

Full Length Research Paper

Management of subclinical mastitis with mastilep gel and herbal spray (AV/AMS/15)

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A clinical trial was conducted to evaluate therapeutic efficacy of Mastilep gel and topical herbal spray (AV/AMS/15) against treatment of sub clinical mastitis. The mastitis suspected bovine cases presented to Teaching Veterinary Clinical Complex (TVCC) and the instructional livestock farm, College of Veterinary and Animal Sciences, Parbhani, Maharashtra state, India were incorporated in study. The cows were screened by performing Mastrip test and Modified California Mastitis Test. All animals were grouped in 3 groups of 10 animals in each. Group A (Control Group) was given no treatment, Group B – treated with herbal spray (AV/AMS/15) sprayed on udder/affected quarters post milking BID for 5 days, Group C- Mastilep gel (applied gently by massaging the udder), post milking BID for 5 days. Somatic cell count (SCC), milk fat content, milk yield was evaluated along with microbiological investigations (isolation and identification). SCC in AV/AMS/15 sprayed and Mastilep gel treated group was reduced significantly on the 5th day of treatment to 2.89 ± 0.83 and 4.01 ± 2.06 respectively. After treatment milk yield in cows suffering from sub clinical mastitis increased significantly with unchanged Milk fat %. Milk samples of all the affected quarters revealed predominance of *Staphylococcus*, *Streptococcus* and *Escherichia coli* as the causative agents of bovine sub clinical mastitis. On the basis of microbial investigation on the 5th day, 60% cure rate was recorded in Mastilep gel treated cows and 70% after treatment with AV/AMS/15 herbal formulation. Affected animals were negative for subclinical mastitis at the end of the experiment. As far as comparison between therapeutic efficacy of AV/AMS/15 herbal spray (Group B) and Mastilep gel (Group C) is concerned with both at par. However, the efficacy of AV/AMS/15 herbal spray, in reducing the SCC in milk was higher as compared to Mastilep gel and the significant difference ($P < 0.01$) was observed between them.

Key words: Mastitis, SCC, mastilep, herbal, udder.

INTRODUCTION

Mastitis is one of the most frequent infectious diseases in dairy cattle and is a reason for heavy economic losses and antimicrobial drug usage in dairy cows (Minst et al., 2012). Despite management advancements, mastitis continues to rob the dairy industry in terms of decreased milk production, veterinary services, diagnostics, drugs, discarded milk, labour, decreased product quality, increased susceptibility to diseases, increased risk of culling and materials for prevention (Hogeveen and Osteras, 2005). Mastitis has remarkably rising impact on Indian economy where overall losses due to mastitis is estimated to be Rs. 7165.51 crores (Bansal and Gupta,

2009), number which increased from 6053.2 crores in less than a decade before (Dua, 2001). 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 sign of inflammation and the milk appears normal. However, micro-organisms and somatic cells are found in elevated numbers in the milk. Bhatt et al., (2012). Subclinical mastitis was found more important in India (varying from 10 to 50% in cows and 5 to 20% in buffaloes) than clinical mastitis (1 to 10%). The incidence was highest in Purebred Holsteins and Jerseys and lowest in local cattle and buffaloes (Joshi and Gokhale, 2006). The incidence rates of sub clinical mastitis reported from different states of India by various workers were 48.7%. Bansal et al., (1995), 53.54% Tiwari et al., (2000), 56.76% Chanda et

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al., (1989). The use of synthetic antibiotics is being increasingly discouraged because their presence in dairy milk may have potential downstream effects on population health and the Agri-food chain (Taga et al., 2012). As demand for higher milk quality standards increases, dairy producers are concerned to improve efforts to control mastitis through prevention and treatment. There comes the need to find sustainable alternatives for control of mastitis. The constituent herbal ingredients of biotherapy viz. *Cedrus deodara*, Thakur et al., (1989). *Curcuma longa* Bone (1991), Ammon et al., (1993). *Glycyrrhiza glabra* Akamatsu et al., (1991) and *Eucalyptus globulus* Satyavati (1976) are used in alternative medicine for their antibacterial, anti-inflammatory, analgesic, anti-histaminic and immunomodulatory properties. These herbal ingredients at various concentrations have been combined in the Mastilep gel and herbal spray (AV/AMS/15). Right from its inception Mastilep gel has been documented for its effective curing potential against the mastitis of dairy animals (Maini et al., 2008; Maiti et al., 2006; Nath and Dutta 2005; Gawai et al., 2002; Rajhora and Pachauri 2002; Sharda et al., 2001; Tiwari et al., 2001; Rahman and Sharma 2000). Topical applications in the form of spray are rapid to use and require minimum effort for application. With raising concern over microbial resistance and to find out a sustainable remedial measure of herbal combination coded product (AV/AMS/15) was developed to curb mastitis problem in dairy cattle. Thus the present study was undertaken with the objective to evaluate therapeutic efficacy of Mastilep gel and topical herbal spray (AV/AMS/15) individually against treatment of sub-clinical mastitis in crossbred cows during lactation.

MATERIALS AND METHODS

The crossbred cows suspected for mastitis were presented to the Teaching Veterinary Clinical Complex (TVCC) and the instructional livestock farm College of Veterinary and Animal Sciences Parbhani, Maharashtra state, India. History pertaining to physiological status of an individual animal e.g. age, milk yield per day, lactation no, lactation stage was collected and conducted detailed clinical examination. Colour, consistency, odour, taste of milk and clinical manifestation if any, was also recorded. The cows were screened for sub clinical mastitis by performing Mastrip test and Modified California Mastitis Test (MCMT).

Modified California Mastitis Test (MCMT)

The Modified California Mastitis Test was used for diagnosis of sub clinical mastitis due to its reliable, easy, rapid and cheap tool helping in diagnosis and controlling

the disease. Viani et al., (1990), Behera and Dwivedi (1992) El- Balkemy et al., (1997). It gives an indirect estimate of SCC because it is based upon a gelling reaction between the nucleic acid of the cells and a detergent reagent. Cows with sub clinical mastitis detected with MCMT were selected and distributed among three groups each containing 10 animals (one control group and two treatments). First treatment group (Group B) was treated with herbal spray (AV/AMS/15), while second treatment group (Group C) was treated with application of herbal topical application of Mastilep gel. Both the herbal formulations were sprayed and applied gently to the cow udders respectively twice daily after every milking for five consecutive days. Control group was not given any treatment. Milk samples were collected before and after spraying and application of herbal preparations. The sampling was done at the milking time in the evening hours from all the four quarters of cow after thorough washing of udder with suitable antiseptic solution and discarding first few strips. Individual samples were collected in sterile glass vials of 100 ml capacity. The samples were immediately subjected to the detection of SCC, estimation of milk fat and microbiological investigations such as, isolation of bacterial agents responsible for clinical & subclinical mastitis. SCC is determined by the method described by Schalm *et al.*, (1971) except staining, which is usually done as per the method described by Newlander and Atherton (1964). Fat content of milk was calculated using Gerber's method, Kleyn *et al*(2001). Isolation of bacteria was done by streaking the samples on blood agar and incubating aerobically at 37^o C, for 24 hours Cruickshank *et al* (1975). The isolates were tentatively identified by Gram's staining and on the basis of morphological characters. The *in vitro* antibacterial sensitivity pattern of these bacterial isolates was determined by a standard disc diffusion technique. Bauer *et al* (1966) using antibiotic discs for recording antibiogram of the isolates. Therapeutic efficacy was determined on the basis of reduction in SCC, milk yield, milk fat content and microbiological investigations (bacterial load in the milk). The data was analyzed according to the methods described by Snedecor and Cochran (1994).

RESULTS AND DISCUSSION

Prevalence

In the present study high incidence of mastitis was observed which may be attributed to humid climate, stress of high milk yield, breed predilection and non-implementation of strict hygienic measures for control of mastitis. The present experiment findings of season also corroborates with the findings of Abdel-Rady and Sayed (2009) who noticed higher prevalence of sub clinical mastitis in hot weather than in winter. As regard to distrib-

Table 1. SCC, milk yield, milk fat content % before and after treatment with AV/AMS/15 herbal spray and Mastilep gel.

Group	SCC ($\times 10^5$ cells/ml)		Milk yield (lit/cow/day)		Milk fat content %	
	Pre treatment	5 th day of treatment	Pre treatment	5 th day of treatment	Pre treatment	5 th day of treatment
Group A	6.01 \pm 4.03 ^a	6.69 \pm 3.52 ^a	9.1 \pm 1.57 ^a	9.0 \pm 2.05 ^b	3.1 \pm 0.62 ^a	3.0 \pm 1.14 ^b
Group B	6.01 \pm 4.62 ^a	3.21 \pm 1.93 ^c	9.3 \pm 0.36 ^a	9.5 \pm 1.07 ^a	3.1 \pm 0.07 ^a	3.1 \pm 0.04 ^a
Group C	6.29 \pm 3.33 ^a	4.01 \pm 2.06 ^b	9.2 \pm 0.32 ^a	9.4 \pm 0.26 ^a	3.1 \pm 0.69 ^a	3.1 \pm 0.71 ^a

Values having same superscripts in the column do not differ significantly from each other. Level of significance (P<0.01).

ution among infected quarters higher prevalence was recorded in right-hind quarter (37.78%) followed by left-hind quarter (26.08%) while it was (21.73%) in right-fore and (17.39%) in left-fore quarters respectively. The similar pattern of affection of quarters i.e. right hind (28.98 %), left hind (27.53%), right fore (23.67%) and left fore (19.80 %) was observed by Shastri (2001). The highest prevalence in hind quarter affection might be more due to continuous soiling of hind teats during defecation and urination and subsequently ready environment for microorganisms to grow.

Somatic Cell Count

Results of SCC, milk yield, milk fat content % before and after treatment with AV/AMS/15 herbal spray and Mastilep gel have been summarized in table 1. SCC in milk is an indication of the presence of udder infection, Radostitis *et al* (2007). Determination of SCC is reliable tool for diagnosis of sub clinical mastitis. In present study, there were no visible changes in udder tissue or gross abnormalities in milk secretion were not obvious but there was complaint of reduction in milk yield and the samples revealed reasonable increase leucocyte count in the milk. The mean SCC of affected quarters was $6.01 \times 10^5 \pm 4.62$ cells / ml which was higher than threshold of 3,50,000 cells/ ml of milk and were identified to be affected with sub-clinical mastitis. Radostitis *et al.*, (2007).

Milk Yield and Milk Fat

On clinical examination there was no visible abnormality in udder and milk. In few of the milk samples abnormalities of slight discoloration of milk were observed. The colour of milk was changed to faint yellow. The milk fat % in the sub clinical mastitis affected animals

ranged from 3.1 \pm 0.07% to 3.1 \pm 0.69%. There was average 0.4 % reduction in milk fat in affected animals. The change in the consistency of milk from normal to watery might have affected the average fat %. Singh *et al.*, (2006).

Microbiological investigation

Milk samples of all the affected quarters were found positive bacteriologically. The findings of the current study revealed predominance of *Staphylococcus*, *Streptococcus* and *Escherichia coli* as the causative agents of bovine sub clinical mastitis. The different isolates included *Staphylococcus aureus* (70%), *Staphylococcus aureus* and *Streptococcus agalactiae* (10%), *Staphylococcus aureus* and *Escherichia coli* (5%), *Staphylococcus aureus* and *Bacillus cereus* (5%), and *Escherichia coli* (5%) were also isolated in the present study. This is in agreement to the earlier reports of Abdel-Rady and Sayed(2009), Bhalerao *et al.*, (2000), Pednekar and Swarup (1991), Singh and Baxi (1982), Rahman *et al.*, (1984).

Herbal Spray (AV/AMS/15) post treatment efficacy

After treatment milk yield and milk fat were optimized and reached near to normalcy. Their values on 5th day of treatment changed from 9.3 \pm 0.36 lit /day to 9.5 \pm 1.07 lit/day and 3.1 \pm 0.071% to 3.1 \pm 0.04% respectively however were non-significant. Mean SCC of milk reduced from pretreatment $6.01 \times 10^5 \pm 4.62$ cells /ml to $3.21 \times 10^5 \pm 1.93$ cells /ml on 5th day of treatment suggested 46.47 % improvement in SCC. Following five days herbal spray therapy, clinical udder manifestations started subsiding and of the 10 bacteriologically positive milk samples, 7 were found negative on 5th day of treatment

giving cure rate 70.00%. The AV/AMS/15 herbal spray contain herbal ingredients with antibacterial, anti-inflammatory, analgesic and immunostimulatory properties and thus might be effective against not any specific organism but plethora of organisms, which are responsible for persistence of disease (Singh *et al.*, 2006).

Herbal Mastilep Gel post treatment efficacy

After treatment average milk yield increased from 9.2 ± 0.32 lit/day to 9.4 ± 0.26 lit/day on 5th day of treatment however the milk fat level remained unchanged (3.1 ± 0.071). Significant difference was observed in SCC in pretreatment ($6.29 \times 10^5 \pm 3.33$ cells/ml) and post treatment ($4.01 \times 10^5 \pm 2.06$) milk samples. On bacteriological examination, out of 10 positive milk samples, six were found negative on 5th day of treatment giving the 60% cure rate. The present results have further confirmed the earlier studies on Mastilep. Joshi *et al.*, (1996). Buragohain and Dutta (1998) reported 100 percent cure of sub clinical mastitis in herbal gel Mastilep treated groups for 7-14 days. As far as comparison between therapeutic efficacy of AV/AMS/15 herbal spray (Group B) and Mastilep gel (Group C) is concerned both were at par. Non-significant difference was observed with any of the parameters when Group B and Group C were compared with each other. The efficacy of the herbal gel is due to the combined action of the ingredient herbs having immunopotentiating activity to enhance the body's defense mechanism along with udder immunity, thereby keeping all sort of intramammary infections at bay. Singh *et al.*, (2006).

CONCLUSION

The improvement in milk yield and milk fat and reduction in SCC after the treatment indicated that herbal therapy by potentiating the udder immunity, not only eliminates udder infection in sub clinical mastitis but also control the mastitis without any side effects. It also augments repair of mammary gland, firmness and normalize udder functioning with improved milk quality. In the present study the overall cure rate and prophylactic efficacy by AV/AMS/15 topical herbal spray was good and at par to Mastilep gel thus may be recommended for treatment and control of sub clinical Mastitis in bovines.

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