

ANALYSIS OF DAIRY PACK FOOD FOR PRESENCE OF BACTERIAL PATHOGENS

Zagare M.S., Deshmukh A.M. *and Patil S.S.

Department of Environmental Science, Dr. Babasaheb Ambedkar Marathwada University,
Aurangabad. Pin - 431001 (M.S.), India.

*Department of Microbiology, Dr. Babasaheb Ambedkar Marathwada University (Subcentre),
Osmanabad-413501, India

E-mail: amdeshmukh1@rediffmail.com

ABSTRACT

Milk is a complete food and dairy products becoming very popular in our society. There are number of packed dairy foods available in market e.g. milk powder, milk, cheese, curd and ice cream etc. Study was carried out by using various microbiological techniques to isolate and identify pathogens. From 41 packed food samples Three isolates of *Escherichia coli* and seven isolates of *Salmonella typhi* and One isolate of *Staphylococcus aureus* were obtained. Thus it seems that dairy packed food samples may cause food borne illness.

KEY WORDS: Dairy products, *Escherichia coli*, *Salmonella typhi*, *Staphylococcus aureus*

INTRODUCTION

Over the last few years food poisoning and food safety have become very topical subject, eliciting a great deal of public concern to many people all over the world. This is a result of emerging food borne pathogens that continue to cause outbreak of food borne diseases caused by eating food contaminated with pathogenic microorganisms or their products (Lefoka, 2009). Milk is a highly nutritious food ideal for microbial growth and the fresh milk easily deteriorates to become unsuitable for processing and human consumption (FAO, 2001). High bacteria counts are indicators of poor production, hygiene or ineffective pasteurization of milk (Harding, 1999). Milk and milk products derived from dairy cows milk can harbor a variety of microorganisms and can be important sources of food borne pathogen (Oliver *et al.*, 2005; Yagoub *et al.*, 2005). The presence of food borne pathogens in milk is due to direct contact with contaminated source in the dairy farm environment and to excretion from the udder of an infected animal (El-Zubeir *et al.*, 2006).

In recent times the dairy industry has been concerned with the presence of this pathogen in dairy products with common sources of contamination as hands and clothing of food processor and dirty surfaces of equipment. A broad spectrum of microbial pathogens can contaminate human food and water supplies and causes illness after they or their toxins are consumed. These include a variety of enteric bacteria, aerobes and anaerobes, viral pathogens and yeasts. (Tauxe *et al.*, 2002). During past decade microorganisms such as *staphylococcus aureus*, *salmonella spp.*, *Escherichia coli*, *shigella spp.* were reported as the most common food borne pathogens that are present in many foods and able to survive in milk and fermented milk products. (alm *et al.*, 1983; Ahmad *et al.*, 1986; Ryser *et al.*, 1988; Schaak *et al.*, 1988; Canganella *et al.*, 1998). In the US reports have show that food borne illness accounted for approximately 76 million illnesses, 325000 hospitalizations and 5000 death each year (Mead *et al.*, 1999). Food borne disease surveillance began in the US in the early 1900s response to morbidity caused by milk transmitted typhoid fever and infantile diarrhea (Cliver *et al.*, 1990). In the present study identification of pathogenic bacteria from the dairy products were carried out to know its hazardous effect and its microbial quality. Pathogenic bacteria in milk have been a major factor for public health concern since the early days of the industry. Many diseases are transmissible via milk products. Traditionally raw or pasteurized milk has been a major vehicle for transmission of pathogens. The source of contamination by microorganisms is unclean teats. The use of unclean milking and transport equipment also contributed to the poor hygienic quality. (Parekh and Subhash, 2008).

MATERIALS AND METHODS

Collection of samples:

41 Samples of dairy products like Milk packet, Milk powder, Shrikhand, Amrakhand, Ice cream, Curd and Chocobar were collected from the market Packed dairy food samples were directly transported to the laboratory in ice box. They were stored in refrigerator and analyzed within 24 hours. Out of 41 samples 13, 3, 8, 11, 3, 1, 1, 1 packed food samples were milkpackets, milk powder, Amrakhand, Shrikhand, Ice cream, ghee, chocobar, curd sample respectively.

Microbiological analysis

A portion (1 g or 1 ml) from each sample was taken aseptically and diluted in 9 ml sterile distilled water
The diluted sample was streak inoculated on sterile selective media as given below

1. Eosine Methylene Blue (EMB) for *Escherichia coli*
2. Mannitol Salt Agar (MSA) for *Staphylococcus aureus*
3. Wilson's and Blair (W& B) for *Salmonella typhi* Inoculated petriplates were incubated at 37 °C for 24 hours.

Identification of pathogens

1. Cultural characteristics, gram nature and colour of colonies were noted.
2. Biochemical examination colonies from each petriplate were picked, subcultured, incubated at 37 °C and then identified by the various biochemical tests.

Biochemical tests were performed to confirm *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi* using, catalase test, indole test, methyl red test, voges prousker test, nitrate reduction test, urease production, citrate utilization test and glucose, lactose, mannitol, sucrose sugar fermentation test.

RESULTS

Morphological

Gram and cultural characteristics of *Escherichia coli*, *Salmonella typhi*, and *Staphylococcus aureus* were studied and results are given in Table- 1. Biochemical test for identification of *Escherichia coli*, *Salmonella typhi*, *Staphylococcus aureus* were performed and results are given in Table- 2. Out of 41 bacterial isolates 3 isolates were confirmed as *Escherichia coli*, 7 isolates were confirmed as *Salmonella typhi*, 1 isolate was confirmed as *Staphylococcus aureus*.

Table: 1. Morphological and culture characteristics of isolated pathogens

Sr.No	Isolated pathogen	Gram staining and morphology	Culture characteristics on selective media
1	<i>Escherichia coli</i>	Gram –ve coccobacilli	Colonies showing green metallic sheen.
2	<i>Staphylococcus aureus</i>	Gram +ve cocci	MSA medium turns red to yellow
3	<i>Salmonella typhi</i>	Gram –ve rod shaped	Black colonies on Wilsons and Blair medium.

Table:2. Biochemical characterization of isolated pathogens

Sr.No.	Biochemical test	Results for		
		<i>Escherichia coli</i>	<i>Salmonella typhi</i>	<i>Staphylococcus aureus</i>
1	Catalase	+	–	+
2	Urease	–	–	–
3	Oxidase	–	–	–
4	Coagulase	–	–	+
5	Citrate utilization	–	+	–
6	Nitrate reduction	–	–	+
7	Indole production	+	–	–
8	Methyl red	+	+	+
9	Voges prousker	–	–	+
10	Glucose	+	+	+
11	Lactose	+	–	+
12	Mannitol	+	+	+
13	Sucrose	+	–	+
14	Maltose	–	+	+

Table: 3 . Distribution of various packed food samples on the basis of bacterial pathogen

Sr. No.	Type of Sample	Total no. of samples	Number of samples showing presence of pathogen		
			<i>Escherichia coli</i>	<i>Salmonella typhi</i>	<i>Staphylococcus aureus</i>
1	Milk packet	13	01	04	
2	Milk powder	3		01	
3	Amrakhand	8	01	01	
4	Shrikhand	11	01	01	
5	Ice cream	3			
6	Ghee	1			
7	Chocobar	1			01
8	Curd	1			

DISCUSSION

The presence of bacteria like *Escherichia coli*, *Staphylococcus aureus*, *Salmonella typhi* in milk suggested contamination from various sources such as animal teats, udder etc. unclean hands of worker, environmental dust and microbes, unclean utensils in dairy, and others. (Murphy *et al.*, 2000). The pathogens in the food also multiply and grow in large number. In the present study the results obtained from the microbial analysis of milk and milk products viz. milk packets, milk powder, Amrakhand, Shrikhand, Ice cream, Ghee, Chocobar and Curd showed that milk and milk products were contaminated with microorganisms of public health concern. The distribution of various packed milk food samples on the basis of presence of pathogens is given in Table-3. It was found that Out of 13 milk packets samples, 1 sample was contaminated with *Escherichia coli* and 4 sample were contaminated with *Salmonella typhi*. Out of 3 milk powder samples, 1 sample was contaminated with *Salmonella typhi*. Out of 8 amrakhand samples, 1 sample was contaminated with *Escherichia coli* and 1 sample was contaminated with *Salmonella typhi*. Out of 11 shrikhand samples, 1 sample was contaminated with *Escherichia coli*, 1 sample was contaminated with *salmonella typhi*. Out of 3 ice creams and 1 ghee sample there was no any contamination found. In chocobar sample 1 sample was contaminated with *Staphylococcus aureus*. *Escherichia coli* was isolated from milk products like mawa, khoa, cream, dahi, cheese, butter and Gulabjaman (Bhat *et al.*, 1948; Kumar *et al.*, 1989; Kulshreshtha *et al.*, 1990).

The presence of coli form or bacteria in milk and milk products may be due to the low level of hygiene maintained during the processing and sale of the products, this includes handlers hand, quality of water used and the utensils used in the dairy farm. The exposure of milk and milk products displayed for sale in bowls and packet can serve as a source of contamination (Roseline *et al.*, 2006).

Escherichia coli in milk products may indicate faecal contamination. Their presence indicates poor hygienic practices among handlers of milk products. Contaminated packed food samples may cause many bacterial food borne diseases. Hygiene measure assume a decisive importance in food safety management (Untermann, 1998). *Staphylococcus aureus* in milk and milk products indicates the spoilage of milk and milk products. At even refrigerator temperature bacteria release toxic chemicals in contaminated food products and cause symptoms of illness in consumer.

Salmonella typhi in milk and milk products cause typhoid fever. Various internal organs such as spleen and liver becomes infected by *salmonella spp.* presence of *salmonella spp.* indicate contamination due to water used in dairy farm. The isolation of coliform and other food pathogens from these dairy products pose a serious threat to food safety, especially, locally processed foods which are consumed without further processing, great attention should therefore be given to the microbiological safety of these products because their direct consumption may cause health hazard to the consumer (Roseline *et al.*, 2006).

Isolation of bacterial pathogen from dairy farms and from outbreaks of human disease substantiates the hypothesis that packed food samples are reservoir of food borne pathogens (Oliver *et al.*, 2005). It is therefore; critically important to ensure high quality milk production from healthy animal, therefore it is recommended that training should be given to farm owner and worker responsible for milking process.

REFERENCES

- Ahmed A.H., Moustafa M. and El-Bassiony T.A. (1986). Growth and Survival of *Yersinia enterocolitica* in yoghurt. *J. Food Prot.* 4: 983-985.
- Alm L. (1983). Survival rate of salmonella and *shigella* in fermented milk products with and without added gastric juice: an invitro study. *Food Nutr. Sci.* 7:19-28.
- Bhat J.V., Sethna K. and Fernandes F. (1948). *Ind. J. Dairy Sci.* 49.
- Canganella F., Ovidi M., Paganini S., Vettraino A. M., Bevilacqua L. and Trovati L. D. (1998). Survival of undesirable microorganisms in fruit yoghurts during storage at different temperatures. *Food Microbiol.* 15: 71-77.
- Clover O. D. (1990). Foodborne Diseases. Academic press, Inc. San Diego, California 92101.
- FAO. (2001). The lactoperoxidase system of milk preservation. Regional lactoperoxidase Workshop in west Africa. *Burkina Faso.* 17-19.
- Griffin P.M. and Tauxe R.V. (1991). The epidemiology of infection caused by *Escherichia coli* 0157:H7, other enterohemorrhagic *E-coli* and the associated hemolytic syndrome. *Epidemiol. Rev.* 13; 60-97.
- Harding F. (1999). Milk Quality . A chapman and Hall Food Science Book. An Aspen Publication. First edition.
- Kulshreshtha S.B. (1990). Prevalence of enteropathogenic serogroups of *E.coli* in milk products samples from Bareilly and their multiple drug resistance. *Ind. J. Dairy Sci.* 43:337-378.
- Kumar, V. and R.N. Sinha, (1989). Incidence of coliforms in indigenous milk products. *Ind. J. Dairy Sci.* 42:579-580.
- Lefoka (2009). The survival of microbial pathogens in dairy products. M.sc. Dissertation, Univ. of Free State, Bloemfontein. 1-157.



- Leyer G. J., Wang, L. and Johnson E.A. (1995).** Acid adaptation of escherichia coli O157:H7 increases survival in acidic foods. *Appl. Environ. Microbiol.* 61: 3752-3755.
- Mead P. S., Slutsker L., Dietz V., McCaig L.F., Bresee J. S. and Shapiro C. (1999).** Food- related illnesses and death in the United State. *Emerg. Infect. Dis.* 5: 607-625.
- Muhamed Mubarack H., Doss R. Dhanabalan. and Balachander S. (2010).** Microbial quality of raw milk samples collected from different villages of Coimbatore District, Tamilnadu south India *Ind. J. Sci. Tech.* 3:1-5.
- Oliver S. P., Jayarao B. M. and Almeida R. A. (2005).** Foodborne pathogens in milk and the dairy farm environment: food safety and public health implications. *Foodborne Pathogenic Dis.* 2 (2): 115-129.
- Roseline E., Uzeh Regina E., Ohenhen, Ayodeji K. and Rojughokan. (2006).** Microbiological and nutritional qualities of dairy products: Nono and Wara. *Nature and Sci.* 4(3).
- Ryser E. T. and Marth E.H. (1988).** Survival of *Listeria monocytogenes* in coldpack cheese food during refrigerated storage. *J. Food Prot.* 51: 615-621.
- Sohaak M. M. and Marth E. H. (1988).** Behavior of *Listeria monocytogenes* in skim milk and in yoghurt mix during fermentation by thermophilic lactic acid bacteria. *J. Food Prot.* 51, 607-614.
- Utermann F. (1998).** Microbial hazards of food. *Food Controll.* 9:119-126.
- Yagoub Sanna O., Nazik E., Awadalla and Ibtisam E.M. and Zubeir EI. (2005).** Incidence of some potential pathogens in raw milk in Khartoum North (Sudan) and their susceptibility to antimicrobial agents. *J. Anim. Vet. Advances.* 4(3): 356-359.
- Zubeir, E. I., Ibtisam E. M., Wigdan, M. Abdalla and EI Owni O.A.O. (2005).** Chemical composition of fermented milk (roub and mish) in Sudan. *Food Control.* 16: 633- 637.