



Original Research

Effect on Milk Composition of Sub Clinical Mastitis in Cows in Organized Dairy Farms

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Abstract

In the present study prevalence of subclinical mastitis (SCM) was assessed on organized dairy farms. The qualitative changes of milk in the cows with subclinical mastitis were assessed on the basis of estimation of fat percent, solid not fat percent, protein percent and lactose percent. A significant reduction in values of fat %, SNF %, protein %, lactose % was recorded in the sub clinically infected cows of Groups as compared to the values of normal healthy cow on the day of screening. Subclinical mastitis altered the composition of milk in cows which had significantly lower values of fat, SNF, protein, lactose. Simultaneously therapeutic efficacy of herbal paste/gel as topical application alone or in combination was also assessed in restoring the milk constituents. Estimation of therapeutic potential of herb alone and in combination as paste or gel was also undertaken. The therapeutic trial was based on the restoration of constituents of milk and the paste of herbal mixture was found to be the most efficacious in subclinical mastitis.

Key words: Cows, Milk Composition, Organized Dairy Farms, Sub Clinical Mastitis

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Introduction

Bovine mastitis is the inflammation of mammary gland, mainly due to microbial infection like bacteria, virus, etc. Numerous pathogens are responsible for mastitis with the majority infections caused by bacteria. Among bacterial species, mastitis is mainly caused by *Staphylococcus spp.*, *Streptococcus spp.*, *Escherichia coli*, *Corynebacterium spp.*, *Klebsiella spp.*, *Pseudomonas spp.* etc. Early diagnosis and prompt treatment of mastitis is imperative for reducing production losses and for enhancing the prospects of recovery. Mastitis could be categorized as clinical and subclinical (Duguma and Yohannes, 2014). Subclinical mastitis is an inflammation of the mammary gland without noticeable signs, with reduction in daily milk



yield and altered milk composition. In addition, it is considered as a prevailing disease in dairy herds with prevalence of 15-40 subclinical mastitis cases for every one case of clinical mastitis. The prevalence of subclinical mastitis has increased enormously in India in the recent years than bovine clinical mastitis (NAAS, 2013).

According to Tiwari *et al.* (2017) early identification of mastitis in sub-clinical stage could be helpful for implementation of effective managemental interventions to reduce the economic losses. Verma *et al.* (2018) studied that milk and its products are excellent source of vital nutrients and nearly perfect food. Milk proteins offer a high quality animal protein in diet. Milk fat fractions are now having interesting anti-cancer properties. Minerals and vitamins contribute significantly to human nutrition. The present research work highlighted the changes in milk composition due to sub clinical mastitis in cows in organized dairy farms.

Material and Methods

The study was undertaken in lactating cows reared in organized dairy farms under different phases of lactation in Durg. The animals were divided in to five Groups having six animals in each Group. Different Groups were presented in Table 1 along with their treatment. Group 1 was the healthy animal Group without mastitis and the rest four Groups were treatment Groups. In all Groups different constituents of milk like fat, solid not fat, protein and lactose was assessed and changes were analysed before and after treatment.

Table 1: Different Groups for estimation of changes in milk compositions

Groups	No. of Animals	Treatment
I	6	Healthy control without SCM
II	6	Cows with SCM and treated with Aloe vera gel
III	6	Cows with SCM and treated with Neem leaves paste
IV	6	Cows with SCM and treated with Tulsi leaves paste
V	6	Cows with SCM and treated with mixture of all herbs (Aloe Vera Gel+ Neem Leaves Paste +Tulsi Leaves Paste)

Fat Content

Milk fat content was estimated by Lactostar-milk analyzer model number-3510, Funke Gerber, Germany and the results were expressed in %.

SNF (Solid Not Fat) Content

Milk SNF content was estimated by Lactostar-milk analyzer model number- 3510, Funke Gerber, Germany and the results were expressed in %.

Protein Content

Milk protein content was estimated by Lactostar-milk analyzer model number- 3510, Funke Gerber, Germany and the results were expressed in %.

Lactose Content

Milk Lactose content was estimated by Lactostar-milk analyzer model number- 3510, Funke Gerber, Germany and the results were expressed in %.

Statistical Analysis

The mean and standard error of the recorded value was calculated. The data was analyzed statistically by using ANOVA using one way analysis of variance followed by DMRT (Duncan's multiple range test) after suitable transformation as per the procedure outlined by Snedecor and Cochran (1994) using SPSS 20 version.

Results and Discussion

Fat Content

The values (Mean±SE) of milk fat (%) recorded in the different Groups during the present study are presented in Table 2.

Table 2: Changes in milk fat % (Mean± S.E.) in sub clinical mastitis in cows

Groups	Days of Observation			P-value (with in Groups)
	0 Day	7 Day	14 Day	
Group-I	3.68±0.087 ^a	3.73±0.095 ^a	3.75±0.109 ^a	0.86
Group-II	2.73±0.071 ^b	2.81±0.091 ^c	3.01±0.102 ^b	0.115
Group-III	2.71±0.066 ^{bb}	3.02±0.079 ^{bcA}	3.12±0.057 ^{bA}	0.002
Group-IV	2.72±0.105 ^b	2.94±0.104 ^{bc}	3.06±0.093 ^b	0.082
Group-V	2.68±0.060 ^{bb}	3.12±0.113 ^{bA}	3.29±0.069 ^{bA}	<0.001

^{ABC} Means with different superscripts within a row differ significantly. ^{abc} Means with different superscripts within a column differ significantly. Group I- healthy control; Group II- cows with SCM and treated with aloe vera gel; Group III- cows with SCM and treated with neem leaves paste; Group IV- cows with SCM and treated with Tulsi leaves paste; Group V- cows with SCM and treated with mixture of all herbs

The obtained values on day 0, day 7th, day 14th in of Group I were 3.68±0.087; 3.73±0.095; 3.75±0.109, in Group II 2.73±0.071; 2.81±0.091; 3.01±0.102, in Group III 2.71±0.066; 3.02±0.079; 3.12±0.057, in Group IV 2.72±0.105; 2.94±0.104; 3.06±0.093 and in Group V 2.68±0.060; 3.12±0.113; 3.29±0.069 respectively. The animals of Group I, II and IV revealed non-significant (P>0.01) increase in milk fat (%) on 7th and 14th days of treatment. However, the animals of Group III recorded significant (P<0.01) increase in milk fat (%) on day 7th and 14th of treatment. In animals of Group V, a highly significant (P<0.001) increase in milk fat (%) was observed on day 7th and 14th of treatment. The comparison of values of fat (%) between Groups, revealed significantly (P<0.01) lower milk fat (%) in the animals of Group II (2.73±0.071), III (2.71±0.066), IV (2.72±0.105) and V (2.68±0.060) as compared to Group I (3.68±0.087) throughout the observation period. Similarly, Comparison of values of fat (%) between Group III and IV as compared to Group II revealed non-significantly (P>0.01) lower milk fat (%) in the animals of Group III and IV on day 0 followed

by non-significantly ($P>0.01$) higher milk fat (%) on day 7th and 14th of treatment in Group III and IV. Comparison between Group II and V revealed non-significant ($P>0.01$) lower milk fat (%) in animals of Group V on day 0 which significantly ($P<0.01$) increased on day 7th and day 14th of treatment. Comparison between Group IV and V as compared to Group III indicated non-significantly ($P>0.01$) higher milk fat (%) on day 0 in animals of Group IV followed by non-significantly ($P>0.01$) higher milk fat (%) on day 7th and 14th of treatment in Group V. Similarly, comparison of the values of fat content between Group IV (2.72 ± 0.105) and V (2.68 ± 0.060) revealed non-significantly ($P>0.01$) lower milk fat (%) on day 0 in animals of Group V followed by non-significantly ($P>0.01$) higher milk fat (%) on day 7th (3.12 ± 0.113) and 14th (3.29 ± 0.069) of treatment in Group V.

A significant ($P<0.01$) decrease in fat (%) was recorded in the animals of all Groups having sub clinical mastitis on the day of screening. However, the milk fat percent value increased gradually on 7th and 14th day following the application of gel and paste on sub clinically affected udder. Reduction in milk fat percent in milk of SCM affected cow has also been reported by (Jones, 2006; Sonea *et al.*, 2009; Souza *et al.*, 2009; Hassan, 2013) and (Jagadeesh *et al.*, 2016). In contrast to the finding of our study and an increase in fat percent in dairy cow affected with SCM has been reported by (Bruckmaier *et al.*, 2004). The increase in milk fat percent might be attributed to gradual reduction of inflammatory process in udder of affected cow. The decrease in fat percent in affected cows might be due to increased activity of enzyme lipase which causes breakdown of milk fat and release of free fatty acid (Nagwa *et al.*, 2000) and (Uallah *et al.*, 2005).

SNF (Solid Not Fat) Content

Mean \pm SE value of milk SNF (%) in healthy and sub clinically infected cows are presented in Table 3.

Table 3: Changes in milk SNF % (Mean \pm S.E.) in sub clinical mastitis in cows

Groups	Days of Observation			P-value (with in Groups)
	0 Day	7 Day	14 Day	
Group-I	9.52 \pm 0.105 ^a	9.54 \pm 0.037 ^a	9.57 \pm 0.065 ^a	0.892
Group-II	7.21 \pm 0.083 ^b	7.45 \pm 0.125 ^b	7.58 \pm 0.069 ^b	0.051
Group-III	7.20 \pm 0.150 ^b	7.55 \pm 0.240 ^b	7.63 \pm 0.232 ^b	0.345
Group-IV	7.21 \pm 0.172 ^b	7.45 \pm 0.180 ^b	7.58 \pm 0.132 ^b	0.292
Group-V	7.18 \pm 0.090 ^{bC}	7.64 \pm 0.291 ^{bAB}	7.81 \pm 0.270 ^{bA}	0.046

^{ABC}Means with different superscripts within a row differ significantly; ^{abc} Means with different superscripts within a column differ significantly. Group I- healthy control; Group II- cows with SCM and treated with aloe vera gel; Group III- cows with SCM and treated with neem leaves paste; Group IV- cows with SCM and treated with Tulsi leaves paste; Group V- cows with SCM and treated with mixture of all herbs

On day 0 SNF % values in Group I, II, III, IV and Group V were 9.52 \pm 0.105, 7.21 \pm 0.083, 7.20 \pm 0.150, 7.21 \pm 0.172, 7.18 \pm 0.090 respectively. Similarly the SNF (%) values in Group I, II, III, IV and V on day 7th and 14th were 9.54 \pm 0.037 and 9.57 \pm 0.065; 7.45 \pm 0.125 and 7.58 \pm 0.069; 7.55 \pm 0.240 and 7.63 \pm 0.232;

7.45±0.180 and 7.58±0.132; 7.64±0.291 and 7.81±0.270 respectively. Comparison within the Group on day 0, 7th and 14th indicated that the animals of Group I, II, III and IV exhibited non-significance (P>0.01) increase in SNF % as compared to value of day 0 with that of the values recorded on 7th and 14th day after the treatment. However, the animals of Group V revealed significant (P<0.01) increase in SNF % as compared to base value on 7th and 14th day post treatment. On the day of screening significantly (P<0.01) higher SNF % in the healthy animals of Group I as compared to animals of Group II, III, IV and V throughout the observation period. Comparison between Group II with Group III and V revealed non-significantly (P>0.01) higher SNF % in animals of Group II on 0 day followed by non-significantly (P>0.01) lower SNF % in the animals of Group II on day 7th and 14th of observation periods. However, on comparison of SNF (%) between Group II and IV revealed non-significant (P>0.01) changes in value of SNF % throughout observation periods. Comparing value of SNF % between Group III and IV revealed non-significantly (P>0.01) higher SNF % in the animals of Group III throughout observation period. Comparing values of SNF % between Group III and V revealed non-significant (P>0.01) higher SNF % value in Group III as compared to Group V on day 0, while the values of SNF % were non-significantly (P>0.01) lower in the animals of Group III as compared to animals of Group V on day 7th and 14th day following the treatment. Comparison between Group IV and V revealed non-significantly (P>0.01) lower SNF % in animals of Group IV as compared to Group V throughout observation. The values of SNF % were significantly lower in Group II, III, IV and V with subclinical mastitis on day 0. When compared with the values of healthy normal cows of Group I.

However, the milk SNF % values increased gradually on 7th and 14th day following treatment. This observation is in accordance with that of (Singh *et al.* 2004) and (Hassan 2013) they found decrease in SNF contents of mastitic milk and its significant increase after treatment of topical application of herb in the form of paste or spray. The decrease in SNF percentage in cow with subclinical mastitis occurred due to invasion of pathogens to mammary tissue leading to decrease in synthesis activity of mammary glands (BenChedly *et al.*, 2009). Since lactose and protein are the major constituents of SNF so, it appears that drop in SNF mainly occurred due to significant decrease in lactose content of mastitic milk.

Protein Content

Table 4 depict the Mean±SE values of milk protein % in various Groups. The values of protein % in the cows of healthy Group I on day 0 was 3.51±0.037 followed by 3.52±0.068 on day 7th and 3.54±0.027 on day 14th. Similarly, obtained values of protein % in Group II animals were 2.76±0.108, 2.86±0.055 and 2.91±0.087 on 0, 7th and 14th day respectively. On day 0, 7th and 14th values of protein % of Group III were 2.73±0.071, 2.93±0.122 and 3.03±0.083 respectively. The respective values of protein % of Group IV on 0, 7th and 14th day were 2.74±0.126, 2.88±0.070 and 2.98±0.054. In Group V on day 0, 7th, and 14th obtained

values were 2.71 ± 0.073 , 2.96 ± 0.080 and 3.15 ± 0.080 respectively. The animals of Group I, II, III and IV revealed non-significant ($P > 0.01$) increase in protein % on 7th and 14th day of treatment. In the animals of Group V, milk protein % increased significantly ($P > 0.01$) on 7th and 14th day of treatment as compared to their base values. The values of protein % between Groups revealed significantly ($P < 0.01$) higher protein % in animals of Group I (healthy control) as compared to animals of Group II, III, IV and V throughout the observation period. Comparison between Group II with Group III and V revealed non-significant ($P > 0.01$) lower milk protein % in the animals of Group V before initiation of treatment (Day 0) followed by non-significantly ($P > 0.01$) higher values of milk protein % on 7th of day of observation.

Table 4: Changes in milk protein % (Mean \pm S.E.) in sub clinical mastitis in cows

Groups	Days of Observation			P-value (with in Group)
	0 Day	7 Day	14 Day	
Group-I	3.51 ± 0.037^a	3.52 ± 0.068^a	3.54 ± 0.027^a	0.885
Group-II	2.76 ± 0.108^b	2.86 ± 0.055^b	2.91 ± 0.087^c	0.437
Group-III	2.73 ± 0.071^b	2.93 ± 0.122^b	3.03 ± 0.083^{bc}	0.109
Group-IV	2.74 ± 0.126^b	2.88 ± 0.070^b	2.98 ± 0.054^c	0.192
Group-V	2.71 ± 0.073^{bB}	2.96 ± 0.080^{bA}	3.15 ± 0.080^{bA}	0.004

^{ABC}Means with different superscripts within a row differ significantly. ^{abc}Means with different superscripts within a column differ significantly. Group I- healthy control; Group II- cows with SCM and treated with aloe vera gel; Group III- cows with SCM and treated with neem leaves paste; Group IV- cows with SCM and treated with Tulsi leaves paste; Group V- cows with SCM and treated with mixture of all herbs

However, a significant ($P < 0.01$) increase in milk protein % was recorded on 14th day of observation. Comparison of protein % of value between Group III and IV non-significant ($P > 0.01$) lower milk protein % was recorded in the animals of Group III on day 0 followed by non-significant ($P > 0.01$) higher milk protein % values on day 7th and 14th after treatment. Comparison between Groups III and V revealed non-significant ($P > 0.01$) higher milk protein % in animals of Group III on day 0 followed by non-significant ($P > 0.01$) lower milk protein % in the animals of Group III on day 7th and 14th of observation period. While comparing the protein % of values between animals of Group IV and V a non-significant ($P > 0.01$) higher milk protein % in animals of Group IV was observed on day 0 followed by non-significant ($P > 0.01$) lower milk protein % in the animals of Group IV on day 7th and 14th of observation periods. The result of present study elucidated a significant ($P < 0.01$) decrease in milk protein % in the animals of all Groups with sub clinical mastitis on the day of screening. The post therapeutic Mean \pm SE milk protein % value increased gradually on 7th and 14th day. The reduction in protein percent in milk in the present study is in accordance with Jones 2006 and Hassan 2013. Contrary to the finding of this study increased protein percent in SCM affected cow has been reported by Sonea *et al.*, 2009. Khan & Khan 2006 opined that the decrease in protein content in milk during SCM might be due to high increase in activity of proteolytic enzyme (plasmin) which causes extensive destruction of milk protein in udder.

Lactose Content

Table 5 comprise the result of changes in lactose % in healthy and sub clinically affected cows. The values of milk lactose % of healthy animals of Group I (5.13 ± 0.067) was significantly higher on day 0 as compared to Group II (4.40 ± 0.105), Group III (4.38 ± 0.101) Group IV (4.39 ± 0.102) and Group V (4.38 ± 0.101). The animals of Group II, III and IV revealed non-significant ($P > 0.01$) increase in milk lactose % throughout the observation period except in Group I, where the milk lactose % value were non-significant by ($P > 0.01$) lower the base value on day 7th of observation period.

Table 5: Changes in milk lactose % (Mean \pm S.E.) in sub clinical mastitis in cows

Groups	Days of Observation			P-value (with in Group)
	0 Day	7 Day	14 Day	
Group-I	5.13 ± 0.067^a	5.11 ± 0.055^a	5.15 ± 0.068^a	0.899
Group-II	4.40 ± 0.105^b	4.42 ± 0.162^b	4.48 ± 0.107^c	0.91
Group-III	4.38 ± 0.101^b	4.45 ± 0.146^b	4.62 ± 0.141^{bc}	0.432
Group-IV	4.39 ± 0.102^b	4.41 ± 0.098^b	4.56 ± 0.095^{bc}	0.446
Group-V	4.38 ± 0.101^{bB}	4.60 ± 0.136^{bAB}	4.83 ± 0.090^{bA}	0.039

^{ABC}Means with different superscripts within a row differ significantly. ^{abc} Means with different superscripts within a column differ significantly. Group I- Healthy Control; Group II- Cows with SCM and treated with Aloe vera gel; Group III- Cows with SCM and treated with Neem leaves paste; Group IV- Cows with SCM and treated with Tulsi leaves paste; Group V- Cows with SCM and treated with Mixture of all herbs

However, in the animals of Group V, treated with a combination of herbal paste revealed non-significant increase in milk lactose % ($P > 0.01$) on day 7th as compared to the value of day 0 followed by significant ($P < 0.01$) increase on day 14 of observation period. Significantly ($P < 0.01$) higher milk lactose % were recorded in the animals of Group I as compared to Group II, III, IV and V throughout the observation period, when compared the values in between the Groups. A non-significant ($P > 0.01$) higher milk lactose % in the animals of Group as compared to Group III and V on day 0 and 14th day was recorded during the observation periods while milk lactose % values were non-significantly ($P > 0.01$) lower in the animals of Groups II as compared to Group III and V on day 7th of treatment. Comparison of the lactose % value between Groups revealed non-significant ($P > 0.01$) higher milk lactose % in the animals of Group II as compared to Group IV on day 0 and 7th followed by non-significantly ($P > 0.01$) lower milk lactose % in the animals of Group II on day 14th of treatment. The range of normal lactose % values in the cow milk has been reported by many workers. During the present study a significant ($P < 0.01$) decrease in milk lactose percent was determined in the animals of all Groups affected with sub clinical mastitis on the day 0 of screening. However, the milk lactose percent value increased gradually on 7th and 14th day following treatment. The present findings are in agreement with the findings of earlier workers who reported reduction in milk lactose percent in SCM affected cows (Riaz *et al.*, 2012). However, Hassan 2013 reported slightly decrease in lactose percent in cows affected with SCM. The decrease in lactose percent might be attributed

to less glucose being available due to decreased synthetic ability of the enzyme system in the secretory cells as a result of the mammary gland infection and destruction of epithelial cell by leucocytes (Coulon *et al.*, 2002). Verma *et al.* (2018) also concluded that to improve the compositional quality of milk, awareness at unorganized dairy farms is needed with regard to scientific feeding and management practices.

Conclusion

It was concluded that the milk compositions were affected with mastitis condition however, it can be prevented by therapeutic treatment by herbs. In present investigation and herbal mixture aloe vera, neem and tulsi paste was found the most efficacious combination in restoration of milk constituents and SCM.

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