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EFFECT OF MASTITIS ON MILK COMPOSITION OF CROSS-BRED (HOLSTEIN FRIESIAN X CHOLISTANI) AND SAHIWAL CATTLE

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ABSTRACT

The present study was designed to investigate the effect of mastitis on various components including fat percentage, protein, total solids, solid not fat, pH, specific gravity and lactose content in milk of cross-bred (Holstein Friesian × Cholistani) and Sahiwal cattle. A total of 376 milk samples (excluding blind and nonfunctional teats) from 100 cows (50 cross-bred and 50 sahiwal) were collected aseptically. Animals of different ages, parity and stages of lactation were selected under field condition. The diagnosis of subclinical mastitis was done on basis of Surf Field Mastitis Test. For bacterial examination of milk, the samples were grown in laboratory and various biochemical tests were performed. The results of the present study indicated that solid not fat (SNF) of mastitic sahiwal and cross-bred cattle and total solids percentage of normal and mastitic cattle were significantly reduced as the severity of mastitis increased in animals. The milk pH of normal and mastitic cattle including both sahiwal and cross-bred was significantly increased in severe cases of mastitis. The specific gravity of normal and mastitic milk samples showed that there was no significant difference in specific gravity as compared to normal. The protein percentage of normal and mastitic cattle including both sahiwal and cross-bred cattle indicated that protein concentration was significantly reduced in severe cases of mastitic animals. The findings of present study highlighted that mastitis cause major alterations in composition of milk leading to poor quality of the milk.

Keyword: cross-bred, mastitis, milk composition, sahiwal cattle

INTRODUCTION

Mastitis is inflammation of functional tissue of udder leading to tissue damage which ultimately results in loss of milk production. Mastitis may be apparent (clinical) or hidden/overt (sub-clinical) (Radostitis *et al.*, 2000). Mastitis also results in decreased quality of milk and risk for health of consumer (DeVleighter *et al.*, 2005).

According to National Mastitis Council Inc. of USA sub-clinical mastitis causes 70-80% loss of milk (Philpot and Nickerson, 1991). The major dairy animals of Pakistan are buffalo and cow share 95% in total milk production. However, mastitis is the most important disease of dairy animals (Khan and Khan, 2006).

Micro-organisms responsible for intra-mammary infection are *Staphylococcus aureus*, *Streptococcus agalactiae*, *Staphylococcus hyicus*, *Staphylococcus epidermidis*, bacillus spp., *Staphylococcus hominis*, *Escherichia coli*, *Staphylococcus xylosus*, *Streptococcus dysgalactiae,* corynebacterial spp. etc. (Ali *et al.*, 2008). *S. aureus* is of very high economic importance (Tollersrud *et al.*, 2000). *S. aureus* along with *Str. agalactiae* contribute 75% of total mastitis which is followed by environmental pathogens especially *E. coli* (Akinden *et al.*, 2001; Katsuda *et al.*, 2005). *S. aureus* is responsible for chronic infection while *E. coli* causes acute infection (Smith and Hogan, 1993). Infections due to staphylococcus cause reduction in milk production, decreases milk quality due to increased somatic cell count in milk and cause further bad effect on milk composition (Bilal and Ahmad, 2004). *S. aureus* has the ability to produce surface associated

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products and protein toxins which may be virulent (Haggar *et al.*, 2003; Harraghy *et al.*, 2003) and resultantly increase somatic cell count of milk.

Sub-clinical mastitis is easily diagnosed by a cheap and on spot field test i.e., Surf Field Mastitis Test (SFMT) (Muhammad *et al.*, 1995). The clinical mastitis is detectable due to the apparent changes in milk and appearance of udder. Discoloration, watery milk, clots in milk, swelling of udder etc. are symptoms of clinical form of mastitis. The processing of milk from mastitic infected dairy animal is also not economical as well as unhealthy (Muhammad *et al.*, 1995). Limited literature is available on effect of sub-clinical mastitis on milk composition in cross-bred and sahiwal cattle. The present study has been planned to investigate the effect of severity of mastitis on milk protein, fat, pH, specific gravity, lactose, sold not fat, total solids and bacteriology of milk in cross-bred and sahiwal cattle.

Objectives

- To observe the effect of mastitis on milk protein, fat, pH, specific gravity, lactose, sold not fat and total solids of milk.
- To observe the effect of mastitis on bacteriological examination of milk.

MATERIALS AND METHODS

Study animals

Out of 400 quarters from 100 cows (50 cross-bred and 50 sahiwal) 24 blind and non-functional teats were excluded and remaining 376 quarter milk samples were collected aseptically. The first few streaks were discarded and 150ml of milk samples were collected from each quarter. All the animals were inspected for clinical and subclinical mastitis. Animals in first week of lactation and those in the last two month of lactation were excluded.

Gross examination of mammary glands

Udder was examined visually for any abnormally, blind quarters, fibrosis, inflammation, atrophy, injury, tick infestation etc. and information was recorded.

Collection of milk samples

Mammary glands were washed with clean fresh water to remove dung material and the quarters were scrubbed with 70% alcohol solution and dried with clean cloth. Quarter milk samples were subjected to SFMT (Muhammad *et al.*, 1995) and processed further for determination of milk fat, protein, pH, specific gravity, lactose, sold not fat, total solids and bacteriology of milk. Each sample was labeled as LF (Left Front), LR (Left Rear), RF (Right Front) and RR (Right Rear).

Diagnosis of sub-clinical mastitis

SFMT was performed as described (Muhammad *et al.*, 1995) for diagnosis of sub-clinical mastitis.

Examination of milk

The collected milk samples were transported to laboratory of Haleeb Foods Limited, Lahore and processed for examination of fat, protein, SNF, total solids, pH using electronic pH meter, specific gravity (Davide, 1973), milk lactose (Egan *et al.,* 1981) and bacteriological examination of milk as described by NMC (2004).

Statistical analysis

The data was analyzed as described by Steel *et al.* (1997). The means were further analyzed by Duncan's Multiple Range as described by Montgomery (1997).

RESULTS AND DISCUSSION

In the present study 376 quarter milk samples (excluding blind and nonfunctional teats) from 100 cows (50 cross-bred and 50 sahiwal) were collected. Abnormalities of mammary glands are shown in (Table 1).

Table 1. Percentage of various abnormalities recorded in Sahiwal and cross-bred cattle

Quarter abnormalities	Cross-bred (%)	Sahiwal (%)
Blind	2	1
Fibrosis	3	1
Atrophied	1	2
Injured	2	1
Aberrations	3	2
Ticks	4	2

Recorded quarter abnormalities of mammary glands having blind, fibrosis, atrophied, injured, aberrations and ticks were 2, 3, 1, 2, 3 and 4% respectively, in cross-bred cattle. Recorded quarter abnormalities of mammary glands having blind, fibrosis, atrophied, injured, aberrations and ticks were 1, 1, 2, 1, 2 and 2% respectively, in sahiwal cattle

SFMT based prevalence of mastitis

Prevalence of sub-clinical mastitis bases on SFMT was conducted Out of 376 milk samples, 217 (57.71%) were found negative, the degree of mastitis as found in between positive and negative (Traces T) were 27 (7.18%), in first degree (P_1) of mastitis were74 (19.68%), in second degree (P_2) were 39 (10.37), and in 3rd degree of mastitis (P_3) were 19 (5.05%). Among the positive cases of SFMT maximum number of cases found in first degree were 74 (19.68%) out of 159 (42.29%) positive quarters. Our results of SFMT for detection of subclinical mastitis indicated the prevalence of 57.71% subclinical mastitis in sahiwal and cross-bred cattle under filed conditions (Table 2).

Table 2. SFMT based prevalence of mastitis

SFMT Score	No. of Quarters	Prevalence (%)
N	217	57.71
T	27	7.18
P_1	74	19.68
P_2	39	10.37
P_3	19	5.05
Total =	376	100

SFMT based quarter wise prevalence of mastitis

Quarter wise prevalence of sub-clinical mastitis based on SFMT was conducted. Out of 92 right rear (RR) quarters, quarters found in N, T, P_1, P_2 and P_3 were 53, 7, 16, 11 and 5 respectively. Out of 95 right front (RF) quarters, quarters found in N, T, P_1, P_2 and P_3 were 57, 6, 21, 7 and 4 respectively. Out of 101 left rear (LR) quarters, quarters found in N, T, P_1, P_2 and P_3 were 58, 9, 18, 8 and 8 respectively. Out of 88 left front (LF) quarters, quarters found in N, T, P_1, P_2 and P_3 were 49, 5, 19, 13 and 2 respectively (Table 3).

Table 3. SFMT based quarter wise prevalence of mastitis

SFMT	RR	RF	LR	LF	Total
Normal	53	57	58	49	217
T	07	06	09	05	27
P_1	16	21	18	19	74
P_2	11	07	08	13	39
P_3	05	04	08	02	19
Total =	92	95	101	88	376

SFMT based result of subclinical mastitis indicated overall 57.71% prevalence in sahiwal and cross-bred cattle under field condition. Our results coincide with the results of previous studies. Fazal-ur-Rehman (1995) found quarter wise prevalence of 64% subclinical mastitis in dairy animals. Bachaya *et al.* (2005) determined the prevalence of 58.75% of subclinical mastitis in lactating animals on the basis of SFMT. Pal *et al.* (1989) determined that subclinical mastitis was 23.08% in cattle on the basis of bacteriological examination.

Fat (%) of normal and mastitic cattle at various scores of SFMT

The mean fat percentage of normal and mastitic cattle is represented in (Table 4). The results showed that fat concentration was significantly reduced as the severity of mastitis increased in animals. Milk fat % of normal and mastitic sahiwal and cross-bred cattle is shown in (Table 4). There was non-significant difference between control and trace group. Results indicated that no significant difference between P_1, P_2 and P_3. However fat contents were significantly reduced in P_3 followed by P_2 and P_1 (Table 4).

Results indicated that fat percentage was significantly reduced as severity of mastitis increased in animals. There was non-significant difference among the normal and traces group. The findings of our study indicated that, there was significant decrease in fat percentage between strongly positive (P_3) mastitic cross-bred and sahiwal cattle. Similarly, Wielgosz-groth and Groth (2003) also indicated that fat percentage was significantly reduced with severity of mastitis. In our study maximum milk fat was recorded in N grade quarters while minimum milk fat was found in P_3 grade quarters. Previously researchers also reported decrease in milk fat with severity of mastitis Abdel-Galil and Nassib (1980) observed decrease in fat percentage in mastitic quarters. Mandal and Raheja (1985) reported decrease in percentage of milk fat from 5.18 to 5.06% with increasing mastitis severity. Shahin and Haggag (1987) reported 7.28% of milk fat in healthy buffaloes while 4.02% in mastitic buffaloes. Mahran *et al.* (1992) reported 18% decreased fat in mastitic quarters as compared to healthy quarters. Hortet *et al.* (1998) reported reduced milk fat due to mastitis in dairy cows. The decrease in milk fat may be due to malfunction of udder parenchyma in mastitis infection.

In our study highest milk protein percentage was found in negative group and the minimum in SFMT P_3 grade mastitic quarters. Mert *et al.* (1992) had reported decreased milk protein percentage from 0.92 to 0.82 gm in mastitis. Researchers also documented decreased milk protein in mastitis (Hortet *et al.*, 1998; Urech *et al.*, 1998; Rawdat and Omaima, 2000) in mastitis infection. The decrease in milk protein percentage in mastitis cases is due to malfunction of udder parenchyma in mastitis infection as a result the protein synthesis is reduced in mastitis cases.

The results indicated that SNF was significantly reduced as the severity of mastitic increased in animals. There was non-significant difference between P_1, P_2 and P_3. However solid not fat contents were significantly reduced in P_3 followed by P_2 and P_1. There was non-significant difference of mean value of solid not fat concentration among the normal and traces group. The findings of our study also indicated that, there was significant decrease among the mean value of solid not fat concentration in strongly positive (P_3) mastitic cross-bred and sahiwal cattle. The results indicated that milk solid not fat of mastitic sahiwal and cross-bred cattle was significantly reduced as the severity of mastitic increased. Similarly, researchers also found a net decrease in SNF with mastitis (Wielosz-groth and Groth 2003; Rowland *et al*., 1959). The decrease in SNF percentage in mastitis cases is due to malfunction of udder functional tissue in mastitis infection as a result the SNF percentage is reduced in mastitis cases.

Table 4. Mean ± SE of milk fat (%) of normal and mastitic Sahiwal and cross-bred cattle

Group	Sahiwal cattle					Cross-bred cattle				
	RR	FR	LR	LF	Mean	RR	FR	LR	LF	Mean
Control	5.03±0.03	4.99±0.08	5.03±0.05	4.87±0.03	4.98±0.03^{a}	4.34±0.06	4.17±0.04	4.17±0.04	4.06±0.01	4.18±0.03^{d}
Traces	4.94±0.03	4.85±0.07	4.85±0.05	4.80±0.02	4.86±0.02^{b}	4.24±0.02	4.11±0.01	4.10±0.05	3.95±0.02	4.10±0.03^{de}
P_1	4.73±0.03	4.75±0.03	4.75±0.04	4.73±0.03	4.74±0.02^{c}	4.02±0.01	4.06±0.02	4.02±0.03	4.01±0.03	4.03±0.01^{e}
P_2	3.69±0.05	3.67±0.05	3.67±0.04	3.73±0.03	3.69±0.02^{f}	3.45±0.03	3.41±0.02	3.51±0.02	3.44±0.02	3.45±0.01^{g}
P_3	3.16±0.02	3.16±0.01	3.08±0.01	3.27±0.13	3.17±0.03^{h}	2.82±0.04	2.70±0.05	2.74±0.02	2.68±0.01	2.74±0.02^{i}
Overall Mean	4.31±0.17^{a}	4.29±0.17^{a}	4.27±0.18^{a}	4.28±0.15^{a}	4.29±0.08^{a}	3.77±0.13^{b}	3.69±0.13^{bc}	3.71±0.12^{c}	3.63±0.12^{c}	3.70±0.06^{c}

Values with different superscripts differ significantly (P<0.001).

Table 5. Mean ± SE of milk protein (%) of normal and mastitic Sahiwal and cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	FR	LR	LF	Mean
Control	3.78±0.04	3.70±0.05	3.68±0.04	3.67±0.01	3.71±0.02^{a}	3.61±0.01	3.54±0.02	3.53±0.03	3.49±0.04	3.54±0.02^{c}
Traces	3.73±0.03	3.64±0.06	3.60±0.02	3.62±0.02	3.64±0.02^{ab}	3.55±0.01	3.48±0.02	3.47±0.03	3.44±0.04	3.48±0.02^{cd}
P_1	3.61±0.02	3.68±0.05	3.57±0.03	3.65±0.07	3.63±0.02^{b}	3.43±0.02	3.43±0.02	3.43±0.02	3.37±0.01	3.41±0.01^{d}
P_2	3.21±0.01	3.25±0.02	3.23±0.02	3.24±0.03	3.23±0.01^{e}	3.16±0.02	3.13±0.01	3.51±0.03	3.11±0.01	3.14±0.01^{f}
P_3	2.83±0.03	2.75±0.02	2.72±0.01	2.60±0.05	2.72±0.03^{g}	2.45±0.01	2.3±0.03	2.27±0.01	2.24±0.01	2.33±0.02^{h}
Overall Mean	3.43±0.08	3.40±0.09	3.36±0.08	3.35±0.10	3.39±0.04^{a}	3.24±0.10	3.19±0.09	3.17±0.11	3.13±0.11	3.18±0.05^{b}

Values with different superscripts differ significantly (P<0.001).

Table 6. Mean ± SE milk solids not fat (%) of normal and mastitic Sahiwal and cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	FR	LR	LF	Mean
Control	8.58±0.11^{a}	8.58±0.03^{a}	8.63±0.03^{a}	8.63±0.03^{a}	8.62±0.03	8.46±0.02^{bc}	8.46±0.04^{bc}	8.42±0.01^{bc}	8.51±0.04^{ab}	8.46±0.02
Traces	8.53±0.10^{ab}	8.54±0.02^{ab}	8.55±0.04^{ab}	8.51±0.04^{ab}	8.53±0.03^{ab}	8.40±0.02^{bc}	8.39±0.02^{bc}	8.34±0.02^{bc}	8.45±0.05^{bc}	8.40±0.02
P_1	8.50+0.05^{bc}	8.46+0.02^{bc}	8.42+0.01^{bc}	8.51+0.01^{bc}	8.47+0.01	8.20+0.02^{c}	8.18+0.01^{c}	8.25+0.03^{c}	8.14+0.02^{d}	8.19+0.01
P_2	7.03±0.02^{e}	7.04±0.01^{e}	7.86±0.04^{d}	7.80+0.08^{d}	7.43+0.10	7.50+0.08^{de}	7.55+0.05^{de}	7.52+0.06^{de}	6.76+0.08^{f}	7.33+0.02
P_3	5.65±0.06^{g}	5.61±0.03^{g}	5.61±0.04^{g}	5.46±0.01^{gh}	5.58±0.03	5.29±0.02^{h}	5.23±0.02^{h}	5.26±0.02^{h}	5.23±0.01^{h}	5.25±0.01
Overall Mean	7.68±0.27^{bc}	7.66±0.27^{bc}	7.81±0.26^{a}	7.78±0.27^{ab}	7.73±0.13^{a}	7.57±0.27	7.56±0.28^{bc}	7.56±0.27^{bc}	7.42±0.03^{c}	7.53±0.14^{b}

Values with different superscripts differ significantly (P <0.001).

Table 7. Mean ± SE milk Total Solids of normal and mastitic Sahiwal and Cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	FR	LR	LF	Mean
Control	13.54±0.13	13.58±0.05	13.33±0.33	13.23±0.52	13.42±0.15	12.96±0.10	12.98±0.16	13.13±0.22	13.18±0.08	13.0±0.07
Traces	13.48±0.14	13.51±0.04	13.23±0.32	13.15±0.52	13.34±0.15	12.85±0.11	12.81±0.07	13.03±0.21	13.11±0.09	12.95±0.07
P_1	13.05±0.06	12.65±0.06	12.60±0.02	12.65±0.04	12.74±0.05	12.40±0.04	12.40±0.05	12.71±0.24	12.89±0.03	12.60±0.08
P_2	10.95±0.06	10.80±0.04	10.79±0.03	10.72±0.16	10.81±0.03	10.35±0.09	10.30±0.09	10.38±0.03	10.38±0.02	10.35±0.03
P_3	10.09±0.05	9.48±0.18	9.63±0.01	9.49±0.06	9.67±0.08	8.24±0.03	8.31±0.20	8.65±0.06	8.35±0.06	8.39±0.06
Mean	12.22±0.33^{a}	12.00±0.37^{ab}	11.91±0.34^{ab}	11.85±0.36^{bc}	11.99±0.17^{a}	11.36±0.42^{d}	11.36±0.42^{d}	11.58±0.41^{cd}	11.58±0.44^{cd}	11.47±0.21^{b}

Values with different superscripts differ significantly (P<0.001).

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Table 8. Mean ± SE milk pH of normal and mastitic Sahiwal and Cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	RF	LR	LF	Mean
Control	6.69±0.06^{h}	6.65±0.02^{h}	6.67±0.03^{h}	6.43±0.09	6.61±0.04^{de}	6.59±0.04^{j}	6.50±0.05^{j}	6.57±0.05^{h}	6.64±0.06^{h}	6.57±0.03^{g}
Traces	6.71±0.01^{g}	6.70±0.01^{g}	6.71±0.01^{g}	6.70±0.01^{g}	6.70±0.004^{de}	6.70±0.01^{g}	6.71±0.01^{g}	6.68±0.004^{g}	6.83±0.05^{ef}	6.73±0.02^{cd}
P_1	6.76±0.04^{ef}	6.78±0.02^{ef}	6.78±0.01^{ef}	6.80±0.01^{ef}	6.78±0.01^{c}	6.83±0.03^{ef}	6.81±0.04^{ef}	6.65±0.06^{f}	6.63±0.05^{f}	6.73±0.03^{cd}
P_2	7.05±0.07^{de}	7.25±0.06^{c}	745±0.06^{g}	743±0.09^{g}	7.29±0.05^{a}	6.94±0.04^{c}	7.06±0.01^{d}	7.39±0.20^{ab}	7.14±0.06^{cd}	7.12±0.04^{g}
P_3	7.24±0.08^{c}	7.35±0.03^{bc}	7.35±0.06^{bc}	7.30±0.07^{bc}	7.31±0.03^{a}	7.33±0.11b^{c}	7.40±0.09^{g}	7.25±0.05^{c}	7.35±0.03^{bc}	7.33±0.04^{a}
Overall Mean	6.89±0.06	6.94±0.07	6.99±0.08	6.93±0.09	6.94±0.04^{b}	6.88±0.06	6.89±0.07	6.90±0.08	6.91±0.07	6.90±0.04^{a}

Values with different superscripts differ significantly (P<0.001).

Table 9. Mean ± SE milk specific gravity of normal and mastitic Sahiwal and cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	RF	LR	LF	Mean
Control	1.028±0.001	1.027±0.002	1.028±0.001	1.029±0.001	1.028±0.002	1.030±0.001	1.030±0.001	1.030±0.002	1.028±0.001	1.029±0.002
Traces	1.020±0.001	1.020±0.001	1.020±0.001	1.20±0.001	1.20±0.001	1.021±0.001	1.021±0.001	1.021±0.001	1.021±0.001	1.021±0.021
P_1	1.027±0.001	1.027±0.002	1.028±0.001	1.027±0.001	1.027±0.001	1.028±0.02	1.029±0.001	1.028±0.001	11.028±0.001	1.028±0.001
P_2	1.027±0.001	1.025±0.001	1.027±0.001	1.026±0.001	1.026±0.001	1.026±0.001	1.026±0.001	1.027±0.001	1.027±0.001	1.026±0.001
P_3	1.024±0.001	1.024±0.001	1.025±0.001	1.024±0.001	1.024±10.001	1.025±0.001	1.025±0.001	1.025±0.001	1.025±0.001	1.025±0.001
Overall Mean	1.025±0.001	1.025±0.001	1.025±0.001	1.025±0.001	1.025±0.001	1.026±0001	1.026±0001	1.026±0001	1.026±0001	1.026±0001

Values with different superscripts differ significantly (*P*<0.001).

Table 10. Mean ± SE milk Lactose of normal and mastitic Sahiwal and Cross-bred cattle

Group	Sahiwal					Cross-bred				
	RR	FR	LR	LF	Mean	RR	RF	LR	LF	Mean
Control	4.68±0.05	4.66±0.06	4.80±0.04	4.83±0.01	$4.74±0.03^{A}$	4.64±0.11	4.84±0.04	4.85±0.03	4.57±0.03	$4.73±0.04^{A}$
Traces	4.3±0.05	4.60±0.05	4.76±0.03	4.80±0.02	$4.70±0.03^{a}$	4.70±0.02	4.81±0.03	4.79±0.03	4.52±0.02	$4.70±0.03^{a}$
P_1	4.61±0.02	4.63±0.03	4.55±0.05	4.60±0.04	$4.60±0.02^{b}$	4.37±0.07	4.25±0.06	4.65±0.07	4.70±0.03	$4.71±0.06^{a}$
P_2	4.12±0.07	4.19±0.08	4.17±0.01	4.18±0.03	$4.16±0.03^{c}$	4.04±0.02	4.04±0.01	4.04±0.02	4.20±0.01	$4.03±0.01^{d}$
P_3	3.68±0.05	3.50±0.05	3.16±0.03	3.12±0.03	$3.67±0.06^{f}$	2.86±0.05	2.82±0.03	2.85±0.02	2.81±0.03	$2.83±0.02^{e}$
Overall Mean	$4.34±0.09^{a}$	$4.32±0.01^{ab}$	$4.29±0.14^{ab}$	$4.31±0.15^{ab}$	$4.31±0.06^{b}$	$4.12±0.16^{d}$	$4.15±0.17^{cd}$	$4.23±0.17^{bc}$	$4.12±0.16^{d}$	$4.16±0.08^{a}$

Values with different superscripts differ significantly (*P*<0.001).

The results about total solids indicated that total solids concentration was significantly reduced as the severity of mastitis increased in animals. There was non-significant difference between P_1, P_2 and P_3. However total solids contents were significantly reduced with severity of mastitis. Similar results are also reported by researchers (Wielosz-groth and Groth, 2003; Rowland *et al*, 1959). The results showed that pH value was significantly increased as the severity of mastitis increased in animals. There was non-significant difference between control and traces, and results also indicated that no significant difference found between P_1, P_2 and P_3. However, pH value was significantly increased in P_3 followed by P_2 and P_1. The milk pH of normal and mastitic cattle including both sahiwal and cross-bred was significantly increased in severe cases of mastitic animals. These values are similar to the results of previous researchers (Haggag *et al.*, 1991; Wielgosz-groth and Gorth, 2003; Batavani *et al.,* 2007) who also reported increased pH in mastitis cases. The normal milk has pH towards acidic trend but in mastitis the pH increases and tends towards basicity. This increase in pH may be due to abnormality in udder function, more influx of salts and free radicals (Na^{+}, K^{+}, Cl^{-}, etc.) in udder during mastitis infection.

The results indicated that, there was non-significant difference between control and traces. Result also indicated that no significant difference found between P_1, P_2 and P_3. In our study on significant difference of milk specific gravity was found at different grades of sub-clinical mastitis. In contrast to our results the previous studies reported that the specific gravity of milk reduced in severe cases of mastitis (Horvath *et al.*, 1980; Haggeg *et al.*, 1991 and Charjan *et al.*, 2000). Mean lactose contents were calculated with respect to severity of mastitis based on score of SFMT. Mean lactose content found in control case i.e., Surf Field Mastitis Test was negative (4.74%), mean lactose content for T, P_1, P_2 and P_3 was 4.70, 4.60, 4.16 and 2.83%, respectively. Previous study also found that there was significant decrease in lactose contents with severity of mastitis. Reichmuth (1975) and Kitchen (1987) observed that mastitis result in tissue damage leading to the decrease synthesis of lactose. Other researchers also described that the lactose decreased during mastitis (Bering and Shook, 1992; Hamann and Kromker 1997; Rawdat and Omaima 2000; Coulon *et al.,* 2002; Pyorala 2003; Silva *et al.,* 2000). The decrease in milk lactose percentage in mastitis cases is due to malfunction of udder functional tissue in mastitis infection as a result the synthesis of milk lactose in udder is reduced in mastitis cases.

Table 11. Microbiological examination of milk, identification and percentage of bacteria

Sr. No.	Name of Bacteria	Number	Percentage (%)
1.	*S. aureus*	66	41.5
2.	*Str. agalactae*	43	27
3.	Bacillus spp.	02	1.2
4.	*E. coli*	08	05
5.	Corynebacterium	05	3.14
6.	Mix	35	22
Total =		159	100

Microbiological examination of milk was done for identification of bacteria in T, P_1, P_2 and P_3 cases of SFMT Score. Out of total 159 quarter milk samples the *S. aureus w*as found in 66 cases (41.5%), *Str. agalactae* in 43 cases (27%), bacillus spp. was found in 2 cases (1.2 %), *E. coli* was found in 8 cases (5%), Corynebacterium was found in 5 cases (3.14%) and Mixed infection was found in 35 cases (22%) as shown in (Table 11). *S. aureus* is one of the most pathogenic bacteria in mastitis and can invade in all types of cells in udder and is difficult to control. Due to intracellular localization in epithelial cells the response of *S. aureus* to antibiotic treatment is low and long lasting and persistent infection occurs (Dhakal *et al.*, 2007).

CONCLUSION

It was concluded that mastitis altered the milk composition of both cross-bred (Holstein Friesian × Cholistani) and Sahiwal cattle resultantly fat, solid not fat, total solids, lactose and protein percentage decreased, while pH increased, however specific remained un-changed.

AUTHOR'S CONTRIBUTION

T. Ahmad: Conceived and designed the experiments.
A. Ghafoor: Performed the experiments.
M. Nadeem: Contributed materials/ tools.
M. Q. Bilal: Analysed the data.
A. Sharif: Wrote the paper.

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