

Efficacy of *Moringa oleifera* leaves infused oil on sub clinical mastitis in goats

Nisha¹, P. N. Panigrahi^{1*}, M. Jaiswal¹, M. Anand², S. Vaswani³, A. K. Tripathy¹

¹Department of Veterinary Medicine, ²Department of Animal Physiology, ³Department of Animal Nutrition, College of Veterinary Science and Animal Husbandry, DUVASU, Mathura, UP-281001

Abstract

The present study was conducted to determine the efficacy of *Moringa oleifera* leaves infused oil on sub clinical mastitis in goats. A total of 217 goats were screened for this study out of which 50 goats were found positive for sub clinical mastitis. For therapeutic trial, six goats (12 quarters) were considered apparently healthy control group (group I) after thorough physical examination and various diagnostic tests. Out of 50 sub clinical mastitis goats, six goats (12 quarters) were taken as treatment group (group II) where topical application of *Moringa oleifera* leaves powder infused oil was given for seven days twice daily. Therapeutic efficacy of *Moringa oleifera* leaves was evaluated based on alteration of somatic cell count of milk and qualitative changes in milk. A significant increase in SCC and pH of milk and significantly decreased concentration of fat and solid not fat % was reported in SCM affected animals (group II) as compared to healthy control (group I) before treatment. *Moringa oleifera* significantly reduced the SCC and pH as well as significantly increased the fat and SNF %. It is concluded that, *Moringa oleifera* leaves infused oil was found effective against sub clinical mastitis as evidenced by restoration of somatic cell count and other milk parameters.

Key words: Sub clinical mastitis; goat; *Moringa oleifera*; somatic cell count

Sub clinical mastitis (SCM) is one of the important constraint in animal husbandry sector throughout the globe especially in growing countries like India. In Uttar Pradesh, India, prevalence of mastitis occurs throughout the year both in clinical and sub-clinical form amounting to huge economic annual loss of more than Rs. 1311.767 crores (Singh *et al.*, 2021). Goat, known as the poor man's cow usually reared for milk and meat in every climatic condition in India. Now-a-days rearing of goat for milk has been increasing as goat milk is wholesome, easily digestible, nutritious and has medicinal properties. Goat milk shares a contribution of 3% in the total milk across the country. The prevalence of SCM in lactating goats had been ranges from 9-50 % (Hall and Rycroft, 2007; Min *et al.*, 2007; Marogna *et al.*, 2012). Diagnosis of subclinical mastitis (SCM) can be made either direct or indirect measurement of milk somatic cell count (SCC) and isolation and identification of mastitis pathogens in milk. California mastitis test (CMT) and White side test (WST) were mostly used as indirect diagnostic tests, whereas SCC was used as direct laboratory diagnostic methods for detection of SCM.

Treatment and management of SCM was usually done with anti-inflammatory, antibacterial, antifungal and analgesic drugs. But, most of them were reported

ineffective, costliest as well as tolerated by mastitis causing pathogens due to their rampant use and abuse. Indeed, SCM is a multi etiological disease and involves in alteration of several factors. Therefore, the present situation demands a newer approach of therapy. Several recent studies have highlighted the effectiveness of medicinal plants and natural compounds for treatment and management of SCM (Mooventhan *et al.*, 2016; Fozia *et al.* 2012). *Moringa oleifera* is a commonly available plant in India considered as miracle plant as whole part of plant like leaves, roots, seed, bark, fruit, flowers and immature pods possess nutritional as well as medicinal properties (Mooventhan *et al.*, 2016). Fozia *et al.* (2012) reported that *Moringa oleifera* possess anti-inflammatory, antimicrobial, antioxidant, anticancer, cardiovascular, hepatoprotective, anti-ulcer, anti-urolithiatic and anthelmintic properties. Aqueous extracts of *Moringa oleifera* were found to be inhibitory against many pathogenic bacterias, including *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, and *Pseudomonas aeruginosa* in dose dependent manner which were the potent pathogens causing mastitis in animals (Saadabi and Abu Zaid, 2011). Methanol extract of leaves has shown effective antioxidant activity (Kumar *et al.*, 2012). Taking into consideration of antibacterial, anti-inflammatory and antioxidant properties of *M. oleifera*, the present study was designed to evaluate therapeutic efficacy of *Moringa*

*Corresponding author: pnpvetmed@gmail.com

oleifera leaves infused oil in goats affected with SCM.

Materials and Methods

Animals

The present study was undertaken at Department of Veterinary Medicine, College of Veterinary Science and Animal Husbandry, DUVASU, Mathura in association with University Goat Farm and Veterinary Clinical Complex, DUVASU, Mathura. Lactating goats of any breed, more than one-year old having owner's complaint and/or history suggestive of SCM i.e., gradual loss of milk yield, salty taste, quick curdling, slimy or watery consistency were taken into consideration for this study.

Screening of animals for subclinical mastitis

Approximately 10 ml milk was collected from each animal for screening the sub clinical mastitis by California Mastitis Test (CMT) as per procedure given by (Schalm and Noorlander, 1957) using a modified CMT reagent (Sharma and Rajani, 1969). A total of 217 lactating goats (Barberi-103, Jamnapari-85, Jakhrana-11, Sirohi-4, Black Bengal-2 and Beetal-12) were screened for SCM by CMT.

Plant material

Fresh green leaves of *Moringa oleifera* were collected from medicinal herb garden, DUVAU, Mathura. The leaves were washed properly with distilled water, air dried and powdered uniformly using an electric grinder. Leaves infused oil was prepared by standard procedure and applied topically on the infected udder.

Therapeutic trial

A total of 217 lactating goats were screened for SCM by CMT out of which 50 goats were found positive for SCM. Out of which, 6 goats (12 quarters) found positive for subclinical mastitis on screening were taken as treatment group (group II) where topical application of *Moringa oleifera* leaves powder infused oil was given for seven days twice daily at 10 mg/kg (EC₅₀ dose). Rest of the sub clinical mastitis goats were taken for other study (Nisha *et al.*, 2021). Six goats (12 quarters) were considered apparently healthy control group (group I) after thorough physical examination and various diagnostic tests. Therapeutic efficacy of *Moringa oleifera* leaves was evaluated based on alteration of somatic cell count of milk and qualitative changes in milk (milk fat, solid not fat, pH, density, protein and

lactose). Therapeutic efficacy was evaluated at weekly interval for 14 days.

Analysis of milk

Approximately 5-10 ml of milk was collected from each quarter of each animal in clean sterilized collection vial on day-0 (before treatment), day-7 (7th day of treatment) and day-14 (14th day of treatment). The milk sample was taken for milk analysis and somatic cell count in automatic Lactoscan ultrasonic milk analyzer. Somatic cell count (SCC), pH of milk, fat percentage, solid not fat (SNF) percentage, density, milk protein content, lactose content was calculated in automatic Lactoscan ultrasonic milk analyzer as per manufacturer's instructions.

Statistical analysis

The data was expressed as mean \pm SEM. Standard error of mean and p-values were used to determine whether there is any significant difference among different treatment groups using one-way analysis of variance (ANOVA) following standard protocol (Snedecor and Cochran, 1994).

Results and Discussion

A total of 217 lactating goats were screened for SCM by CMT out of which 50 goats were found positive for SCM. Twenty goats were found positive for SCM in Barbari breed out of 103, 24 out of 85 in Jamnapari, 4 out of 11 in Jakhrana breed, 1 out of 4 in Sirohi, 1 out of 12 in Beetal and 0 out of 2 in Black Bengal breed.

Table 1 summarizes the alteration in different milk parameters in goats of different groups at different intervals of study. A significant ($p < 0.05$) increase in SCC was recorded in milk of goats affected with sub clinical mastitis (group II) as compared to healthy control before treatment (day 0). *Moringa oleifera* reduced the SCC significantly and the mean value was statistically similar to that of healthy control animals after treatment. On contrary, milk fat and solid not fat (SNF) % reduced significantly in SCM goats as compared to healthy control at day 0. *Moringa oleifera* significantly increased the above concentration. No significant differences were recorded in mean values of milk density, milk protein concentration and milk lactose concentration either between the groups or within groups at different observation periods of study. A significant ($p < 0.05$) increase pH of milk was recorded in group II animals on day 0 as compared to untreated healthy control. The

pH of milk decreased significantly post therapy on day 7 as well as day 14 and the values had no significant difference as that of healthy control.

In present study, somatic cell count (SCC) concentration of goats affected with sub clinical mastitis increased significantly as compared to that of the apparently healthy control group at day 0. The increase in Somatic cell count (SCC) can also be attributed to increased polymorph nuclear cells and neutrophils (Kitchen *et al.*, 1970). Whereas, after treatment with *Moringa oleifera* infused oil there was significant reduction in somatic cell count (SCC) at different interval of study indicated reduction of inflammation as a result of treatment which was in accordance with the finding of Raikwar (2012). It might be due to potent anti-inflammatory and anti-bacterial effect of *Moringa oleifera* leaves (Gilani *et al.*, 1997; Nikkon *et al.*, 2003; Dangi *et al.*, 2002).

Sub clinical mastitis causes many changes in milk composition. In the present investigation, fat and SNF percentage of milk reduced significantly in sub clinical mastitis animals but, the values were increased significantly at day 14 after the treatment. These results are in accordance with Souza *et al.* (2009) and Kifaro *et al.* (2009), but disagreement with Leitner *et al.* (2004), Merin *et al.* (2004) and Min *et al.* (2007). The observed high fat content in milk could be attributed to increased fermentation efficiency, enhanced by phyto-chemicals

rich *Moringa* in conjunction with amino acids and P, Ca, and Mg in *M. oleifera* leaves, which are essential for high milk synthesis (Kholif *et al.*, 2015).

In this investigation, density of milk, milk protein and lactose concentration followed a non significant change at different intervals of study before and after the therapy. The density of milk and lactose decreased non significantly in animals affected with SCM and these results were closed to the findings of Merin *et al.* (2004), Rulan and Shangguan (2010) and disagreement with that of Kifaro *et al.* (2009) and Moroni *et al.* (2005). Lactose is synthesized in the gland cells of udder from glucose and galactose. Milk of infected glands has a significantly lower concentration of lactose, which is accompanied by significantly higher whey and albumin concentrations. The accumulation of whey proteins probably results from a breakdown of the caseins and other milk proteins, which down-regulate milk secretion (Silanikove *et al.*, 2000; Shamay *et al.*, 2003) and explain the lower lactose concentration.

The pH was significantly higher in SCM affected goats than healthy control group at day 0 and it was reduced significantly after treatment with *Moringa oleifera*. Increased pH of SCM milk was also reported by other researchers (Hassan, 2013; Hussain *et al.*, 2012; Batavani *et al.*, 2007). The rise in pH was due to the leakage of blood bicarbonate into the milk following

Table 1: Effect of *Moringa oleifera* leaves infused oil on milk parameters in goats

Parameters		Day 0	Day 7	Day 14
SCC ($\times 10^3$ /ml)	Group I	74.33 ^{aA} ±21.72	77.83 ^{aA} ±48.06	82.83 ^{aA} ±38.08
	Group II	1476.7 ^{bB} ±224.75	323.33 ^{aA} ±82.90	189.67 ^{aA} ±55.48
Fat (%)	Group I	4.69 ^{aB} ±0.35	4.85 ^{aB} ±0.72	4.61 ^{aA} ±0.74
	Group II	3.27 ^{aA} ±0.45	3.79 ^{abA} ±0.24	4.24 ^{bA} ±0.22
SNF (%)	Group I	8.45 ^{aB} ±0.15	8.59 ^{aB} ±0.17	8.57 ^{aA} ±0.17
	Group II	7.41 ^{aA} ±0.26	7.77 ^{aA} ±0.15	8.07 ^{bA} ±0.18
Density	Group I	31.26±1.23	30.08±0.59	29.19±0.64
	Group II	26.21±0.87	26.09±1.08	26.89±0.93
Protein (%)	Group I	3.13±0.05	3.21±0.12	3.09±0.10
	Group II	3.72±0.15	3.42±0.13	3.22±0.08
Lactose (%)	Group I	4.81±0.19	4.81±0.17	4.75±0.35
	Group II	4.34±0.09	4.36±0.10	4.52±0.12
pH	Group I	6.35 ^{aA} ±0.02	6.37 ^{aA} ±0.05	6.30 ^{aA} ±0.04
	Group II	6.83 ^{bB} ±0.04	6.58 ^{aA} ±0.05	6.43 ^{aA} ±0.07

Group I= healthy control goats, group II = goats treated with *Moringa oleifera* leaves infused oil

Values (Mean ± SE) within same column for a particular parameter (capital letters) and in same row (small letter) bearing similar superscript do not differ at $P < 0.05$.

damage to the mammary epithelium (Kumhar *et al.*, 2010). The decrease in pH after *Moringa oleifera* treatment might be due to reduction in alkalinity resulted due to inflammation by increased number of Na⁺ and Cl ions in the milk (Ahmed *et al.*, 2005). The present curative results are in agreement with Aruna, (2019); Mooventham *et al.*, (2016) who studied the effect of *Moringa oleifera* leaves in cow and reported reduction in pH of milk after *Moringa* supplementation.

Based on the results of the study it was concluded that, *Moringa oleifera* leaves infused oil was found effective against sub clinical mastitis as evidenced by restoration of somatic cell count and other milk parameters.

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