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Debiprasanna Das
Department of Veterinary Pathology, CVSc & AH, Bhubaneswar, Odisha, India

Susen Kumar Panda
Deptt. Of Vety Physiology, CVSc & AH, Bhubaneswar, Odisha, India

Akshaya Kumar Kundu
Deptt. Of Vety Physiology, CVSc & AH, Bhubaneswar, Odisha, India

Basanti Jena
Deptt. Of ARGO, CVSc & AH, Bhubaneswar, Odisha, India

Bhabesh Chandra Das
Deptt. Of Vety. Extension, CVSc & AH, Bhubaneswar, Odisha, India

Rajesh Kumar Sahu
Department of Veterinary Public Health & Epidemiology, CVSc, Rajendranagar, Hyderabad, Telangana, India

Correspondence
Debiprasanna Das
Department of Veterinary Pathology, CVSc & AH, Bhubaneswar, Odisha, India

Haematological and metabolic profile test of mastitis affected bovines in Odisha

Debiprasanna Das, Susen Kumar Panda, Akshaya Kumar Kundu, Basanti Jena, Bhabesh Chandra Das and Rajesh Kumar Sahu

Abstract
Present study was undertaken to evaluate the haemato- biochemical parameters of cattle affected with mastitis. Blood samples were collected from jugular vein of 20 apparently healthy, 60 subclinical mastitis and 40 clinical mastitis affected cattle. Blood samples were analyzed for Hb, PCV, TEC, TLC, ESR and DLC, serum samples were analyzed for ALT, AST, Ca, Cl, K, Mg, Na, P and TP. There was an increase of Hb, PCV, TEC, lymphocyte, monocyte and ESR in SCM compared to CM, whereas in CM there was increase of TLC and granulocyte compared to SCM. Higher values of ALT and TP were observed in CM cases, whereas AST and Ca values were higher in SCM cases. Haemato-biochemical parameters can be used as important indicators for pathological state of mastitis animals.

Keywords: Mastitis, haematology, TLC, Neutrophil, Leucocyte

1. Introduction
Mastitis is multi factorial and a costly problem which affects all milk producing ruminants in India. It reduces milk yield, alters its composition and serious increase of leucocyte counts in milk. The magnitude of these changes in individual animal varies with the severity, duration of the infection and the causative microorganisms [1]. High yielding milch animals are more prone to mastitis especially exotic and cross breed animals. Cross breeding for better milk production, insanitary condition of cattle shed, improper management status of dairy equipments, unhygienic conditions of milkers and bosy condition of animals are the main cause for mastitis [2]. Clinical mastitis where abnormal milk is readily detected and sub-clinical mastitis where no change in the milk is apparent but in both the cases there is reduction of milk production. The reduction in milk production attributed to sub-clinical mastitis may account for 70%-80% of the total losses [3]. In mastitis, there is a break in the blood-milk-barrier along with impaired synthesis and secretary activity of udder epithelial cells, which alters the level of most components. Keeping in view of the above facts present study was undertaken to evaluate the haemato- biochemical parameters of cattle affected with mastitis.

2. Materials and methods
Blood samples were collected from 20 apparently healthy, 60 subclinical and 40 clinical mastitis affected cattle by professional Veterinary clinicians without harming the animal and as per the established guidelines. 10 ml of blood was collected aseptically from jugular vein of animals. 2 ml of blood was kept in an anticoagulant (EDTA) treated vial for haematological examination on the same day of collection. The remaining 8ml was used to harvest serum for biochemical estimations. Blood samples were transported to the laboratory within one hour in a thermo flask with ice & then fresh blood was analyzed for Hb, PCV, TEC, TLC, ESR and DLC were estimated as per method described by Schalm (1965)[4]. The serum separated from blood was analyzed for ALT, AST, Ca, Cl, K, Mg, Na, P and TP as per the Modified IFCC method as per the procedure described by Burtis and Ashwood (1999) [5], using the diagnostic kits supplied by M/s Crest Biosystem™, a division of Coral clinical systems, Goa. The data obtained from analysis were analyzed through one way analysis of variance (ANOVA).

3. Results and discussion
3.1 Haematological parameters
The haematological profile in clinical and subclinical mastitis affected cows in the present study has been represented in Table-1.

The mean values of haemoglobin (Hb) (g/dL), Packed cell volume (PCV) (%), Total erythrocyte count (TEC) ($\times 10^6$/mm^3), Total leukocyte count (TLC) ($\times 10^3$/mm^3), Erythrocyte sedimentation rate (ESR) (mm/2hrs) and differential leucocyte count i.e. granulocyte(%), lymphocyte (%), monocyte (%) has been recorded as 9.46 ±0.13, 26.53 ±0.68, 6.69±0.08, 11.28 ±0.28, 1.81±0.07, 63.02 ±0.82, 33.78 ±0.81, 3.28±0.16 respectively in subclinical mastitis (SCM) affected animals. The corresponding mean values were 9.27 ±0.17, 25.12 ±0.74, 6.59±0.09, 12.57c±0.49, 1.16 ±0.08, 69.18 ±1.34, 28.07 ±1.38 and 2.75±0.20 respectively in clinical mastitis (CM) affected animals. In control group, the respective values were 10.89 ±0.23, 33.4 ±0.84, 7.1±0.12, 6.84 ±0.41, 1.11±0.12, 36.6 ±0.84, 60.2 ±0.86 and 3.2±0.28. Statistical analysis revealed that there was significant (P<0.05) decrease in Hb, PCV, TEC and relative lymphocyte count in mastitis affected cows as compared to healthy cows. There was no change observed in values of Hb and PCV between subclinical and CM affected animals. Relative lymphocyte count was decreased significantly (lymphopenia) in CM affected animals as compared to SCM.

TLC and Granulocyte count increased significantly (P<0.05) in CM as compared to SCM as well as healthy control group. ESR was increased significantly (P<0.05) in SCM as compared to normal and CM; however no significant change was observed between CM infected and healthy animals. No significant difference was observed among the 3 groups with respect to relative monocyte count.

There was significant (P<0.05) decrease in Hb, PCV, TEC and relative lymphocyte count with increase in TLC in mastitis affected cows as compared to healthy cows. There was no change observed in values of Hb and PCV between subclinical and CM affected animals. Relative lymphocyte count was decreased significantly (lymphopenia) in CM affected animals as compared to SCM.

Higher leukocyte and granulocyte count along with lymphopenia and anaemia in mastitic cows was earlier reported [2]. They also opined that the changes in haemato-biochemical parameters and milk leukocyte count can be used as important indicators of the physiological or pathological state (mastitis) of the animal. These observations were also in accordance with earlier report where significant reduction in RBC, Hb and PCV values leading to anaemia in animals affected with mastitis was reported [8]. However, No significant trends in PCV and Hb in mastitic cases were observed earlier [6] Higher leukocyte and granulocyte count was recorded in the present study in clinical as well as SCM affected animals. Increase in TLC in affected animals along with a higher monocyte, neutrophil and eosinophil count [7]. The ESR of SCM infected animals was found to be higher than healthy animal however, no significant (P>0.05) change was observed in CM affected animals. These findings are in agreement with the earlier reports [8, 9]

Table 1: Haematological parameters of mastitis affected cattle

Parameters	Control (n= 20)	Subclinical mastitis (n=60)	Clinical mastitis (n=40)
Hb (g/dL)	10.89^{a}±0.23	9.46^{b}±0.13	9.27^{b}±0.17
PCV (%)	33.4^{a} ±0.84	26.53^{b}±0.68	25.12^{b}±0.74
TEC x10^6/µL	7.1±0.12^{a}	6.69±0.08^{b}	6.59±0.09^{b}
TLC x10^3/µL	6.84^{a}±0.41	11.28^{b}±0.28	12.57^{c}±0.49
Granulocyte (%)	36.6^{a}±0.84	63.02^{b}±0.82	69.18^{c}±1.34
Lymphocyte (%)	60.2^{a}±0.86	33.78^{b}±0.81	28.07^{c}±1.38
Monocyte (%)	3.2±0.28	3.28±0.16	2.75±0.20
ESR (mm/2hrs)	1.11^{a}±0.12	1.81^{b}±0.07	1.16^{a}±0.08

* Means ±SE bearing different superscripts in a row vary significantly (P<0.05).

3.2 Serum Biochemical parameters

The serum biochemical profile in clinical and subclinical mastitis in the present study has been represented in Table-2. The mean values of Alanine Amino Transferase (ALT) (U/L), Aspartate Amino Transferase (AST) (U/L), Total protein (TP) (g/dL), Calcium (Ca) (mg/dL), Phosphorus P (mg/dL), Magnesium (Mg) (mg/dL), Chloride (Cl) (mEq/L), Sodium (Na) (mEq/L) and Potassium (K) (mEq/L) were found to be 18.49±1.09, 89.23 ±2.16, 7.39 ±0.07, 10.41±0.16, 6.15 ±0.09, 2.25±0.09, 99.86 ±0.96, 134.02 ±1.98 and 4.47±0.15 respectively in healthy control group animals. The respective values were found to be 19.03±0.63, 124.84 ±1.62, 7.84 ±0.14, 13.24 ±0.24, 5.12 ±0.11, 2.11±0.11, 86.28 ±1.02, 153.16 ±2.25, 5.10 ±0.12 respectively in SCM affected animals. In CM affected animals the corresponding mean values were 19.16±0.64, 101.76 ±1.34, 7.91±0.16, 12.98 ±0.38, 5.38 ±0.18, 2.25±0.16, 88.32 ±1.47, 156.74±2.12 and 5.38±0.19. Statistical analysis revealed that there was significant (P<0.05) increase in levels of AST, TP, Ca, Na and K in mastitic cows as compared to healthy cows. There was significant (P<0.05) decrease in levels of P and Cl mean values in mastitis affected cows as compared to healthy animals. There was significantly higher AST level in SCM affected animals than CM. There was no significant difference between the SCM and CM affected animals with respect to other parameters. There was significant (P<0.05) increase in levels of AST, TP, Ca, Na and K in mastitic cows as compared to healthy cows. There was significant (P<0.05) decrease in levels of P and Cl mean values in mastitis affected cows as compared to healthy animals. There was significantly higher AST level in SCM affected animals than CM. There was no significant difference between the SCM and CM affected animals with respect to other parameters.

Biochemical estimation revealed significantly (P<0.05) higher values of Ca, P, Na, Cl, and K in mastitis infected cows compared with healthy animals with no significant change in Mg and ALT levels [2, 10]. AST and TP values were significantly increased in SCM infected as compared to healthy animals. Higher levels of globulin and total protein was reported in serum of mastitic cows [11, 12]. However, reduced TP values in mastitic cases were also observed [7]. This may be attributed to the decreased albumin levels after the immune response to the udder infection [13]. Serum calcium level of the SCM and CM infected animals were significantly (P<0.05) higher than the healthy animals which is attributed to the reduced milk production in affected animals which causes decreased Ca excretion in milk. Singh *et al*. (2014) reported higher plasma levels of Ca in mastitis affected buffaloes which is similar to our findings [14]. But in contrast to the present study reduction in the Ca values in the infected animals were earlier reported [8]. Average value of phosphorous of the healthy animals were significantly (P<0.05) higher than the SCM and CM infected animals which could be attributed to its higher secretion in milk, due

to injury to the udder wall resulting in increased loss in milk which is in accordance with the earlier observation [11]. In the present study, no significant change in magnesium levels were recorded in mastitis affected animals which is in agreement with the earlier [11]. However, higher levels in serum Mg of buffaloes suffering from acute mastitis were reported by Singh (1999) [15] and decreased serum Mg levels were recorded by Siddiqe *et al*. (2015) [16]. Sodium, Chloride and Potassium levels of the SCM and CM infected animals were significantly (P<0.05) higher than the healthy animals. An increased level of sodium and potassium in serum of mastitic cases were observed which was in line with earlier findings [17]. This was attributed to the reduced milk yield during mastitis which elevates the level of sodium, potassium and chloride in blood due to minimal loss from the mastitis affected animal. Increased levels of plasma K in buffaloes suffering from mastitis was also reported [15]. The highly significant increases detected in AST values of SCM are in line with the reports of Bayumi *et al*. (2005) [18] and Chandrasekaran *et al*. (2015) [19] which could be due to stressful conditions.

Table 2: Serum biochemical parameters of mastitis affected animals

Parameters	Control (n=20)	Subclinical mastitis (n=60)	clinical mastitis (n=40)
ALT (U/L)	18.49±1.09	19.03±0.63	19.16±0.64
AST (U/L)	89.23^{a}±2.16	124.84^{b}±1.62	101.76^{c}±1.34
TP (g/dL)	7.39^{a}±0.07	7.84^{ab}±0.14	7.91^{b}±0.16
Ca (mg/dL)	10.41^{a}±0.16	13.24^{b}±0.24	12.98^{b}±0.38
P (mg/dL)	6.15^{a}±0.09	5.12^{b}±0.11	5.38^{b}±0.18
Mg (mg/dL)	2.25±0.09	2.11±0.11	2.25±0.16
Cl (mEq/L)	99.86^{a}±0.96	86.28^{b}±1.02	88.32^{b}±1.47
Na (mEq/L)	134.02^{a}±1.98	153.16^{b}±2.25	156.74^{b}±2.12
K (mEq/L)	4.47^{a}±0.15	5.10^{b}±0.12	5.38^{b}±0.19

* Means ±SE bearing different superscripts in a row vary significantly (P<0.05).

4. Conclusion

The changes in haemato-biochemical parameters can be used as important indicators of the physiological or pathological state (mastitis) of the animal.

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