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Effect of subclinical mastitis in compositional change in milk and blood parameter of crossbred dairy cow

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Abstract
This study was designed to evaluate the haemato-biochemical alterations in mastitis cow and normal cow along with compositional change in milk. Overall 53.33% incidence of mastitis on the basis of clinical signs, palpation in udder, CMT test and SCC with 81.25% subclinical mastitis and 18.75% clinical mastitis. Milk composition in mastitis cow Significant (p<0.01) increase in pH, Na, Whey Protein and electrical conductivity and a significant (p<0.01) decrease in specific Gravity, Fat, acidity, total protein and SNF value observed mastitis milk. Hematology performance in mastitis cows significant (p<0.01) observed increase in AST, Na^+, Total Protein and K^+ concentration and significant decrease (p<0.01) in TEC, Hb, PCV, Nutrophilia and Lymphopaenia.

Keywords: Subclinical mastitis, CMT, SCC, udder etc

Introduction
Mastitis (inflammation of udder) is one of the most devastating disease conditions leading to significant economic losses globally (Kumar *et al*., 2010, AbdEllah, 2013) [12, 2] because of reduced milk production, increased labor, treatment costs, milk with holding following treatment, death and premature culling (Miller *et al*., 1993, Szweda *et al*., 2014) [13, 18]. India is the largest milk producer in the world with a production of 176.3 million tonnes of milk per annum and per capita availability of milk in India is 375g/day in 2017-18 (NDDB 2017-18) as against the recommended level of 280 g/per capita per day by (ICMR). Milk is one of the most important foods of human beings. It is universally recognized as a complete diet due to its essential components (Battaglia, 2007; Javaid *et al*. 2009) [7, 9]. Liquid milk demand increased tremendous worldwide in recent year due to population growth. (Klaas, 2000) [11]. The mastitis was first reported in India by land (1926). The mastitis affects the productivity of milk up to 30% and production up to 15% mastitis significantly decreased the production and alters the milk composition. Several species of bacteria are able to invade the mammary gland, produce harmful substance that result is an inflammatory response. Signs of clinical mastitis include alterations in milk composition, decreased milk production and appearance, elevated body temperature and swelling, heat or redness in infected quarters. The main disease responsible for affecting productivity of milk is mastitis up to 30% reduction in productivity and 15% loss of its production. Subclinical mastitis was found more important in India (varying from 10-50% in cows and 5-20% in buffaloes) than clinical mastitis (1-10%) Joshi *et al*. 2006 [10].

Material and methods
Screening of cow
The present investigation was carried out in the department of Animal Husbandry and Dairying and outdoor Veterinary hospital of Chandra Shekhar Azad University of Agriculture & Technology, Kanpur. Different Physical, Chemical and Clinical test were employed to screen out the cows suffering from mastitis. After screen- out the cows affected from the mastitis disease was 80 out of 150 cows. The cows were divided in to two groups sub-clinical and clinical mastitis on the bases of somatic cell count (SCC) and California mastitis test (CMT) score and also signs of inflammation of udder and abnormality in milk composition.

Collection of Milk Sample for Laboratory Test

The udder was cleaned with a cloth/duster and dried prior to the collection of milk sample. Teat surface was swabbed with 70% alcohol or Dettol solution. The milk sample of each quarter from the same cow and bulk samples was collected in sterile McCartney bottles. First few stripping of milk work always avoided and discarded. The collected milk samples were examined within an hour's of collection.

California Mastitis Test

The test was done as per the standard procedures described by Pandit and Mehta (1969) [14]. Testing individual quarter sample requires use of a plastic paddle having four shallow cups marked A, B, C and D.

About 3 ml of milk from each quarter was drawn in to one of the four shallow cups to the paddle and equal volume of test reagent was added to the milk sample. The milk and reagent was mixed by gentle circular rotation of paddle held horizontally

The results are calibrated as under

- Trace Slight precipitate formed which dissolved with mixing.
- 1^+ Slimy gel formed.
- 2^+ Gel becomes thick and flocculent.
- 3^+ Gel becomes viscous and tacky.

Somatic cell count

The Somatic cell count of milk was performed by the method as described by Schalm *et al.,* (1971) [16]. The reactions occurring in CMT have been correlated with cell counts as follows.

- 1^+ 400,000 – 1500,000 cells/ml
- 2^+ 800,000 – 5000,000 cells/ml
- 3^+ > 500,000 cells/ml

Composition change in milk

Analysis of physico-chemical characteristics of healthy cow and suffering from mastitis disease cows were done by on the basis of these method, Moisture (AOAC, 1995) [3], Fat (AOAC, 1995) [3], Lactose (Nielssen, 1994), Ash (AOAC, 2005) [5], SNF (AOAC, 2005) [5], Acidity (AOAC, 2000) [4] and pH Beckman pH meter – Schmar and Company), specific gravity (AOAC, 2000) [4]. Nitrogen content (*N*) in the milk samples was estimated by the Kjeldahl (1983)'s method and crude protein content was calculated as *N*×6.25, Electrical conductivity (meter Draminski® Electronics in Agriculture) was used to measure the electrical conductivity of milk.

Statistical analyses of data were done by one-way ANOVA with SPSS 20 package software.

Result and Discussion

After screen-out 53.33% cow suffering from mastitis disease on basis of clinical signs, California mastitis test and somatic cell count. Sub clinical mastitis considered on 2.5-4.0lakhs/ml SCC without any abnormality of udder and more than 4lakhs/ml of SCC with abnormality of udder considered as clinical mastitis and below 2.5 lakhs/ml obvious changes in milk. Out of 80 cows screened positive for mastitis, 65 cows (81.25%) were of subclinical mastitis and only 15 cows (18.75%) were of clinical mastitis.

Table 1: Mean ± SE of compositional change in normal milk and mastitis milk.

Parameters	Normal Milk	Mastitis Milk
pH	6.5±0.03^b	6.9±0.1^a
Fat	4.01±0.07^a	3.2±0.45^b
Acidity	0.12±0.010^a	0.0791±0.004^b
Specific gravity	1.029±0.001^a	1.026±0.002^b
Solid not fat	8.28±0.06^a	7.58±0.08^b
Protein	3.58±0.007^a	3.51±0.003^b
Electrical Conductivity	13.38±.04^b	14.32±0.13^a
Na	466.4±23.14^b	810.4±11.27^a
Whey Protein	0.71±.07^b	1.24±0.04^a
Ash	0.68±0.07^a	0.63±0.04^b
Lactose	4.53±0.24^a	4.21±0.17^b

^{a,b} Mean with different superscripts in a row differ significantly (P<0.01)

Table 2: Mean ± SE of Hematological attributes in cows affected with mastitis and normal cow.

Attributes	Normal	Mastitis
TLC	7061±180.5^b	10720±160.8^a
TEC	5.60±0.14^a	4.77±0.10^b
Hb (gm/dl)	10.64±0.41^a	8.9±0.17^b
MCV (fl)	58.47±0.14^a	57.52±0.17^b
PCV (%)	32.2±0.62^a	26.9±0.21^b
MCHC (g/dl)	32.85±0.01^a	32.52±0.07^b
Lymphocyte (%)	60.84±1.72^a	45.32±0.74^b
Monocyte (%)	1.50±0.18^b	1.51±0.07^a
Eosinophils (%)	1.68±0.28^b	3.21±0.25^a
Neutrophils (%)	36.14±1.52^b	49.86±0.71^a

^{a,b} Mean with different superscripts in a row differ significantly (P<0.01)

Table 3: Mean ± SE of serum biochemical alterations in cows with mastitis and apparently healthy cows reared in natural field conditions.

Attributes	Normal	Mastitis
Protein	8.01±.06[b]	9.56±0.05[a]
AST	89.74±1.32[b]	107.1±1.62[a]
ALT	10.65±0.04[a]	10.35±0.02[b]
Glucose	59.04±0.14[a]	57.41±0.10[b]
P	5.8±0.12[a]	5.01±0.01[b]
Ca	10.80±0.12[a]	8.08±0.07[b]
Na	138.24±0.92[b]	151.4±1.07[a]
K	4.01±0.01[b]	5.18±0.08[a]

[a,b] Mean with different superscripts in a row differ significantly (P<0.01)

Compositional change in milk

In table no. 1 data of compositional change in milk showed a significant (p<0.01) increase in Whey Protein, Na, pH, Electric Conductivity in mastitis milk compared with normal milk and significant decrease (p<0.01) in Specific gravity, SNF, Fat and Protein value in mastitis milk. These results were also found in cow as reported by Badran *et al*., (1986) [6] and Haggag *et al*., (1991) [8]. Decrease in the specific gravity in mastitis affected cows attributed to the increase in chloride and decrease in the lactose contents. The study also reported that there is a positive correlation with the severity along with pH of the milk in mastitis. The decreasing trend in the specific gravity in mastitis milk owes to the reduced lactose contents and increased chlorides.

Hematological result

Table no. 2 presented various parameters of hematological in mastitis suffering cow and compared with healthy cow. There was a significantly (p<0.01) increase in TLC in mastitis suffering cow compared with healthy cow and significantly (p<0.01) decrease in HB, TEC, PCV and MCV. Different type of leucocyte count (DLC) that is Neutrophil, Eosinophil and Monocyte significantly (p<0.01) increase in mastitis suffering cow compared with healthy cow. The Same result also obtained by Sarvesha *et al*., (2017) [15].

Serum biochemical result

Table no. 3 showed a significantly (p<0.01) increase in various serum biochemical attributes like AST, Na^+, Protein and K^+ concentrations and a significantly decrease (p<0.01) attributes like Ca^+, Glucose and P^+ concentration in mastitis suffering cow compared with healthy cow. Kumar (2012) and Jain *et al*., (2013) found that non-significant decrease in total protein and calcium and only obvious change found in serum phosphorus value in mastitis suffering cow compared with healthy cow. Decreased albumin levels may be caused by increase in total protein may be after the immune response to the udder infection (Singh, 2000) [17].

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