

## COMPARATIVE STUDY ON ISOLATION AND BIOCHEMICAL CHARACTERIZATION OF STAPHYLOCOCCUS AUREUS FROM MILK AND MILK PRODUCTS OF MATHURA

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### ABSTRACT

This investigation presents comparative data on detection of *Staphylococcus aureus* in milk products of Mathura & nearby region. A total of 30 samples including 5 each from milk vendors, household milk, dairy farm, Gaushala, butter and Dahi were tested for the presence of *Staphylococcus aureus* and identified by isolation using nutrient agar media, colony characteristics, morphological characteristics (Gram staining) and biochemical tests. Out of the 30 samples tested 8 (26.7%) were found to be *S. aureus* positive. Furthermore, Gaushala milk was having highest *S. aureus* contamination (60%) followed by dairy farm (40 %), milk vendors on bicycle (20%), household milk (20%), Dahi (20%) and butter (0%), respectively. The results of the present study showed that hygienic methods of milk collection should be adopted especially in Gaushalas and dairy farms to ensure contamination free milk and good health of consumers. However, a comprehensive study including large sample size and more accurate techniques like PCR are required to ascertain the actual confirmed status of *S. aureus* contamination in milk and milk products of this particular region.

**Keywords:** Nutrient agar, Gram staining, colony, morphological characteristics, PCR.

### INTRODUCTION

India's milk production has continuously increased because of its growing livestock population, better feedstock, and better breeds. Milk is supposed to constitute a complex ecosystem for various microorganisms including bacteria. There is an increased demand by the consumer for high quality natural food, free from artificial preservatives, and contaminating microorganisms. Contamination of milk and milk products with pathogenic bacteria is largely due to processing, handling and unhygienic conditions. The contaminants reach the milk products either

during cooking or handling after cooking<sup>1</sup>. A wide variety of bacteria can be involved, but the most common pathogen is *Staphylococcus aureus*. Despite implementing intensive control measures, it is difficult to eradicate the intramammary infections caused by this pathogen and it remains a substantial economic problem<sup>2</sup>. Enterotoxigenic *S. aureus* in raw milk possess a potential health hazard to consumers and the identification of such strain should be used as a part of analysis of milk and milk products<sup>3</sup>. Illness through *S. aureus* range from minor skin infection such as pimples, boils, cellulitis, toxic shock syndrome, impetigo, and abscesses to life threatening disease such as pneumonia, meningitis, endocarditis, and septicemia<sup>4,5</sup>. Considering the economic loss and zoonotic importance of *S. aureus* the present study was designed to detect the prevalence of *Staphylococcus aureus* contamination in bovine milk samples of different sources and milk products viz. Dahi, butter of Mathura and nearby regions.

### MATERIALS AND METHODS

**Collection of samples:** A total of 30 milk samples and milk products were collected from different sources (viz; Milk vendor on cycle, Household milk, Dairy farm, Gaushala, Butter and Dahi) from Mathura and nearby regions. 5 samples of each product (5ml) were collected aseptically, transferred to sterile test tubes and were transported to the biotechnology laboratory, Mangalayatan university, Beswan, Aligarh under cold conditions. They were stored at 4 °C and tested within 24 hours for isolation and identification of *Staphylococcus aureus*.

**Isolation and identification of *Staphylococcus aureus*:** Each sample was enriched in Peptone water and incubated at 37°C for 24 hours. Each inoculum was cultured on Nutrient agar medium and again incubated at 37°C for 48 hours. The

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colonies appeared as pale yellow colored. Single isolated colony was then picked and streaked on Nutrient agar slant and further the organism was identified on the basis of their cultural, morphological, staining and various biochemical characteristics<sup>6</sup>. All positive samples were subjected to coagulase test for confirmation of *Staphylococcus aureus*<sup>7</sup>.

managerial conditions of milk producing animals<sup>11</sup>.

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**Table-1: S. AUREUS CONTAMINATION OF VARIOUS COLLECTED SAMPLES OF MATHURA AND NEARBY REGIONS**

S. No.	Type of sample collected	No. of tested samples	No. of Positive samples	% positive sample
1.	Milk vendor on cycle	5	1	20%
2.	Household milk	5	1	20%
3.	Dairy farm milk	5	2	40%
4.	Butter	5	0	0%
5.	Dahi	5	1	20%
6.	Gaushala milk	5	3	60%
	Total	30	8	26.7%

### RESULTS AND DISCUSSION

Table 1 shows the sampling data which consists of different numbers of samples analyzed and the number *S. aureus* isolates out of the total number of samples tested. In the present investigation, out of all the milk samples tested the highest *S. aureus* contamination was found to be in the milk collected from Gaushala (60%) followed by dairy farm (40%). The milk samples from bicycle vendors and household showed same contamination (20%). This may be due to unhygienic conditions in Gaushalas and dairies and improper handling of milk<sup>8</sup>. In case of milk products, the highest contamination was recorded in Dahi (20%) followed by Butter (0%), suggesting that it could be due to contaminated environment and unhygienic handling or preparation. Out of the total 30 samples tested 8 (26.7%) were found to be contaminated with *S. aureus*. However, in case of Brazilian Dairy farm earlier published reports<sup>9</sup> revealed slightly lower *S. aureus* contamination (10.8%) in bulk milk samples. Similar to our findings Priyanka and Alka<sup>10</sup> also could not detect *S. aureus* contamination in Dahi samples of Agra region. Recent findings have suggested differential *S. aureus* contamination in various products i.e., 13.33% each in milk samples from bicycle vendors and Dairy farm, 6.67 % each in household milk, Dahi and butter<sup>1</sup>. The difference in results might be chiefly due to the variation in the number of samples analyzed and the difference in

### REFERENCES

1. Rajeev Kumar and Amit Prasad, 2010. Detection of *E. coli* and *Staphylococcus* in Milk and Milk Products in and around Pantnagar. *Veterinary World*, 3(11):495-496.
2. Salmon, S.A., 2002. Use of antimicrobial susceptibility data to assist in determining the best therapy for clinical mastitis. *Proceedings of the NMC 41st Annu Mtg., Orlando, FL. National Mastitis Council Madison, W.I., pp: 36.*
3. Zouharova M and Rysanek D., 2008. Multiplex PCR and RPLA Identification of *Staphylococcus aureus* enterotoxigenic strains from bulk tank milk. *Zoonoses Public Health*, 55(6):313-319.
4. Soomro, A.H et al., 2003. Isolation of *Staphylococcus aureus* from Milk Products Sold at sweet-meat Shops of Hyderabad. *Online Journal of Biological sciences*, 3(1):91-94.
5. Masud, T. Ather, H.I. Azhar Chushti, M. Amim Shah, M., 1988. Microbiological studies on indigenous dahi with special reference to public health. *Aust. J. Dairy Technology*, 8-13.

6. Cruickshank, R., 1970. Medical Microbiology. 11th Edn., The English Language Book Society and E. and Livingston Ltd., UK.
7. Monica, C., 1991. Medical Laboratory Manual for Tropical Countries. Vol. II, ELBS, USA., pp:60-63.
8. Sharma, S.D., 1982. Studies on bovine mastitis with special reference to mycotic infections of udder. Thesis submitted to CSA univ. Kanpur (U.P.).
9. Helena Fagundes, Luciana Barchesi, Antonio Nader Filho, Luciano Menezes Ferreira, Carlos Augusto Fernandes Oliveira, 2010. Occurrence Of *Staphylococcus aureus* In Raw Milk Produced in Dairy Farms in Sao Paulo State, Brazil. Brazilian Journal of Microbiology, 41:376-380.
10. Priyanka Singh and Alka Prakash, 2008. Isolation Of *Escherichia coli*, *Staphylococcus aureus* And *Listeria monocytogenes* From Milk Products Sold Under Market Condition At Agra Region *Acta agriculturae Slovenica*, 92(1), 83–88.
11. Blood, D.C. and Henderson, J.A. and Radostits, O.M. , 1979. Veterinary medicine, Ed. V ELBS & B.T.