Comparison of Efficacy of Three Cow Side Tests for the Diagnosis of Subclinical Mastitis in Dairy Cattle

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Abstract: Bovine mastitis due to bacterial origin is considered as the most economically important disease among livestock worldwide. The subclinical form is the most serious due to the gradual loss of milk production it causes. The early diagnosis is important for enhances prospects of recovery and reduces production loss. This work aimed at finding the prevalence of subclinical mastitis (SCM) in a herd and the comparison of three different cow side tests California Mastitis Test (CMT), electrical conductivity (EC) and somatic cell count (SCC) for the diagnosis of subclinical mastitis with the standard bacterial culture. There was high prevalence (57.73%) of SCM in the herd. SCC was found to be more accurate (58.56%) than CMT (54.05%) and EC (24.77%). The sensitivity of SCC (59.89%) was higher while EC showed more specificity (94.74%). The κ (Kappa) values of all the three tests indicated poor agreement with bacterial culture. The study poses the need for more advanced diagnostic tools for the early detection of cows with SCM.

Introduction

Bovine mastitis is defined as the inflammation of mammary gland of cows and in most cases has a bacteriological origin (Nederbragt, 2015). The direct as well as indirect losses caused by the disease to the farmer makes mastitis the most economically important disease of livestock worldwide. In comparison to clinical mastitis the subclinical form is more serious because of the absence of overt clinical signs and the gradual damage caused leading to permanent loss of production. Usually subclinical mastitis goes unnoticed by the farmers and hence untreated. The changes in the udder takes place much before the disease become evident (Sharma *et al.*, 2010). Thus early diagnosis is important for reducing the production loss and enhancing the prospects of recovery (Kamal *et al.*, 2014).

The present study aims to find the prevalence of subclinical mastitis in a dairy farm using three field level diagnostic techniques, California Mastitis Test (CMT), Electrical Conductivity (EC) and Somatic Cell Count (SCC) and compare the findings with the results of the gold standard test, the culture and isolation.

Materials and methods

Quarter milk samples were collected from 110 milking cows from a farm aseptically. The cows belonged to cross bred category and were in various stages of lactation. Machine milking was practiced in the farm with hygienic measures like teat dipping.

California Mastitis Test:

CMT was employed for the initial screening of the herd. The test was conducted as per procedure described by Schalm *et al.* (1971) using hand held paddle and CMT reagent. The results were interpreted as follows:

CMT score	Interpretation
0	No reaction or trace
1	Weak positive
2	Moderate positive
3	Strong positive

The CMT positive samples were subjected for further detailed investigation.

Measurement of electrical conductivity:

A commercial multi quarter electrical conductivity meter (Draminski, Poland) was used for the detection of EC as per manufacturer's instruction. Based on the EC, values were assigned to the samples by the machine. The values below 300 were taken as positive for subclinical mastitis.

Somatic Cell Count (SCC):

The somatic cell count was estimated as per the procedure suggested by Schalm *et al.*

(1971). Manual counting of cells after staining milk smears with Newman's stain was practiced. Bacteriological culture and isolation:

The samples with CMT score 2 and above were selected for bacteriological culture. The samples were streaked on Brain Heart Infusion Agar and incubated at 37^0C for 24 hours. The colonies were stained using Gram's stain and morphologically identified.

Result and discussion

High prevalence of subclinical mastitis was noticed in the farms with 66 (57.73%) cows out of the 110 tested cows being positive for CMT. Milk from these animals (222 quarter milk samples) when subjected to EC yielded 9.46% positive result. The SCC values were above 200000 cells/ ml for 128 samples.

The accuracy, sensitivity, specificity, PPV, NPV, PLR and NLP of the routine tests were calculated with the results of bacterial culture result as the standard. The results are given in the following tables.

Table 1: The three cow side diagnostic tests for determination of subclinical mastitis among dairy cattle

Tests	Samples	Positives	TP	FP	TN	FN	Accuacy(%)
CMT	222	115	98	17	22	85	54.05
EC	222	21	19	2	165	36	24.77
SCC	222	128	109	19	21	73	58.56

TP- true positive, FP- false positive, TN- true negative, FN- false negative

SCC was found to be more accurate (58.56%) than CMT (54.05%) and EC (24.77%). The sensitivity of SCC (59.89%) was higher as reported by Sharma *et al.* (2010) while EC showed more specificity (94.74%) which was in accordance with Langer *et al.* (2014). The **κ** (Kappa) values of all the three tests are below 0.4 indicating poor agreement with the gold standard test.

Table 2: Comparison of efficacy of the three tests.

Tests	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	PLR (%)	NLR (%)	κ	r, %
CMT	53.55	43.59	85.22	20.56	5.76	3.86	0.06	6
EC	10.33	94.74	90.48	22.34	9.5	4.58	0.16 9	16.9
SCC	59.89	52.5	85.16	17.91	5.73	3.48	0.08 8	8.8

PPV-Positive Predictive Value, NPV-Negative Predictive Value, PLR-Positive Likelihood Ratio, NLR-Negative Likelihood Ratio, κ-Kappa, r-Regression value.

Conclusion

From the above results there was 57.27% prevalence of subclinical mastitis in the herd. SCC was the test with highest accuracy and sensitivity while the kappa statistics analysis shows poor agreement with bacterial culture for all the three tests indicating the need for a more advanced technique of diagnosis.

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