risk of microbial resistance caused by antibiotics used in mastitis treatment. It is important to track trends over time for effective antibiotic use and to help veterinarians select the best treatment for mastitis, given antibiotics are often administered before susceptibility testing. The susceptibility results in the study may not fully represent current bacterial susceptibility in mastitis but serve as a reference point. This research aimed to compare how effective ceftizoxime and marbofloxacin are in treating clinical mastitis (CM) in terms of bacteriological, clinical and pathogen cure.

MATERIALS AND METHODS

Study population

The research on evaluating the effectiveness of ceftizoxime and marbofloxacin are in treating clinical mastitis (CM) was carried

out as a randomized study that spanned from September 2021 to March 2022. It focused on lactating buffaloes affected by clinical mastitis, primarily at the farmers' homes and included organized buffalo farms from Bidar in Karnataka. Buffaloes identified with clinical mastitis in one or more of their quarters at any stage of lactation were randomly assigned to either the Treatment group (T1), or the Treatment group (T2), making 24 animals in each group. Animals with clinical mastitis in the Treatment group (T1) received Ceftizoxime injection on day 0 and repeated after day 3. The Treatment group (T2, 24 animals) also received marbofloxacin injection on day 0 and repeated after day 3. Both groups received supportive treatment for 5 days. The study data was first analysed after the first and second follow-up periods, which were conducted at day 7 and day 14 respectively.

Sample collection

The study animals were observed for up to two weeks after treatment and gathered milk samples. Before treatment, right before starting the therapy and then again on the 7th day and 14th days after treatment. The California mastitis test (CMT) was performed on the side of the buffalo where the infection was found before taking the milk samples at day 0. The CMT score was considered zero (negative) if the milk and reagent mixture looked normal and it reached its highest score of 3 (maximum) when a gel was formed and the mixture was hard to stir. To prepare the udder for taking the samples, the teats were cleaned and dried with a paper towel, followed by disinfecting the teat ends with gauze soaked in 70% ethanol. The initial flow of 3 to 4 strips of foremilk was discarded to remove any bacteria in the teat canal and then 10 ml of milk was carefully collected from the infected teat using

Table 1. Grades of mastitis based on associated clinical signs.

Grades of mastitis	Clinical signs
Grade 1	Abnormal milk
Grade 2	Abnormal milk, inflamed udder

Table 2. Treatment protocol for Treatment and Control group.

Groups	Period	No. of animals	Grades of mastitis	SCC	Milk and Clinical recovery
Group 1	Day 7	5	I	4.6±0.24	15
	Day 14	8	II	5.7±0.36	18
Group 2	Day 7	5	I	5.26±0.82	17
	Day 14	8	II	6.37±0.35	19

Table 3. Overall distribution of bacterial species and prevalence of clinical mastitis (n=48) at day 0, day 7 and day 14.

Period	Day 0				Day 7				Day 14			
	<i>Staph aureus</i>	<i>Streptococcus</i>	<i>E. coli</i>	<i>Pseudomonas</i>	<i>Staph aureus</i>	<i>Streptococcus</i>	<i>E. coli</i>	<i>Pseudomonas</i>	<i>Staph aureus</i>	<i>Streptococcus</i>	<i>E. coli</i>	<i>Pseudomonas</i>
Treatment group (n=24)	8	6	5	3	3	2	2	0	2	1	2	0
Treatment group (n=24)	8	7	5	3	3	1	1	0	1	1	0	0

sterile, disposable plastic tubes that could hold 15 ml each.

In Department of Microbiology at Veterinary College, Bidar bacterial culture, identification and isolation was carried out as per standard procedures. The segregation of microscopic organisms was carried out on the premise of standard methods (Schukken *et al.*, 2013). A loopful of overnight incubated milk sample was streaked on fibrinated sheep blood agar and MacConkey's agar plates and incubated at 37°C for 24 to 48 h. Plates with the development of three or more colony sorts of bacteria were considered for further isolation of bacterial species. Sample colonies were decontaminated through sub culturing on selective media and subjected to beginning recognizable proof by colony morphology and gram staining. Assist recognizable proof of the microbes was done as per the rules of national mastitis chamber (Seeger *et al.*, 2003). Gram-positive microscopic organisms were separated as *Staphylococci* and *Streptococci*. Gram-negative microscopic organisms were distinguished and separated by oxidase test, KOH test, IMViC tests (indole, methyl ruddy and Voges-Proskauer responses and Simmons's citrate), nitrate diminishment, triple sugar press agar test as well as by development highlights on MacConkey agar, eosin methylene blue agar and green pigmentation on supplement agar by *pseudomonas* (Schukken *et al.*, 2013).

Definition of infection

Bacteriological cure

The treated quarters were stated as bacteriologically cured based on that an udder pathogen displayed in the pre-treatment drained milk sample and absent in the post-treatment drain milk sample.

Clinical cure

The affected quarters were said to be clinical cured, when its clinical scores such as abnormal milk, inflamed hard udder and systemic infection were present on day 0 pre-treatments and followed signs disappear on day 7 and/or day 14 post treatment.

Pathogen cure

When both follow-up tests of milk sample (at day 7 and day 14) contained no pathogens in culture, the creature was characterized as a pathogen remedy.

RESULTS AND DISCUSSIONS

Prevalence of clinical mastitis and udder pathogens at day 0 (pre-treatment)

Based on the result of cultural examination of milk sample (Table 3) clinical mastitis in buffalo was at the prevalence rate of 10.32% at day 0 pre-treatment. The present study for clinical mastitis is in agreement with Hussain *et al.* (2018) recorded 10.20% (106/1036) of clinical mastitis in two districts, Lahore and Bhimber Azad Kashmir Pakistan in buffaloes. In present study found that the foremost predominant causative organism was *Staphylococcus aureus* speaking to 52.45%, *Escherichia coli* (22.95%), *Streptococcus spp.* (14.20%), *Pseudomonas* (4.91%), Present findings were in agreement with Didugu *et al.* (2015); Verma *et al.* (2018).

Bacterial cure rate for IMI was 70.84% at day 7 after treatment in Treatment group 1, it increased to 79.17% at day 14 in the same group. Disposition kinetics of ceftizoxime in *S. aureus*-induced acute mastitis in Murrah buffaloes following 4 administration of ceftizoxime (25 mg/

kg) was studied by Kumar *et al.* (2016). This is the first published study of disposition and efficacy of ceftizoxime in Indian mastitis crossbred cows under field conditions. Since the introduction of marbofloxacin for Veterinary use, it is being first line of treatment of acute clinical mastitis (Pillet *et al.*, 2013). In present investigation the overall cure rate of 87.50% was observed 7 days after treatment in Treatment group 2, while a rate of 91.66% was observed 14 days post-treatment, which is substantiated by earlier reports of Patil *et al.* (2021); Rahimiyan *et al.* (2021).

The Treatment group T2 exhibited slightly superior clinical and pathogen cure rates to the Treatment group T1.

Bacteriological cure rate

In Treatment group T1 had *Staphylococcus aureus* to (62.50%), *Escherichia coli* (66.66%), *Streptococcus* spp. (60.00%) and *Pseudomonas* (100%) whereas in *Staphylococcus aureus* (75.00%), *Escherichia coli* (60.00%), *Streptococcus* spp. (83.33%) and *Pseudomonas* (100%) on day 7 and day 14 respectively. Similarly in Treatment group T2 *Staphylococcus aureus* to (62.50%), *Escherichia coli* (80.00%), *Streptococcus* spp (85.71%) and *Pseudomonas* (100%). whereas in *Staphylococcus aureus* to (87.50%), *Escherichia coli* (100%) and *Streptococcus* spp. (85.71%) on day 7 and day 14 respectively.

Clinical cure

In Treatment group T1, 62.50% of buffaloes (n=15/24) experienced clinical remedy or zero clinical signs (clinical signs such as abnormal milk drain, inflamed hard udder and systemic infection) at day 7 and 75.00% (n=18/24) at day 14 post-treatment. Of the marbofloxacin treated (T2) buffaloes, 70.83% (n=17/24) experienced clinical

remedy at day 7 while 79.16% (19/24) on day 14 of post-treatment. (Ramesh *et al.*, 2015; Yadav *et al.*, 2022) clinical cure rate noticed in was 82.6%.

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