

# **Bacteriological Testing of Milk Samples Suspected of Sub-Clinical Mastitis: A Case Study**

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**Abstract:** *There are two forms of mastitis viz., clinical and sub-clinical forms. The present article reports the microbiological investigation of a clinical case of mastitis in dairy cattle by following the proper dose regimen and schedule of recommended antibiotics for treatment. Mastitis is usually caused by bacteria that invade the udder, multiply and produce toxins which are harmful to the mammary gland. It remains the most economically important disease of dairy industries around the world producing great economic loss to farmers.*

**Keywords:** *Antibiotics, Antibiogram, Mastitis*

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## **1. INTRODUCTION**

The indiscriminate and injudicious administration of antibiotics and irrational treatment of bovine mastitis with different antibiotics have invited serious complications like multiple drug resistance. Mastitis is characterized by the chronic inflammation of the mammary gland of cattle and can have infectious and non-infectious etiologies. It is characterized by physical, chemical and usually bacteriological changes in the milk and pathological changes in the glandular tissue of the udder and affects quality and quantity of milk. Till date different types of antibiotics have been tried against the pathogens in bovine mastitis with or without identification and drug sensitivity testing.<sup>[1-5]</sup> The present study was conducted to test whether the suspected test milk samples were infected from sub-clinical mastitis and the antibiotics/ antibacterial drugs which may prove effective for its treatment.

## **2. MATERIALS AND METHODS**

Four (04) milk samples collected from the suspected four quarters of the udder of a crossbred cattle exhibiting clinical symptoms of mastitis were produced at the Teaching Veterinary Clinical Complex (T.V.C.C.) of Arawali Veterinary College. The collected milk samples were then forwarded to the Department of Veterinary Microbiology during March, 2017 for bacteriological investigation and reporting. The collected milk samples looked apparently normal in appearance.

The milk samples were examined bacteriologically<sup>[6]</sup> for the colony characteristics by nutrient agar plate culturing. Bacterial staining was done by Gram's Method<sup>[7]</sup>. The antibiotic sensitivity test was performed as per Kirby-Bauer antibiotic disc diffusion assay method on Mueller-Hinton agar plates with certain modifications<sup>[8-10]</sup> using antibiotic discs provided by the supplier (Microbes & Diagnostic & Research Centre, Ahmedabad, India). The concentration of antibiotic in each filter paper disc was as per the specification of the manufacturer required for laboratory purpose. Incubation of the petridishes layered with the agar containing antibiotic discs was done at 37°C for 24 h in a B.O.D. incubator installed at the department.

## **3. RESULTS AND DISCUSSION**

The overnight incubated nutrient broth cultures of the milk samples were subjected to spread plate culture on Nutrient agar media plates. After incubation at 37°C for 24 h it revealed the presence of circular, convex, glistening colonies with full regular edges on the agar media. Grams' staining revealed the presence of Gram positive cocci arranged in the form of clusters when examined under the high power magnification of the compound microscope. The bacteria was bacteriologically determined to be grouped under *Staphylococcus* spp.<sup>[6-10]</sup>

Antibiotic disc diffusion assay revealed the bacterial isolates to be highly sensitive to the minimum inhibitory concentration (MIC) of the antibiotics namely, ceftriaxone (30 mcg), tetracycline (30 mcg) and gentamicin (10 mcg) with moderate sensitivity to amoxicillin (10 mcg) and ceftazidime (30 mcg) respectively. The degree of sensitivity was determined on the basis of zone of inhibition produced by the isolated bacteria after exposure to the particular antibiotics and after comparison with the minimum inhibitory concentration of the respective antibiotic. The results obtained on cultural properties of the bacteria and their antibiotic disc diffusion assays revealed in the present study were in correlation with the findings of earlier investigations. <sup>[11-18]</sup>

#### 4. CONCLUSION

The present study revealed the presence of *Staphylococcus* spp. responsible for causing sub-clinical mastitis in dairy cattle. The bacteria was found to be sensitive to broad spectrum antibiotics which was reported and recommended to the T.V.C.C. for their administration in divided doses on alternate daily intervals in mixed preparations.

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