

## A Study on Detection of Pathogens in Milk of a Mastitis Effected Buffalo

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### ABSTRACT

Mastitis is a detrimental common problem in dairy industry which when left untreated is a potential health hazard and economic loss to farmer. The pathogens generally responsible for mastitis are Staphylococcus aureus, E. coli,

Streptococcus agalactia, Streptococcus dysgalactiae. Mastitis can be effectively treated by identification of appropriate antibiotic to be administered given the antibiotic resistance in field conditions. The objective of the paper includes detection of mastitis and etiological agent responsible. The milk sample was found positive for Staphylococcus aureus.

**Keywords:** Mastitis, pathogens, Staphylococcus aureus, antibiotic.

### I. INTRODUCTION:

Mastitis is the inflammation of mammary gland which may be infectious or noninfectious. Organisms such as bacteria, mycoplasma, yeast and algae can cause mastitis. Among all pathogens the most common cause of mastitis is bacteria. Mastitis pathogens have been classified as: Contagious or

environmental. The major contagious pathogens are: Streptococcus agalactia, Staphylococcus aureus, Mycoplasma.

Environment pathogens are: coliforms (E. coli, Klebsiella, Enterobacter), Streptococcus dysgalactia, Streptococcus uberis. Among all these, Staphylococcus is the chief etiological agent of mastitis in cattle and buffaloes. It can colonize in the skin as well as in the udder. Mastitis can be clinical or subclinical. Staphylococcus aureus is gram positive, anaerobic bacteria, catalase positive, oxidase negative. Production of coagulase by Staphylococcus is the important indication of pathogenicity.

A case of Buffalo (Breed-Graded Murrah) two months parturient and in its first lactation was presented to the local Veterinary Hospital with a chief complaint of swollen udder and teats since one week. Owner reported that there was a rapid reduction in milk yield. Flakes were noticed in milk. Milk sample was screened by California Mastitis Test for which it was found positive. The sample was further evaluated for presence of any pathogen.

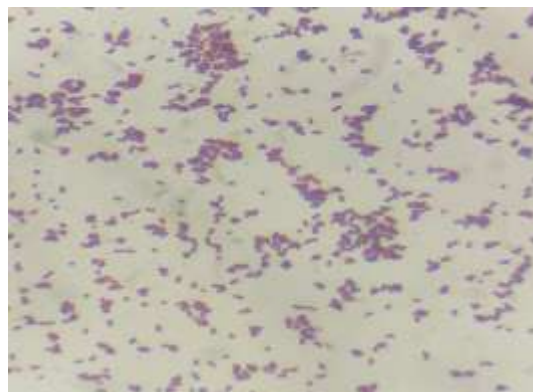
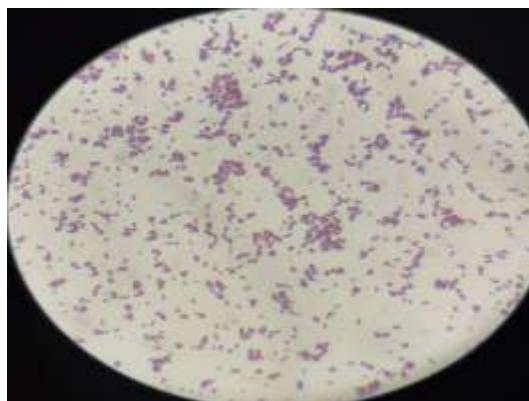


## II. MATERIALS:

Sterile glassware(Conical flask,Petridish).Sterile nutrient broth.Sterile media(Mannitol salt agar,Mueller Hinton agar).

### Procedure:

Nutrient broth was prepared and kept in bacteriological incubator at 37°C. A loopful of milk sample was inoculated onto nutrient broth . It was kept in bacteriological incubator for 24 hours to observe the desired bacterial growth. Upon Gram staining, Gram positive cocci were observed in the sample.



11.1-grams of Mannitol salt agar was mixed with 100ml of distilled water in a conical flask which was later autoclaved at 121°C,15lbs pressure for 15 minutes. The media was later transferred into petri plate in the laminar air flow near flame.The petri plates were incubated in incubator at 37°C for 24hours.

The following day,a loop full of culture was taken from nutrient broth and was streaked on Mannitol salt Agar media plate.This was later incubated at 37°C for 24 hours.

## III. OBSERVATION:

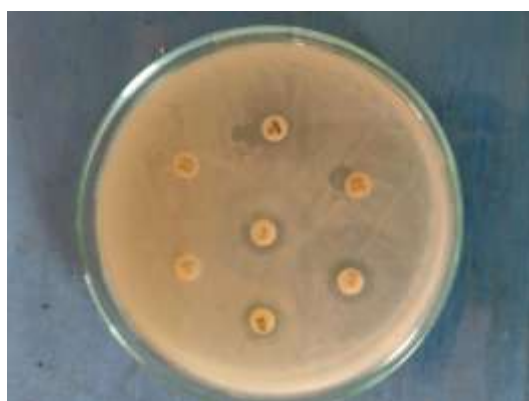
Yellow coloured pin point colonies were observed.



**Antibiotic sensitivity test:**

3.8 grams of Muller Hinton Agar (MHA) was mixed with 100ml of distilled water in a conical flask and was autoclaved at 121°C, 15 lbs pressure for 15 minutes. The media was then transferred onto petriplates in laminar airflow near flames. A loop full of bacterial culture was streaked on MHA media. Antibiotics discs were placed in the petriplate. The

discs used were of : Amikacin, Ampicillin, Enrofloxacin, Ofloxacin, Ceftriaxone, Cefixime, Cefotiofur, Penicillin, Tetracycline, Gentamicin, Ciprofloxacin. The petriplate was placed in bacteriological incubator at 37°C for 24 hours. By the following day, higher zone of inhibition was seen for Amikacin.



**IV. RESULT :**

From the above experiment, the pathogen was found to be bacteria *Staphylococcus aureus*.

## V. DISCUSSION:

Mastitis in dairy animals is the most important disease and poses a serious socio-economic impact worldwide. The inflammatory response in the udder of the dairy animals is called mastitis. Clinical mastitis can be detected by clinical examination such as inspection and palpation. At laboratory, mastitis can be detected by using California Mastitis Test (CMT) of milk sample from mastitis affected buffalo. *Staphylococcus aureus* is an important food borne pathogens that may be transmitted to flocks, an important mechanism for the spread of this organism.

In the current study, the prevalence and probabilities of isolating bacteria (*Staphylococcus aureus*) which is the main cause of mastitis in dairy animals, its association with animal (quarter) status, Antibiotic sensitivity test, were assessed while Amikacin has been recorded as the most effective antibiotic against isolated bacteria (*Staphylococcus aureus*) and Ofloxacin, Pefloxacin, Ciprofloxacin were found to be less effective against isolated bacteria from mastitis milk of the affected animal. This is due to increased indiscriminate and frequent use of those antibiotics which leads to the development of antibiotic resistant bacteria. Biosecurity against contagious mastitis pathogens such as pre-milking teat dip disinfection and post milking teat disinfection reduce the bacterial numbers on teat skin and spread of pathogens in the herd.

## VI. CONCLUSION:

The study was conducted to isolate the bacteria that cause mastitis in buffalo which was reported at the Local Veterinary Hospital. It was found *Staphylococcus aureus*.

## REFERENCES:

- [1]. Holt J, Krieg R, Senath A, Staley T, Williams, Bergey's manual of determinative Bacteriology. Hensley RW and Forlifer EL(eds). 9<sup>th</sup> edition. Williams & Wilkins Baltimore, USA, 1994.
- [2]. Sugiri, YD and Anri A 2010 prevalence of mastitis causing pathogens (*Staphylococcus aureus*, *Streptococcus agalactiae*) in small holder dairy farms. Research report of west Java livestock department.
- [3]. Hosseinzadeh S & Saei HD 2014. Staphylococcal species associated with bovine mastitis in the North West of Iran.
- [4]. Myllysv, Asplund K, Brofeldt E, Hirvela Koski V, Honkanen-Buzalskit, Juntilla J, et al. Bovine mastitis in Finland in 1988 and 1955 - changes in prevalence and antimicrobial resistance. PubMed abstract / Google scholar.
- [5]. Adkins PR, Middleton JR Laboratory Handbook on Bovine Mastitis: New Prague, MN: National Mastitis Council Inc. (2017).
- [6]. Dingwell RT, Leslie KE, Schukken YH, Sargeant JM, Timms LL. Evaluation of the California Mastitis test to detect an intramammary infection with a major pathogen in early lactation dairy cattle. Google Scholar.
- [7]. CLSI performance standards for Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria isolated from animals, Approved standard fourth edition Wayne, PA: CLSI document (2013) Google scholar.
- [8]. PJ Quinn, BK Markey, FC Leonard, ES Fitzpatrick, S Fanning, PJ Hartigan, Veterinary microbiology and microbial disease second edition. Wiley-Blackwell.
- [9]. Zadoks R, Fitzpatrick J. Changing trends in mastitis Ir. Vet J (2009). Google Scholar.
- [10]. D. Scott Mevey, Melissa Kennedy and M.M Chengappa. Veterinary Microbiology 3<sup>rd</sup> edition. Wiley-Blackwell.
- [11]. Prof. S.N. Sharma, Dr. S.C. Adlaka. Textbook of Veterinary Microbiology.