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PREVALENCE OF BOVINE MASTITIS IN NORTH KARNATAKA, INDIA

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Abstract

The aim of this study was to determine the prevalence of bovine mastitis in North Karnataka Region. The samples were collected from March 2012 to August 2012. A Total of 392 herds (cows and buffaloes) milk samples were collected fortnightly and considered in this study. The study includes Holstein Freshein, Jerscy, Dharwari and Murrah. The overall prevalence was 69.89%, from which 48.21% and 21.68% were subclinical and clinical mastitis respectively. A total of 148 buffalos were tested, among which 96 (64.86%) were affected. A total of 244 cows were tested, among which 178 (72.95%) were affected. The age based prevalence of mastitis in buffalos showed that the maximum affect was at the age of 10 years (86.66%) and 11 years (15.38%) for subclinical and clinical mastitis respectively. Whereas 6 years (57.44%) and 11 years (15.78%) for subclinical and clinical mastitis respectively in cows. The lactation based prevalence of mastitis in buffalos showed that the maximum affect was seen in 3rd month (70.83%) and 7th month (25%) for subclinical and clinical mastitis respectively. Whereas 1st month (76.19%) and 4th month (28.12%) for subclinical and clinical mastitis in cows. The quarter based prevalence of mastitis in buffalos showed that the maximum affect was 39.86% and 9.37% in one quarter in subclinical and clinical mastitis respectively. Whereas 47.13% and 6.96% in one quarter for subclinical and clinical mastitis respectively in cows. A total of 06 groups of bacteria's (*Staphylococcus aureus*, Coagulase negative *staphylococcus* (CNS), *Bacillus* spp. *Pseudomonas* spp. *Aerococcus* spp. *Enterobacter* spp.) were isolated based on conventional methods. The present study showed that there was high prevalence of mastitis in the North Karnataka, India and hence, it is recommended to take necessary control measures.

Keywords: Bovine Mastitis, Prevalence, North Karnataka.

Introduction

Mastitis in dairy cows is a serious problem as it is an economically devastating disease causing immense economic losses in the dairy industry[1,2] and is the worldwide costliest production disease in dairy herds[3]. It stands second to Foot and Mouth Disease as a most challenging disease in high yielding dairy animals in India[4] as documentary, but as per reports of occurrence of mastitis in dairy animals, it stands at first position because prevalence of mastitis had been reported more than 90% in high yielder cross bred dairy cows[5]. Bovine mastitis is caused by entry of bacteria in the mammary gland leading to inflammation[6]. This disease, characterized by an increase in somatic

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cells, especially leukocytes, in the milk and by pathological changes in the mammary tissue[7], causes colossal economic losses, but also hold the risk for the transmission of zoonotic diseases like tuberculosis, brucellosis, leptospirosis and streptococcal sore throat to human beings[8]. Bovine mastitis is generally classified into clinical and subclinical mastitis. Clinical mastitis is characterized by local (e.g. swelling of the udder, heat and pain) or systemic (e.g. fever, anorexia, depression) symptoms with milk abnormalities (e.g. milk clots, flakes, watery secretions, blood)[9]. Subclinical mastitis is the most serious type as the infected animal shows no obvious symptoms and secrets apparently normal milk for a long time, during which causative organisms spread infection in herd, so it is an important feature of the epidemiology of many forms of bovine mastitis[10].

Since the quality and quantity of the milk is influenced by mastitis, it is considered to be one of the most important causes of economic losses in the dairy industry worldwide. India is the second largest milk producer in the world with a milk production of around 112 million tons, which is worth more than Rs. 1,70,000 Crores. Buffalo milk accounts for approximately 57 % of total milk production. In India, bovine mastitis in has become extremely complex and the costliest disease. It affects more than 50% of the herd population[11]. It has been estimated that the mastitis alone can cause approximately 70% of all avoidable losses incurred during milk production[12]. The first comprehensive report on mastitis caused losses in India published in 1962 indicated annual losses of Rs. 52.9 crore [13]. However, tremendous thrust on cross breeding programme and launching of operation flood in later years resulted in tremendous increase in high yielding bovine population, leading to many fold increase in economic loss. This is evidenced from a recent report where in annual economic losses incurred by dairy industry in India on account of udder infections have been estimated about Rs.6053.21 crore. Out of this, loss of Rs. 4365.32 crore (70 % - 80 % loss) has been attributed to sub clinical version of udder infections [14]. Production of high quality milk requires an efficient mastitis control programme. Herds with a high prevalence of mastitis as in this study are incapable of producing high quality milk until the inflammation and infection in the udder are brought under control. This has severe economic implications for the milk producer, as the milk is no longer marketable and other animals are easily infected. Treatment and decrease in milk volume also cause considerable losses per animal[15]. The aim of this study was to determine the prevalence of bovine mastitis in North Karnataka, India.

Materials and Methods

Study area

The North Karnataka is located within 15°00' North (N) and 18°30' North (N) latitudes and 74° East (E) and 77°50' (E) East longitude. The border is bounded by Maharashtra and Goa States in the north and northwest and the State of Andhra Pradesh in the east. This region is mainly called as Bayaluseeme region comprising the plains of the Deccan plateau.

Source of Study Animals

The present study was carried out on bovine mastitis in North Karnataka, India (09 districts) from March 2012 to August 2012. A Total of 392 herds (cows and buffaloes) milk samples were collected fortnightly and considered in this study. The study includes Holstein Freshein (H.F), Jerscy, Dharwari and Murrah. The lactating cattles of the dairy farms of the North Karnataka Region has been examined from dairy herds in different smallholder farms as well as large scale farms randomly. Information on age, lactation stage and previous history of mastitis has been gathered. The prevalence of mastitis in herds were determined by examination of changes in the udder viz., swelling, redness and hardness of udder, increase in temperature, changes in milk colour and reduction in quality of the milk. Further, Surf Field Mastitis Test (SFMT) and increased $_{P}$H of the milk have been done to confirm the clinical and subclinical mastitis.

Sampling method

Quarter foremilk samples were collected aseptically for bacteriological assay as described by Honkanen-Buzalski. Before sampling, teat ends were disinfected with cotton swabs soaked in 70% ethanol and allowed to dry and the first streams of milk were discarded. Milk samples were collected in sterile 15 ml tubes. The milk samples were transported in a cold container to the laboratory of the P. G. Department of studies in Microbiology and Biotechnology, Karnatak University, Dharwad for further analysis.

Surf field mastitis test

The samples were subjected to Surf Field Mastitis test (SFMT). The principle of the test is that when detergent is added into milk sample, it causes rupture of somatic cell and release DNA and other cell contents. DNA is acid in nature, while detergent contains alkyl-arylsulfonate, which is basic in nature. DNA and detergents unite to form a gel; consistency of gel depends upon the number of somatic cells. More cells more thick gel and vice versa. For this purpose, three percent surf solution (pH = 10.3) was prepared by adding three grams of commonly used detergent powder (Surf Excell, Uniliver, India) in 100 mL of water. Quarter milk samples and surf solution were then mixed in equal quantities in petri-dishes separately for each quarter. The change in consistency of milk indicated mastitis, while no change in consistency of milk indicated healthy samples. The mastitis was graded into further four categories based on the severity of disease from lower to higher intensity as, + = moderate, + + = severe, + + + = more severe, + + + + = very severe (16, 17). The percentage of prevalence was calculated.

Breed based prevalence of bovine mastitis

The breed based prevalence of mastitis was studied by using different breeds like Cows (*Holstein freshen* (H.F) and Jersey), Buffalo's (Dharwari and Murrah).

Age based prevalence of bovine mastitis

Cows and buffalo's aged 5 years to 11 years were used in the present study to know the age based prevalence of clinical mastitis.

Lactation based prevalence of bovine mastitis

Cows in between 1^{st} to 7^{th} lactation were tested to know the lactation based prevalence of clinical mastitis.

Quarter based prevalence of bovine mastitis

The quarter based prevalence of clinical mastitis was considered in the study.

Statistical analysis

The data was statistically analyzed to know the breed, age, lactation and quarter based prevalence of subclinical and clinical mastitis. Chi-square test was used to know if statistically significant association existed between the breed, age groups, lactation period. For all the analysis performed $P < 0.001$ was taken as statistically significant[18].

Isolation and identification of bacteria from bovine mastitis milk

From the collected milk samples 0.01 ml of milk was cultured on nutrient agar and incubated for 48 hrs at 37°C. The plates were examined for growth and colony morphology. Isolated organisms were streaked on differential medium and identification was carried out by conventional methods like grams staining, microscopic observation and various biochemical tests.

Results

A total of 392 animals from North Karnataka Region were tested for mastitis. The numbers of confirmed subclinical and clinical mastitis from North Karnataka Region showed that total 69.89% animals were affected. From which 48.21% and 21.68% were subclinical and clinical mastitis respectively (Table 1). A total of 244 cows were tested, among which 178 (72.95%) were affected, whereas 61.47% and 11.47% were subclinical and clinical respectively. The breed based prevalence in cows showed that 78.78% and 66.07% were H.F and Jersey, among which 46.21% and 9.84% were subclinical and clinical mastitis respectively in H. F and 79.46% and 13.39% were subclinical and clinical mastitis respectively in Jersey. A total of 148 buffalos were tested, among which 96 (64.86%) were affected, whereas 56.08% and 8.78% were subclinical and clinical respectively. The breed based prevalence in buffalos showed that 47.82% and 72.54% were Dharwari and Murrah, among which 43.47% and 4.34% were subclinical and clinical mastitis respectively in Dharwari and 61.76% and 10.78% were subclinical and clinical mastitis respectively in Murrah. The statistical analysis of prevalence of bovine mastitis between different breeds showed that there is a significant effect of mastitis among the different breeds in the study area, the χ^2 for the analysis is 15.46 ($P < 0.001$), where degree of freedom (df) is 3 (Table 2).

The age based influence on prevalence of mastitis in cows showed that the maximum affect was seen at the age of 6 years (57.44%) and 11 years (15.78%) for subclinical and clinical mastitis respectively. Whereas minimum affect was observed at the age of 9 years (24.24%) and 5 years

(2.94%) for subclinical and clinical mastitis respectively (Table 3). The age based influence on prevalence of mastitis in buffalos showed that the maximum affect was at the age of 10 years (86.66%) and 11 years (15.38%) for subclinical and clinical mastitis respectively. Whereas minimum affect was observed at the age of 6 years (5.55%) and 8 years (4.34%) for subclinical and clinical mastitis respectively (Table 4). The statistical analysis of prevalence of bovine mastitis between age group showed that there is a significant effect on older age group comparatively than younger age of the animals in the study area, the χ^2 value is 19.47 and 21.38 for cows and buffalos respectively, where degree of freedom (df) is 6.

The lactation based influence on prevalence of mastitis in cows showed that the maximum affect was seen in 1st month (76.19%) and 4th month (28.12%) for subclinical and clinical mastitis respectively. Whereas minimum affect was showed in the 4th month (53.12%) and 1st month (4.76%) for subclinical and clinical mastitis respectively (Table 5).

The lactation based influence on prevalence of mastitis in buffalos showed that the maximum affect was seen in 3rd month (70.83%) and 7th month (25%) for subclinical and clinical mastitis respectively. Whereas minimum affect was showed in the 7th month (41.66%) and 1st month (3.57%) for subclinical and clinical mastitis respectively (Table 6). The statistical analysis of prevalence of bovine mastitis between lactation period showed that there is a significant effect on earlier months and middle months comparatively than later months of the animals in the study area, the χ^2 value is 5.61 and 8.62 for cows and buffalos respectively, where degree of freedom (df) is 6.

A total of 592 quarters of 148 lactating buffaloes, 174 (29.39%) quarters were mastitic and a total of 976 quarters of 178 lactating cows, 237 (24.28%) quarters were mastitic. (Table no 7) The quarter based influence on prevalence of mastitis in cows showed that the maximum affect was 47.13% and 6.96% in one quarter for both subclinical and clinical mastitis respectively. Whereas minimum affect was 0.81% in four quarter and 1.63% in two quarter for subclinical and clinical mastitis respectively (Table 8).

The quarter based influence on prevalence of mastitis in buffalos showed that the maximum affect was 39.86% and 9.37% in one quarter for both subclinical and clinical mastitis respectively. Whereas minimum affect was 6.25% in three quarter and 2.70% in two quarter for subclinical and clinical mastitis respectively (Table 9). A total of 06 groups of bacteria, i.e. *Staphylococcus aureus*, Coagulase negative staphylococcus (CNS), *Bacillus* spp., *Pseudomonas* spp., *Aerococcus* spp., *Enterobacter* spp. were isolated based on conventional methods like grams staining, microscopic observation and various biochemical tests (Table 10).

Table No. 01: Prevalence of clinical and sub clinical mastitis from north Karnataka.

S.No	District	Total prevalence			Sub-Clinical		Clinical	
		No of animals tested	No of animals affected	Percentage (%)	Positive	Percentage (%)	Positive	Percentage (%)
1	Dharwad	104	88	84.61	70	67.30	18	17.30
2	Belgaum	44	33	75.00	21	47.72	12	27.27
3	Bijapur	31	15	48.38	08	25.00	07	22.58
4	Bagalkot	32	21	65.62	15	46.87	06	18.75
5	Gadag	56	41	73.21	34	60.71	07	12.50
6	Koppal	33	24	72.72	16	48.48	08	24.24
7	Bidar	26	14	53.84	11	42.30	03	11.53
8	Gulbarga	48	27	56.25	06	12.50	21	43.75
9	Raichur	18	11	61.11	08	44.44	03	16.66
	Total	392	274	69.89	189	48.21	85	21.68

Table No. 02: Breed based prevalence of subclinical and clinical mastitis in cows and Buffalos

Breed		Total Number of Animals			Subclinical		Clinical	
		Examined	Affected	Percentage (%)	Posit-ive	Percentage (%)	Positive	Percentage (%)
Cow	H.F	132	74	66.07	61	46.21	13	9.84
	Jersey	112	104	78.78	89	79.46	15	13.39
	Total	244	178	72.95	150	61.47	28	11.47
Buffalos	Dharwari	46	22	47.82	20	43.47	02	4.34
	Murrah	102	74	72.54	63	61.76	11	10.78
	Total	148	96	64.86	83	56.08	13	8.78
$\chi 2$ values = 15.46*, df=3, P < 0.001								

H. F= Holstein Friesian * Significant

Table No. 03: Age based prevalence of subclinical and clinical mastitis in cows

Age (Years)	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
05	16	06	37.50	01	6.25
06	27	13	48.14	00	0.00
07	36	17	47.22	03	8.33
08	46	33	71.73	05	10.86
09	44	27	61.36	06	13.63
10	52	40	76.92	07	13.46
11	23	14	60.86	06	26.08
Total	244	150	61.47	28	11.47
$\chi 2$ values = 19.47*, df=6, P < 0.001					

* Significant

Table No. 04: Age based prevalence of subclinical and clinical mastitis in buffalos

Age (Years)	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
05	12	03	25.0	01	8.33
06	18	01	5.55	02	11.11
07	18	08	44.44	00	00
08	23	10	43.47	01	4.34
09	21	17	80.95	02	9.52
10	30	26	86.66	03	10.0
11	26	18	69.23	04	15.38
Total	148	83	56.08	13	8.78
$\chi 2$ values = 21.38*, df=6, P < 0.001					

* Significant

Table No. 05: Lactation based prevalence of subclinical and clinical mastitis in cows

Lactation (Months)	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
1	42	32	76.19	02	4.76
2	36	23	63.88	04	11.11
3	43	27	62.79	03	6.97
4	32	17	53.12	09	28.12
5	41	22	53.65	05	12.19
6	26	14	53.84	02	7.69
7	24	15	62.50	03	12.5
Total	244	150	61.47	28	11.47
$\chi 2$ values = 5.61*, df=6, P < 0.001					

* Significant

Table No. 06: Lactation based prevalence of subclinical and clinical mastitis in Buffalo

Lactation (Months)	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
1	28	15	53.57	01	3.57
2	18	12	66.66	00	00
3	24	17	70.83	04	16.66
4	26	13	50.00	02	7.69
5	23	11	47.82	01	4.34
6	17	10	58.82	02	11.76
7	12	05	41.66	03	25.00
Total	148	83	56.08	13	8.78
	$\chi 2$ values = 8.62*, df=6, P < 0.001				

* Significant

Table No. 07: Mastitis prevalence of Quarters examined

Breeds	Total No. of quarters	Affected animals	Mastitic Quarters	Prevalence (%)
Buffalos N=148	592	96	174	29.39
Cows N=244	976	178	237	24.28

Table No. 08: Quarter based prevalence of subclinical and clinical mastitis in cows

Quarter	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
One Quarter	244	115	47.13	17	6.96
Two quarters	244	31	12.70	04	1.63
Three quarters	244	9	3.68	00	00
Four quarters	244	2	0.81	00	00
Total	244	157	64.34	21	8.60

Table No. 09: Quarter based prevalence of subclinical and clinical mastitis in buffalo

Quarter	No. of animals Examined	Subclinical		Clinical	
		Positive	Percentage	Positive	Percentage
One Quarter	148	59	39.86	09	9.37
Two quarters	148	18	18.75	04	2.70
Three quarters	148	6	6.25	00	00
Four quarters	148	00	00	00	00
Total	148	83	86.45	13	13.54

Table No. 10: Bacterial isolates from the bovine mastitis milk

S. No	Bacterial species
1	*Staphylococcus aureus*
2	Coagulase negative staphylococcus (CNS)
3	*Bacillus* spp.
4	*Pseudomonas* spp.
5	*Aerococcus* spp.
6	*Enterobacter* spp.

Discussion

The present study was carried out on selected dairies located in North Karnataka Region to determine status of mastitis among cows and buffalos. This area has numerous smallholdings (farms) and a small number of larger herds. The standard of hygienic milking procedure was poor on the majority of the farms sampled. Preventive measures, such as the use of udder disinfectants, post-milking teat dipping and dry cow therapy,

were observed to be infrequent in these herds. The local information on the prevalence of bovine mastitis in the study area is extremely inadequate. This information is imperative for planning an intervention strategy for this costly disease. Without knowing the epidemiology, it is very difficult, rather impossible to control the disease.

The present study adds to the scarcity of information to the prevalence of mastitis in lactating herds. The overall prevalence of mastitis observed in this study was 69.89% which is higher than the previous findings 34.9% by[19]; 33.6 % by[20] and 40.4 % by[21]. The higher prevalence rate in the present study may be due environmental factors such as unhygienic management of herds. Our results revealed that the sub clinical and clinical mastitis was 48.21% and 21.68% respectively, which is higher than the results reported by [22] that the overall prevalence of subclinical and clinical mastitis was 33.8% and 3.6 % respectively.

The present study revealed a highly significant association (P< 0.001) between the mastitis status and different breeds (Cows and Buffalos). The overall prevalence of mastitis among cows was 72.95%, whereas 61.47% and 11.47% were subclinical and clinical respectively. These finding are higher than previous findings, 33.6 % by[20], 34.9 % by[19], 40.4 % by[21], 59.7 % by[23], the prevalence report of both clinical and subclinical mastitis is also higher than the findings of [23] who reported 38.2 % subclinical and 21.5% clinical mastitis. The difference of mastitic prevalence in breeds may be due to habit as well as due to more development of udder and teats in cows [24].

In the present study the overall prevalence of mastitis among buffalos was 64.86%, whereas 56.08% and 8.78% were subclinical and clinical respectively. The breed based prevalence in buffalos showed that 47.82% and 72.54% were Dharwari and Murrah, among which 43.47% and 4.34% were subclinical and clinical mastitis respectively in Dharwari and 61.76% and 10.78% were subclinical and clinical mastitis respectively in Murrah. Similarly, an incidence of mastitis in buffalos 54.7%, 32.85% and 23.18% has been reported by [20, 25, 26] respectively. Different breeds of cattle are known to differ in their susceptibility to mastitis. The buffalo breed Dharwari is significantly more resistant to the mastitis, comparatively to the other breeds. Variation in prevalence of mastitis might be due to the different regions, breeds, therapeutic practices, management conditions and presence of microorganisms in environment.

In the present study the prevalence of bovine mastitis between age group showed that there is a significant effect on older age group comparatively than younger age of the animals in the study area. In cows showed that the maximum affect was seen at the age of 6 years (57.44%) and 11 years (15.78%) for subclinical and clinical mastitis respectively. Whereas age based influence on prevalence of mastitis in buffalos showed that the maximum affect was at the age of 10 years (86.66%) and 11 years (15.38%) for subclinical and clinical mastitis respectively. Similar observations[27] have made that the prevalence of mastitis increased as the age advanced. Whereas [28] reported 57.5% prevalence of mastitis in the age group of higher than 9 years old and 40.1% in the age group of 7 and 8 years old. The present findings is fairly similar to the findings of [29, 30] who have recorded higher prevalence of mastitis in 4-9 years of age. Similarly[19] have suggested that older cows are at more risk (44.6%), for the incidence of mastitis than younger cows (23.6%). This may be possible due to the injured glandular tissue in the previous lactations and it has been revealed that high-yielding and aged cows are more susceptible for mastitis. In the high-yielding cows the glandular tissues are more susceptible to infection. Moreover, generally the immune mechanism in old cows is poorer than in younger cows[31].

In the present study prevalence of bovine mastitis between lactation period showed that there is a significant effect on earlier months and middle months comparatively than later months of the both Buffalos and Cows in the study area, In cows the maximum affect was seen in 1st month (76.19%) and 4th month (28.12%) for subclinical and clinical mastitis respectively. The lactation based influence on prevalence of mastitis in buffalos showed that the maximum affect was seen in 3rd month (70.83%) and 7th month (25%) for subclinical and clinical mastitis respectively. Similarly it has been reported by[27] that the incidence of 8.5% in first lactation and 26% in greater than or equal to fifth

lactation. High milk yielding animals were more susceptible to mastitis when compared to low yielding animals, which was in agreement with the observations reported by[32]. It was also found that none of the farmers practiced teat dipping as a preventive measure. Hence, it is essential to educate the farmers regarding the risk factors of mastitis and also about teat dipping.

Conclusion

In Conclusion the present study showed that there was high prevalence of mastitis in the study area. Overall 69.89% animals were affected. From which 48.21% and 21.68% were subclinical and clinical mastitis respectively. In cows the prevalence is more in Jersey and in buffalos the prevalence is more in Murrah. Based on the results of this study, it is recommended that in order to reduce the high prevalence of mastitis in the area, improved milking hygiene, prevention of skin lesions, culling of chronic mastitis carriers, and treating of clinically infected herds should be practiced.

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