Study the incidence of *Bacillus cereus* isolates from dairy foods

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Abstract

Among 200 selected dairy samples 50 each of pasteurized milk, cream, butter and paneer were analyzed for the presence of *Bacillus cereus* the paneder samples were found to have the highest incidence of *Bacillus cereus* (52.94%) followed by cream sample (29.41%) and pasteurized milk sample (17.64%) however none of the butter samples were found to be contaminated with *Bacillus cereus*. These isolates were found to be gram positive bacilli with spores, they were found to ferment sucrose, glucose, dextrose; they gave the positive result for NO3 reduction, oxidize, catalase, and for starch hydrolysis.

Keywords: *Bacillus cereus*, Pasteurized milk, Butter, Cream.

1. Introduction

The genus *Bacillus* is comprised of a diverse array of Gram-positive, aerobic and facultative anaerobic and facultatively anaerobic endospore forming rods. *Bacillus* species are found in a wide range of habitats, and include species possessing environmental, industrial and clinical significance. Many species are common soil inhabitants and may frequently contaminate foods, including dairy products, meats, infant food, rice dishes, vegetables, spices and cereals (Christiansson et al., 1999) [9]. Members of the *Bacillus* genus are ubiquitous soil microorganisms and are generally considered harmless contaminants. The vegetative cells range from 0.5 by 1.2 to 2.5 by 10 µm in diameter and can grow at optimal temperatures ranging from 25 to 37 °C, although Thermophile and psychrophilic members are capable of growth at temperatures as high as 75 °C or as low as 3 °C. Some species can flourish at extremes of acidity and alkalinity, ranging from pH 2 to 10. *Bacillus* species play a very important role in the keeping quality of milk and dairy products. These organisms survive heat treatment and high temperature used for processing of the products, activates spore germination and outgrowth, resulting in spoilage of products. *Bacillus cereus* is important as it affects the shelf life of pasteurized milk and heat treated dairy products. The organism is associated with defects such as off flavours, sweet curdling and bitty cream caused by proteinase, lipase and phospholipase enzymes (Meer et al., 1991) [15]. In addition to these defects in dairy products, *B. cereus* has also been associated with outbreaks of food poisoning (Johnson1984 [12] Kramer and Gilbert, 1989) [13].

*Bacillus cereus* is a widely distributed bacteria, have been isolated from rice, spices, meat, and egg and dairy products. The presence of *Bacillus cereus* spores in milk is unavoidable. This pathogen produces both a toxin and spore capable of surviving pasteurization. The growth of *B. cereus* significantly spoils the quality of dairy products, causing sweet curdling and "bitty" cream.

*Bacillus cereus* causes problems in the food industry both by deteriorating the products (TeGiffel et al., 1996; [20] Pirjarvi et al., 2000; [16] Eneroth et al., 2001), [8] and by endangering people’s health upon consuming contaminated foods (Granum et al., 1993; [10] Ghelardi et al., 2002) [9]. Under certain conditions, strains of this species produce haemolysins, phospholipases C, also emetic toxins and enterotoxins that cause food poisoning (Rusul and Yaacob, 1995; [17] Andersen-Borge et al., 2001; [2] Agata et al., 2002) [1]. It causes two different types of food poisoning: the diarrhoeal type and the emetic type. The diarrhoeal type of food poisoning is caused by different enterotoxin complexes produced during the growth of *Bacillus cereus* in the small intestine while the emetic toxin is produced by the growing cells in food. The organism has ability to adapt to different chemical (Willinghan et al., 1996; [24] Ulette et al., 2002) [22]. Heat (Brown and Dowds, 2001) [3] and cold environment (TeGiffel et al., 1997), [21]
as well as by toxin production (Andersen-Borge et al., 2001)
[2]. The ability of spores of B. cereus to resist against the high
pasteurization temperature, which very frequently
contaminates the dairy products. The food poisoning caused by
B. cereus is categorized by a number of different syndromes
(emetic and diarrheal) with a variable intensity (Christian-Son
et al., 1989) [4]. B. cereus produces a number of toxic products
that are important virulence factors and participate in the
course of foodborne human gastrointestinal diseases.

2. Materials and Methods
2.1 Place of Work
The present study entitled “Study the incidence of Bacillus
cereus isolates from dairy foods” was conducted in the lab
of Dairy Microbiology, Warner school of Food & Dairy
Technology, Sam Higginbottom Institute of Agriculture,
Technology and Sciences Deemed- to – be University,
Allahabad.

2.2 Collection of Sample
Two hundred samples of dairy products were collected which
include 50 samples each of cream, butter, paneer and
pasteurized milk from the local market of Allahabad and
Student’s Dairy in Sam Higginbottom Institute of Agriculture,
Technology and Sciences Deemed- to – be University,
Allahabad.

2.3. Isolation of Bacillus cereus
Isolation of Bacillus cereus was performed by streaking the
samples on nutrient agar. These plates are incubated at 37°C for
24-48 hrs.

2.4 Identification of The Isolates
The isolates observed from the nutrient agar were identified on
the basis of cultural, morphological and biochemical
characteristics given in Bergey’s Manual of Systematic
Bacteriology (Holt et al., 1984) [11],

3. Results and Discussion
3.1 Incidence of Bacillus cereus
Among the 200 dairy samples which include 50 samples each of
Paneer, Pasteurized milk, Cream and Butter sample, screened for growth of Bacillus cereus on nutrient agar plate,
17 (8.5%) Bacillus cereus was isolated. The incidence of
Bacillus cereus was 17 (8.5%) in which 9 (52.94%) were
isolated from Paneer sample, 3 (17.64%) were isolated from
Pasteurized milk sample, 4 (29.41%) were isolated from
Cream sample, and no Bacillus cereus isolated from Butter
sample. However a lower incidence of B. cereus was recorded
in the study of Wong et al., (1987) [25] and Schlegelove et al.,
(2003) [18]. In comparison to the incidence of B. cereus
recorded in the selected dairy products in present study a
higher incidence was reported in the study of Schlegelove et al.
(2003) [18] in spreading butter (65.07%). In the present study a
higher incidence of B. cereus was recorded which was in
agreement with the studies of De Santis et al., (2008) [7].
However Vaishnavi et al., (2001) [23] reported contrasting
results. The higher incidence of B. cereus in pasteurized milk
could be attributed to the contamination of teats with soil
(Christiansson et al., 1999), [5] or the entry of B. cereus spores
in raw milk through air after pasteurization process (Lin et al.,
1998) [14]. Similarly the higher level of contamination with B.
cereus in cream and paneer sample tested in the present study
could be occurred due to poor processing condition (TeGiffel
and Beumer, 1998). [19] However, none of the butter samples
tested showed the presence of B. cereus. Since the high salt-
in moisture content is found in butter, it would have had the cidal
effect on the cells (Champagne et al., 1994), [6] or it may be
due to over pasteurization of the samples also during the
homogenization of spreading butter, and 65-70°C, the
contaminating micro flora is eliminated (Schlegelove et al.,
2003) [18].

Table1: Incidence of Bacillus cereus from selected dairy samples

<table>
<thead>
<tr>
<th>Total samples</th>
<th>No. of isolates</th>
<th>Incidence of bacillus cereus (%)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Pasteurized milk</td>
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<tr>
<td>200</td>
<td>17 (8.5%)</td>
<td>3(17.64%)</td>
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</tbody>
</table>

Fig 1: Distribution of Bacillus cereus in different dairy products
3.2. Conclusion
In conclusion the fairly higher incidence of Bacillus cereus recorded in the different dairy products is quite alarming as these organisms are causative agents of food poisoning and spoilage. It is a major threat in food industry both by deteriorating the products and by endangering people’s life upon consumption. Additionally, the products contaminated by these organisms, may be, vectors of resistance to antibiotics. Since it is of vital concern, it is highly recommended that proper control measures during various stages of production and processing should take to control the entry of this ubiquitous organism in the dairy industry. It is further emphasized that implementation of HACCP in the different dairy industry is the best way to assure the lowest possible risk of Bacillus cereus.

4. References