

# EFFICACY OF AMALGAMATION OF HERBS ON GROWTH AND SURVIVAL OF YOGHURT CULTURE ON FREEZING

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## KEYWORDS

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## ABSTRACT

The present research was planned with an aim to increase the viable count of yoghurt culture and to maintain the viable count in the final product by protecting the yoghurt culture from adverse effect of freezing and subsequent storage at low temperature. The product was fermented with traditional yoghurt culture (*Streptococcus salivarius ssp. Thermophilus*, *Lactobacillus delbrueckii ssp. Bulgaricus*) and formulated with different levels of herbal extract at 0.5%, 1.0%, 1.5% and 2.0% for experimental treatment T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> respectively with 10° Brix. On the basis of selective enumeration, herbal extract with the level of 1.5% in frozen yoghurt has maximum viable count (cfu/g) followed by 1.0%, 2.0% and 0.5%.

## INTRODUCTION

The nutritional and potentially therapeutic value of food is a key characteristic in the development of new value-added products manufactured for health conscious consumers (Garcia *et al.*, 1998; Corte, 2008). Frozen yoghurt has been the fastest growing product in the frozen dessert market in recent years (Opdahl and Baer, 1991; Guinard *et al.*, 1994). Frozen yoghurt dessert is a complex fermented frozen dairy dessert that combines the physical characteristics of ice cream with the sensory and nutritional properties of fermented milk products. Yogurt, which is consumed by a wide cross section of people throughout the world, has an established market as a functional therapeutic food (Adhikari *et al.*, 2000). Frozen yoghurt is a low acid product (Schmidt *et al.*, 1997) and can be regarded as a healthy alternative to ice cream for people suffering from obesity, cardiovascular diseases and lactose intolerance due to its low fat content (the fat percentage of regular frozen yoghurt ranges from 3.5% to 6%) (Milani and Koocheki, 2011).

In the present scenario, the herbal products are gaining more popularity over synthetic products in the world market. This is occurring due to some side effect of synthetic products on the body (Amirdivani, 2008). In spite of well-practiced knowledge of herbal medicines and occurrence of a large number of medicinal plants, the share of India in the global market is not up to the mark (Rajkumar, 2009).

Yoghurt contains substantial amounts of live lactic acid

bacteria (Ghosh and Raymond, 2003). However, numbers of viable yogurt cultures differ widely among brands of frozen yogurt, for the retention of the usefulness of lactic acid bacteria, it is necessary not only to promote its growth but also to inhibit death of its cells and further, it is required to maintain a high viable cell count in the final product during storage, for maintenance of these physiological effects at high levels, it is important to retain useful bacteria, such as lactic acid bacteria, in a viable state (Schmidt *et al.*, 1997).

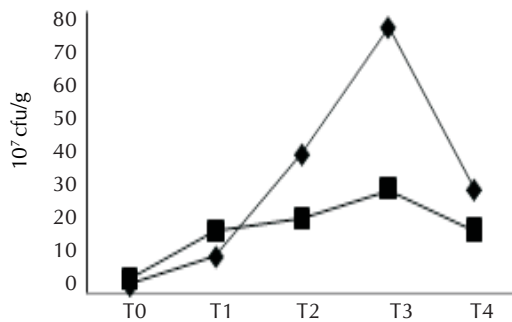
One reason for low counts in frozen yogurt is death of the culture by freezing (Donkor *et al.*, 2006). Freezing with agitation kills a large portion of added lactobacilli. Death of yogurt bacteria in frozen desserts reduces the potential to produce health benefits. The freezing step is especially critical as it negatively affects both viability and physiological state of the bacteria. The formation of ice crystals induces mechanical damage that leads to cellular death (Dave and Shah, 1996).

The objective of the present study was to increase the viable count of yoghurt culture and to maintain the viable count in the final product by protecting the yoghurt culture from adverse effect of freezing and subsequent storage at low temperature.

## MATERIALS AND METHODS

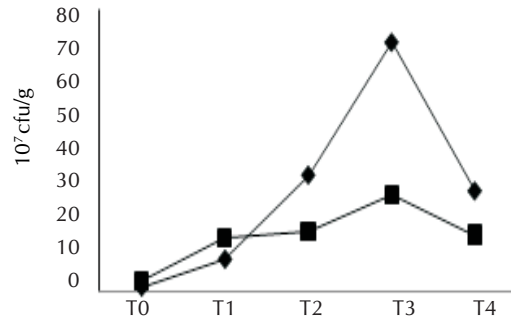
This study was carried out at Warner School of Food and Dairy Technology, Sam Higginbottom Institute of Agriculture, Technology and Sciences, Allahabad, U.P., India.

Viable count of yoghurt culture before freezing



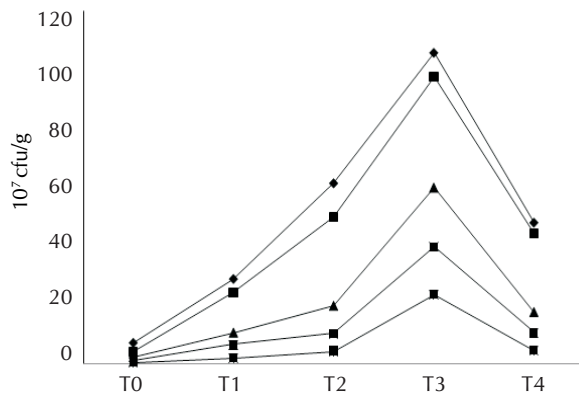
**Figure 1: Selective Enumeration of Yoghurt Culture ( $10^7$  cfu/g) in Frozen Yoghurt before Freezing**

Viable count of yoghurt culture after freezing (0 days)



**Figure 2: Selective Enumeration of Yoghurt Culture ( $10^7$  cfu/g) in Frozen Yoghurt after Freezing**

Average count of yoghurt culture



**Figure 3: Sum of viable count of yoghurt culture for before and after freezing (0, 10, 20 and 30 days)**

### Yoghurt Culture

(*Streptococcus salivarius* ssp. *Thermophilus* NCDC 074 and *Lactobacillus delbrueckii* ssp. *Bulgaricus* NCDC 009) were obtained from National Collection of Dairy Culture (NCDC), Dairy Microbiology Division at NDRI Karnal, Haryana, India.

### Herbs

(Ajowan, Black Pepper, Cardamom, Garlic, Ginger and Mint) were obtained from the local market in Allahabad, U.P., India.

### Selection of Herbal Extract

Six herbs viz., Ajowan, Black Pepper, Cardamom, Garlic, Ginger and Mint extract of 10° brix were selected to enhance the viability of probiotic culture by the application of Disc Diffusion method (Baur and Kirby, 1966).

### Procedure Adopted for Manufacturing Frozen Yoghurt

Both control (T<sub>0</sub>) and experimental (T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub>) frozen yoghurt mix were standardized to 5.0 % fat, 8.7% serum solids, 12 % sugar, stabilizer and emulsifier 0.5% and total solids adjusted to 26 %. Herbal extract (10° brix) was added at 0.0 %, 0.5%, 1.0%, 1.5% and 2.0% for treatments T<sub>0</sub>, T<sub>1</sub>, T<sub>2</sub>, T<sub>3</sub> and T<sub>4</sub> respectively.

The mix was homogenized and then pasteurized and cooled to 42°C and yoghurt starter culture was added at 2.5% (1:1 ratio). The mix was incubated at 42°C till we achieve an acidity of 0.45%. Before the mix is aged at 5°C and frozen in a batch freezer to an overrun of 70%. The frozen yoghurt was filled in cups and kept in deep freezer for hardening at a temperature of -18°C.

### Analysis of Yoghurt Culture

Selective enumerations of yoghurt culture (*Streptococcus salivarius* ssp. *thermophilus* NCDC 074, *Lactobacillus delbrueckii* ssp. *bulgaricus* NCDC 009) were done according to Shah (2000).

## RESULTS AND DISCUSSION

The highest mean selective enumeration ( $10^7$ cfu/g) of *Streptococcus thermophilus* score before freezing was 76 in group T<sub>3</sub> followed by 40, 30, 11 and 2.8 in groups T<sub>2</sub>, T<sub>4</sub>, T<sub>1</sub> and T<sub>0</sub> respectively (Fig. 1). The differences in the values were significant (P < 0.05), except groups T<sub>0</sub>-T<sub>1</sub> and T<sub>2</sub>-T<sub>4</sub>.

The highest mean selective enumeration ( $10^7$ cfu/g) of *Lactobacillus bulgaricus* score before freezing was 29.8 in group T<sub>3</sub> followed by 21.5, 18.1, 18 and 4.5 in groups T<sub>2</sub>, T<sub>4</sub>, T<sub>1</sub> and T<sub>0</sub> respectively (Fig. 1). The differences in the values were significant (P < 0.05), except groups T<sub>1</sub>-T<sub>2</sub>, T<sub>1</sub>-T<sub>4</sub> and T<sub>2</sub>-T<sub>4</sub>.

The highest mean selective enumeration ( $10^7$ cfu/g) of *Streptococcus thermophilus* count after freezing (0 days) was 70.5 in group T<sub>3</sub> followed by 33, 28.5, 9.2 and 1.3 in groups T<sub>2</sub>, T<sub>4</sub>, T<sub>1</sub> and T<sub>0</sub> respectively (Fig. 2). The differences in the values were significant (P < 0.05).

The highest mean selective enumeration ( $10^7$ cfu/g) of *Lactobacillus bulgaricus* count after freezing (0 days) was 27 in group T<sub>3</sub> followed by 17, 15.8, 15.1 and 3 in groups T<sub>2</sub>, T<sub>4</sub>, T<sub>1</sub> and T<sub>0</sub> respectively (Fig. 2). The differences in the values were significant (P < 0.05), except groups T<sub>1</sub>-T<sub>2</sub> and T<sub>2</sub>-T<sub>4</sub>.

The highest mean selective enumeration ( $10^7$ cfu/g) of sum of yoghurt culture (*Lactobacillus bulgaricus* + *Streptococcus thermophilus*) count before freezing was 105.8 in group T<sub>3</sub> followed by 61.5, 48.1, 29 and 7.3 in groups T<sub>2</sub>, T<sub>4</sub>, T<sub>1</sub> and T<sub>0</sub>

respectively (Fig. 3). The differences in the values were significant ( $P < 0.05$ ).

The highest mean selective enumeration ( $10^7$ cfu/g) of sum of yoghurt culture (*Lactobacillus bulgaricus* + *Streptococcus thermophilus*) count after freezing (0 days) was 97.5 in group  $T_3$  followed by 44.3, 24.3 and 4.3 in groups (Fig. 3). The differences in the values were significant ( $P < 0.05$ ).

The highest average total viable count ( $10^7$ cfu/g) after 10 days of storage between control and experimental treatment was 60 in group  $T_3$  followed by 19.9, 17.8, 10.8 and 2.5 in groups  $T_2$ ,  $T_4$ ,  $T_1$  and  $T_0$  respectively (Fig. 3). The differences in the values were significant ( $P < 0.05$ ).

The highest average total viable count ( $10^7$ cfu/g) after 20 days of storage between control and experimental treatment was 39.8 in group  $T_3$  followed by 10.6, 10.3, 6.5 and 1.1 in groups  $T_4$ ,  $T_2$ ,  $T_1$  and  $T_0$  respectively (Fig. 3). The differences in the values were significant ( $P < 0.05$ ) except groups  $T_2$ - $T_4$ .

The highest average total viable count ( $10^7$ cfu/g) after 30 days of storage between control and experimental treatment was 23.6 in group  $T_3$  followed by 4.6, 4.5, 2.1 and 0.64 in groups  $T_4$ ,  $T_2$ ,  $T_1$  and  $T_0$  respectively (Fig. 3). The differences in the values were significant ( $P < 0.05$ ).

Rajkumar (2009) reported that ginger extract and honey with cryoprotective agent (Glycerol) enhanced the viable cell count of frozen yoghurt. There is an adverse effect on total viable count of frozen yoghurt culture during storage due to death of cells (Donkor *et al.*, 2006; Singh, 2009). By analysing different levels of herbal extract we have found that viable count of frozen yoghurt was enhanced by the use of herbal extract.

Singh (2009) suggested that herbal extract (Ajowan, Cardamom, Garlic and Mint) enhances the growth of yoghurt culture. During storage the death of yoghurt culture was found non-significant and herbal extract can be recommended as growth promoter (Heenan *et al.*, 2004; Rajkumar, 2009; Radhika, 2010) for the yoghurt culture.

Chowdhury *et al.* (2008) prepared yoghurt with tulsi leaf (*Ocimum sanctum*), pudina leaf (*Mentha arvensis*) and coriander leaf (*Coriandrum sativum*) based. Tulsi yogurt had the maximum  $\alpha$ -galactosidase activity than other herbal yoghurt.

Some researchers have attempted to improve the viable count, sensory and nutritive characteristics of frozen yoghurt by adding probiotics during storage. Krasaekoopt *et al.* (2003) studied that encapsulation of probiotic culture protects the bacteria in the product's environment and improve their survival during freezing. Donkor *et al.* (2006) reported that presence of probiotic organisms (*Lactobacillus acidophilus* LAFTI® L10, *Bifidobacterium lactis* LAFTI® B94, and *L. paracasei* LAFTI® L26) enhanced proteolysis significantly in comparison with the control batch the increased proteolysis improved survival of *L. delbrueckii* ssp. *bulgaricus* Lb1466 during storage resulting in lowering of pH and production of higher levels of organic acids, which might have caused the low cell counts.

Frozen yoghurt with high viable count can be manufactured by addition of different level of herbal extract. These herbal extracts improve the rate of survival of yoghurt culture on freezing and storage of frozen yoghurt. Therefore, we can

recommend that use of herbal extract in frozen yoghurt enhances the viable count of yoghurt culture

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