The Pharma Innovation Journal 2022; SP-11(9): 3114-3116

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(9): 3114-3116

www.thepharmajournal.com
Received: 17-07-2022
Accepted: 20-08-2022

Vittal
PhD scholar department of veterinary medicine veterinary College, Bidar, Karnataka, India

Vivek R Kasaralikar
Professor and Head, Department of veterinary medicine veterinary college Bidar, Karnataka, India

Patil NA
Director of Extension, KVAFSU, Bidar, Karnataka, India

Ravindra BG
Associate professor and Head VCC Veterinary College, Shivmoga, Karnataka, India

Sandeep H
Associate Professor and Head VCC, Veterinary College, Bidar, Karnataka, India

Dilipkumar D
Dean Veterinary College, Bidar, Karnataka, India

Shrikant Kulkarni
Professor and Head, Department of veterinary physiology and Biochemistry, Veterinary College, Bidar, Karnataka, India

Corresponding Author:
Vittal
PhD scholar, Department of Veterinary Medicine Veterinary College, Bidar, Karnataka, India

Prevalence, antibiotic sensitivity and haemato-biochemical changes in subclinical mastitis of buffaloes

Vittal, Vivek R Kasaralikar, Patil NA, Ravindra BG, Sandeep H, Dilipkumar D and Shrikant Kulkarni

Abstract
The study was conducted to determine the prevalence, antibiotic sensitivity and haemato-biochemical changes in sub-clinical mastitic buffaloes of Bidar district. The sub-clinical mastitis (SCM) was diagnosed by California Mastitis Test (CMT). The overall prevalence of subclinical mastitis was 54.68 percent. Antibiotic sensitivity of milk and haemato-biochemical assay on blood/ serum was done. Enrofloxacin and gentamicin showed higher sensitivity whereas highest resistance was recorded for Cefotaxime and tetracycline. Haematology revealed significantly (p<0.05) lower values of Hb, PCV and TEC whereas significant increase in TLC in SCM infected animals compared to healthy animals. Differential leucocyte count revealed higher neutrophil and monocyte count in infected animals than healthy animals. Serum mineral estimation revealed significant (p<0.05) increase in calcium level and significant decrease in phosphorus level in mastitis buffaloes as compared to healthy control.

Keywords: Subclinical mastitis, prevalence, haemato-biochemical, antibiotic sensitivity assay

Introduction
Mastitis is an inflammation of the parenchyma of the mammary gland. This condition is a major endemic disease that can affect lactating animals (Rinaldi *et al*., 2010) [8]. India is one of the leading milk producing countries in the world with a total milk production of 211 million tons (NDDB 2020-21). Costs associated with mastitis include reduced milk production and quality, increased veterinary cost, increased risk of culling of infected animals and deaths (Seegers *et al.,* 2003) [9]. Riekerink *et al.*, (2008) [7] categoried mastitis into subclinical and clinical form wherein minimal or no apparent signs are present in subclinical form however there is decrease in milk production and change in milk composition (Guidry, 2007) [15].

Materials and Methods
320 buffaloes were screened in and around Bidar and cases presented to Veterinary Clinical Complex, Veterinary College Bidar, in that 175 animals which were positive for SCM, 30ml of milk samples from these animals were collected aseptically (after discarding first few streaks of milk) in a sterile containers. The collected milk samples were immediately transported on ice to the bacteriology laboratory. Upon arrival, milk samples were kept incubated at 37 °C for 12 h then cultured in 5% sheep blood agar, MacConkey agar, Eosin Methylene (EMB) agar and Mannitol salt (MS) agar by striking method. Blood sample was collected from jugular vein in EDTA and clot activator vials. TEC, DLC, Hb, RBC, WBC and PCV was estimated with haematology auto analyser (Blood Cell Counter PCE-210 VET) and calcium, phosphorus, total protein, AST and ALT estimations were done using semi automatic biochemical analyser (Microlab-300) using commercial kit.
Antibiotic sensitivity assay was done on overnight culture of individual bacteria in nutrient broth spread on a Muller Hinton Agar plate and different antibiotic discs viz., amoxyclav (30 mcg), ampicillin+cloxacillin (10 mcg), tetracycline (30 mcg), Pefloxacin (5 mcg), gentamicin (10 mcg), enrofloxacin (10 mg), cefepime (10 µg), cefotaxime (10 mcg), ceftriaxone (30 mcg) were placed equidistantly and incubated overnight. After incubation, the zone of inhibition was measured using scale and recorded in millimeters (mm). Later, these reading were compared with the manufacturers chart. The data generated for haemato-biochemical parameters from the study were analyzed by two-way ANOVA with the application of Bonferroni post-test using 'Graph Pad Prism' version 5.01 (2007) computerized software.

The values were expressed as Mean ± Standard Error and the level of significance was determined at P value of 0.05.

Results and Discussion

In the present study, the prevalence of subclinical mastitis was found 54.69 percent in buffaloes. In comparision the prevalence of subclinical was higher than the clinical mastitis in buffaloes. These findings are in close alignment with the findings of Ali *et al.*, (2021) [1] which reported that prevalence of subclinical mastitis was 66% and clinical mastitis was 11.00% in buffaloes. Variation in prevalence of mastitis might be due to the different regions, breeds, differences in management conditions, especially milking management, hygienic condition, care of teat injuries, presence of microorganisms in environment and adaptation of mastitis control program.

Table 1: Prevalence study:

Total number of animals screened	Positive for SCM	Prevalence (%)
320	175	54.69

The results of antibiotic sensitivity assay of the bacterial isolates are shown in Table 2. Enrofloxacin and gentamicin showed the highest sensitivity to various bacterial isolates from milk of buffaloes suffering mastitis. Overall, 86 percent of the bacterial isolates were susceptible to gentamicin, followed by 80 percent susceptibility to enroflfloxacin. The result of present study were in accordance with the findings of Ali *et al*, (2021) [1], wherein highest sensitivity was recorded for gentamicin and enrofloxacin. Antibiotic sensitivity showed substaintial decline in sensitivity of following antibiotics viz., Pefloxacin (71%), Ceftriaxone (57%), Ampicillin + Cloxacillin (51%), Cefotaxim and Amoxyclav (34%) and the least sensitivty was found for Tetracycline (23%). This may be due to indiscriminate and frequent use of these antibiotics (Sharma *et al.*, 2007) [11].

Table 2: Antibiotic sensitivity assay:

Antimicrobial agents	Concentration (µg)	Susceptible isolates		Resistant isolates	
		n	%	n	%
Enrofloxacin	10	30	86	5	1
Gentamicin	10	28	80	7	2
Pefloxacin	05	25	71	10	28
Ceftriaxone	30	20	57	15	43
Ampicillin+Cloxacillin	10	18	51	10	29
Cefotaxime	10	12	34	23	66
Amoxyclav	30	12	34	16	46
Tetracycline	30	8	23	22	62

Table 3: Mean ±SE values of haematological parameters of healthy and mastitic Buffaloes

Parameter	Healthy (n=6)	Sub clinical (n=175)
Hb (g/dL)	9.34^{a}±0.284	7.16^{b}±0.10
PCV (%)	29.95^{a}±0.98	22.16^{b}±0.36
TEC (x106 /µL)	6.14^{a}±0.43	4.26^{b}±0.07
TLC (x103 /µL)	8.35^{a}±0.63	11.7^{b}0±0.12
Neutrophils (%)	37.73^{a}±1.34	57.97^{b}±0.27
Lymphocyte (%)	58.55^{a}±1.05	30.34^{b}±0.24
Monocyte (%)	2.00^{a}±0.36	3.42^{a}±0.06

Means marked with different superscript differ significantly (p<0.05) in a row

Haemoglobin (Hb g/dL), PCV (%) and TEC of SCM animals revealed significant (p<0.05) decrease than control group. These findings are in accordance with the findings of Zaki *et al.*, (2008) [14] and Krishnappa *et al.*, (2016**)** [16] who also reported anaemia in mastitic buffaloes due to decrease in Hb, RBC and PCV levels which could be due to chronic infection noticed in SCM.

There was significant (p<0.05) increase in total leukocyte count (TLC) and neutrophil count, along with significant (p<0.05) decrease in the lymphocyte count were observed in buffaloes of subclinical mastitis. Simillar finding were recorded by Khan *et al.*, (1997) [5] and Krishnappa *et al.*, (2016**)** [16] which may be due to persistent prolong microbial infection in udder and these were the major cells of the immune response of the infection (Gargouri *et al.*, 2008) [3].

Table 4: Mean ±SE values of biochemical parameters of control and mastitic Buffaloes

Parameter	Healthy (n=6)	Sub clinical (n=175)
Total Protein (g/dL)	7.05^{a}±0.30	6.32^{b}±0.07
Calcium (mg/dL)	8.61^{a}±0.31	10.63^{b}±0.07
Phosphorus (mg/dL)	5.94^{a}±0.41	4.04^{b}±0.06
Alanine transaminase (U/L)	8.60^{a}±0.20	7.91^{a}±0.07
Aspartate transaminase (U/L)	76.98^{a}±1.26	74.85^{a}±0.43

Means marked with different superscript differ significantly (p<0.05) in a row

There was significant (p<0.05) decrease in total protein (TP) level of SCM cases compared to healthy group. These findings are in accordance with the findings of Mosallam *et al.*, (2006) [17] who reported a notable decrease in TP values of SCM infected buffaloes and this may be attributed to the decreased albumin levels after the immune response to the udder infection (Singh, 2000) [10].

Serum calcium level of the SCM infected animals were significantly (p<0.05) higher than the healthy animals. These findings were in accordance with Singh *et al.*, (2014) [18] who reported significantly (p<0.05) increased levels of plasma Ca in buffaloes suffering from mastitis which is attributed to the reduced milk production in affected animals which causes decreased Ca excretion in milk (Wegner and Stull, 1978) [13].

Serum phosphorus was significantly (p<0.05) lower in SCM infected animals compare to healthy animals this may be due to injury to the udder wall result in increased loss in milk

which is in accordance with the observation of other researchers *viz.*, Siddiqe *et al*., (2015) [12]. the findings were in Contrary to Zaki *et al.*, (2008) [14] who reported no significant change in the values of P in serum of mastitis infected as compared to healthy buffaloes. There was no significant difference observed in the ALT and AST levels of SCM animals compared to healthy animals.

Conclusion

The present study showed that there was high prevalence of SCM in the study area. The antibiotic sensitivity assay of milk revealed Gentamicin and enrofloxacin showed high sensitivity whereas more resistant to Cefotaxime and tetracycline. Their was decrease in the levels of Hb, PCV, TEC and lymphocyte count and increase in TLC, neutrophil and monocyte count in SCM. Increase in calcium levels and decrease in total protein and phosphorus levels in SCM animals.

Reference

1. Ali T, Kamran, Raziq A, Wazir I, Ullah R, Shah P, Ali MI, *et al*. Prevalence of Mastitis Pathogens and Antimicrobial Susceptibility of Isolates From Cattle and Buffaloes in Northwest of Pakistan. Front. Vet. Sci. 2021;8:746-755.
2. Bilal MQ, Iqbal MU, Muhammad G, Avais M, Sajid MS. Factors affecting the prevalence of clinical mastitis in buffaloes around Faisalabad district (Pakistan). Int. J Agri. Biol. 2004;6:185-187.
3. Gargouri A, Hamed H, Elfeki A. Total and differential bulk cow milk somatic cell counts and their relation with lipolysis. Livest. Sci. 2008:113:274-279.
4. Gruet P, Maincent P, Berthelot X, Kaltsatos V. Bovine mastitis and intramammary drug delivery: review and perspectives. Adv. Drug. Deliv. Rev. 2001;50:245-249.
5. Khan MZ, Muhammad G, Umar A, Khan SA. A preliminary comparison of plasma fibrinogen concentrations, leukocyte numbers and erythrocyte sedimentation rate as non-specific indicators of inflammatory conditions in buffalo (*Bubalis bubalis*). Vet. Res. Commun. 1997;21:265-271.
6. Park HR, Hong MK, Hwang SY, Park YK, Kwon KH, Yoon JW *et al*. Characterization of Pseudomonas aeruginosa related to bovine mastitis. Acta Veteśśśrinaria Hungarica. 2013;62:1-12.
7. Riekerink RO, Barkena HW, Kelton DF, Scholl DT. Incidence rate of clinical mastitis on Canadian dairy farms. Journal of Dairy Science. 2008;91:1366-1377.
8. Rinaldi M, Li RW, Capuco AV. Mastitis associated transcriptomic disruptions in cattle. Vet. Immunol. Immunopathol. 2010;138(4):267-279.
9. Seegers H, Fourichon C, Beaudeau F. Production effects related to mastitis and mastitis economics in dairy cattle herds. Vet. Res. 2003;34(5):475-491.
10. Singh SV. Udder health profiles with special reference to acute phase proteins and supplementation therapy. M.V.Sc thesis submitted to the GB Pant University of Agriculture and Technology, Pantnagar, India, 2000.
11. Sharma N, Maiti SK, Sharma KK. Prevalence, etiology and antibiogram of microorganisms associated with subclinical mastitis in Durg, Chhattisgarh State (India). Int. J Dairy Sci. 2007;22:145-151.
12. Siddiqe ZF, Islam S, Islam SS, Islam S, Islam S, Das BC. Haemato-biochemical changes in subclinical mastitis affected high yielding dairy cows in Chittagong district. Int. J Nat. Soc. Sci. 2015;2(4):30-34.
13. Wegner TN, Stull JW. Relationship between mastitis test score, mineral composition of milk and blood electrolyte profiles in Holstein cows. J Dairy Sci. 1978:61(12):1755-1759
14. Zaki MS, Sharaf NE, Mostafa SO, Fawzi OM, El-Battrawy N. Effect of subclinical mastitis on some biochemical and clinic-pathological parameters in buffalo. Am-Euras. J Agric. Environ. Sci. 2008;3(2):200-204.
15. Singh A, Guidry L, Narasimhulu KV, Mai D, Trombley J, Redding KE, *et al*. Mycobacterium tuberculosis WhiB3 responds to O2 and nitric oxide via its [4Fe-4S] cluster and is essential for nutrient starvation survival. Proceedings of the National Academy of Sciences. 2007 Jul 10;104(28):11562-7.
16. Boregowda SV, Krishnappa V, Haga CL, Ortiz LA, Phinney DG. A clinical indications prediction scale based on TWIST1 for human mesenchymal stem cells. EBioMedicine. 2016 Feb 1;4:62-73.
17. Rizk DE, Mosallam M, Alyan S, Nagelkerke N. Prevalence and impact of premenstrual syndrome in adolescent schoolgirls in the United Arab Emirates. Acta obstetricia et gynecologica Scandinavica. 2006 May;85(5):589-98.
18. Singh S, Prasad SM. Growth, photosynthesis and oxidative responses of Solanum melongena L. seedlings to cadmium stress: mechanism of toxicity amelioration by kinetin. Scientia Horticulturae. 2014 Sep 11;176:1-0.