ISOLATION AND BIOCHEMICAL IDENTIFICATION OF *E. COLI* FROM WASTE EFFLUENTS OF A DAIRY INDUSTRY

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ABSTRACT

Microbial assessment of waste effluents from dairy industry revealed the presence of gram positive and gram-negative bacteria. Maximum percentage of *E.coli* was observed in milk and yogurt waste as compared to cheese and butter waste effluent. There is significant difference in number of bacterial colonies among dairy waste effluents. Some strains of *E.coli* are pathogenic and can cause serious health effects to humans by direct or indirect waste such as through water bodies to which waste effluent is discharged.

Key words: Waste effluents, E. coli, Dairy product,

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INTRODUCTION

Industrialization had a pivotal role in the developmental strategies due to its significant contribution to the economic growth and human welfare. Industrialization, like other human activities with an impact on the environment, often results in pollution and degradation. It carries certain costs and problems in terms of pollution of the air, water resources and general degradation of the environment (Suflita *et al.*, 1983; Thomas *et al.*, 1992). Industries turn out wastes, which are uncharacteristic in terms of type, volume and frequency depending on the type of industry and population that uses the product (Odumosu, 1992). Wastewater originated from dairy operations may wharf human pathogens including *Escherichia coli*

The extent of the problem of bacterial contamination justify more elaborative studies from the point of production of milk and milk products to the point of consumption including all intermediary levels. The aim of the present research was to identify the presence and identification of *E.coli* from dairy waste effluents.

METHOD

Sample Collection and Processing

Different samples collected in two successive months which includes waste effluents of butter, milk, yogurt and cheese, released after processing of milk for making different dairy products All samples were kept at 4°C immediately after taking from site.

A 'Theodor Escherich 1885' technique was used for the isolation of *E.coli*. Area was swabbed with 70% ethanol prior to opening any sample container. The samples (0.5 ml) were taken in 10 ml Lauria Bertini (LB) broth medium in autoclaved test tube and vortexed for one minute until a homogenized mixture was obtained. It was then kept for thirty minutes at room temperature. After half an hour, supernatant (1ml) was taken from the test tube with the help of micropipette and added the supernatant into another sterile test tube containing 9 ml autoclaved water in order to make the 2-fold dilution. Then vortexed the dilution tube, 500 microliter from the final dilution tube were spreaded on the Petri dishes of MacConkey medium and

LB medium using spreader. Petri dishes were kept in the incubator for 24 hours at 37°C. After 24 hours, plates were studied for the colonies of microbes grown on the media. Different biochemical tests were carried out for the identification of E.coli including Indole test, Indole spot test, Kovac's indole test, Voges-Proskauer test and Simmon's test.

RESULT AND DISCUSSION

E.coli is a common gut occupant in the gastrointestinal tracts of almost all mammals. It is usually outnumbered 100-fold or more by strictly anaerobic gut bacteria (Weber *et al.*, 2005). *E.coli* causes a variety of diseases in both humans and animals (Russo and Johnson, 2000). It is evident that Shiga toxic Escherichia Coli (STEC) causes human diseases, ranging from uncomplicated diarrhea to life-threatening complications such as the Hemolytic uremic syndrome. Several studies have shown a high prevalence of STEC, with a wide variety of serotypes in animals and food products. However, very few serotypes have been associated with human disease (Pradel *et al.*, 2008).

E.coli from various selected samples was cultured on LB medium and MacConkey medium for morphological characterization. After 24 hrs the examination under microscope isolated two types of colonies on the basis of colony characteristics. These colonies were pink in color while creamy yellow in LB medium. *E.coli* grown on MacConkey Agar was capable of metabolizing lactose, which produces acid by-products that lower the pH of the media.

The highest numbers of *E.coli* isolates were found in milk and yogurt samples. The waste effluent milk collected in March from the local brands (packed in commercial non-tetra pack) and butter samples collected from local bakeries. The sample of waste effluent yogurt collected in April from the local brands (packed in commercial non-tetra pack) and cheese waste effluents collected from local bakeries. *Escherichia* species were found in 49 out of 64 samples studied.

The average value of gram-negative bacteria in waste effluents of milk, butter, yogurt and cheese was 4.81 ± 0.35 , 3.37 ± 0.30 , 4.56 ± 0.32 and 4.00 ± 0.22 respectively.

The results of milk samples showed that more than 50% samples (57 out of 100) were contaminated with *E.coli* and confirmed the works of Martin and his colleagues, 1986. The results of the milk product samples revealed that highest numbers of milk products were contaminated with *E.coli*. Two cases of pediatric HUS investigated in 1986 were the first to be associated to raw milk consumption (Martin *et al.*, 1986) and confirmed that cow's milk as a vehicle for *E.coli* 0157:H7 infection.

CONCLUSION

Microbial assessment of waste effluents from dairy industry confirmed the presence of certain microorganism. Maximum percentage of *E.coli* was observed in milk and yogurt waste effluents and lowest percentage was observed in cheese and butter waste effluent, which can cause serious health effects to human.

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