

**Isolation of probiotics
Lactobacillus
acidophilus from
commercial yoghurt**

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Abstract

Probiotic *Lactobacillus* strain isolated from yogurt, after enrichment in MRS broth, was identified using a rapid screening method. The *Lactobacillus* strain was identified as *Lactobacillus acidophilus*. The viable number of the cell in the yogurt was about 107 CFU/ml.

Introduction

The relationship between certain food and health benefits has been investigated for many years. These studies have allowed the development of the concept of probiotic with positive effects on the host health or well-being beyond its nutritional value (Huggett and Verschuren, 1996).

Probiotic microorganisms are often incorporated in food in the form of yogurt and yogurt type fermented food. Recently, there are probiotic ice cream, cheese, infant formulas, breakfast cereals, sausages, luncheon meats, chocolate and puddings. In fact, there are probiotic products in the form of capsules and tablets (Chan et al., 2000). However, there are problems in determining the efficacy of probiotics as a whole. Although there are a wide range of species and strain used as probiotics, the efficacy of some of them remains in doubt or has not been fully proven. Furthermore, there are also problems of variation in viability or activity of the cells in the various preparations, the use of mixtures of microorganisms and their differential survival, and to ensure that the probiotic cells have a long shelf-life and reach the site of action. Therefore, in order to resolve some of these problems, research work has to be carried out on the formulation, stabilization and targeted delivery of probiotic microorganisms.

Materials and Methods

Chemicals

Man-Rogosa –Sharpe (MRS) broth (Criterion, U.S.A)

Man-Rogosa –Sharpe (MRS) Agar (Criterion, U.S.A)

Gram staining reagents (Becton, Dickinson and company, USA)

Micro plate TM (Biolog Hay Ward U.S.A)

Carbon dioxide generating system (Oxoid, U.K)

Sodium chloride (Oxoid, U.K)

Isolation of Lactobacillus acidophilus

Five ml of yogurt drink (Vitagen, Denmark) was inoculated into 95 ml of MRS broth (Criterion, USA) and incubated at 37°C for 24 hours and at 100 rpm of agitation. One loopful of the suspension was then streaked on MRS and nutrient agar media and the inoculated plates were incubated at 37°C for 24-72 hours. The single isolated colonies formed on both plates were examined and maintained on MRS agar medium.

For enumeration of viable cells, 0.1 ml of the yogurt drinks that was diluted serially in ten fold (10⁻¹-10⁻⁸, MRS broth as diluent), were spread plate on MRS medium. The plates were incubated at 37°C for 72 hours. The numbers of colony were counted and recorded.

Identification of Lactobacillus acidophilus

A single colony was identified using gram stain, scanning electron microscopy and BioLog identification system.

Results and Discussion

Only one type of cells was observed on the isolation media. To achieve preliminary identification, the isolates (15 altogether from 15 samples) were subjected to sugar fermentation pattern analysis using BioLog identification system and the results identified the isolates as Lactobacillus acidophilus. Figures 1 and 2 show the shape and arrangement of Lactobacillus acidophilus as seen under light microscope and scanning electron microscope, respectively.

The results of the enumeration showed that the viability of *L. acidophilus* in the yogurt drink was about 107 cells/ml.

Although other species such as *L. casei* and *L. paracasei* are increasingly being used in the manufacture of probiotic yogurts (Sarem-Damerdji et al., 1995) *L. acidophilus* is expected to be the main *Lactobacillus* species involved in the manufacture of yogurt and yogurt drinks. This observation has indeed been supported by this study using the Biolog identification system and electron microscopy. It seems reasonable to assume that adequate numbers of the probiotic bacteria need to be consumed to exert a health-promoting effect for the consumer and it has been suggested that to have any therapeutic effect, the minimal number of probiotic bacteria in a product should be above 10⁵ or 10⁶ per gram (Pennachia et al., 2004).

References

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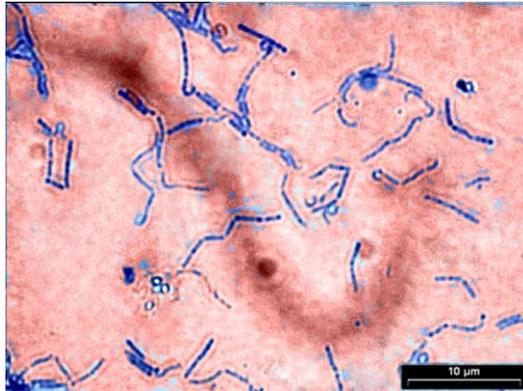


Figure 1: *Lactobacillus acidophilus* as observed under light microscope.
(Magnification 1000x)

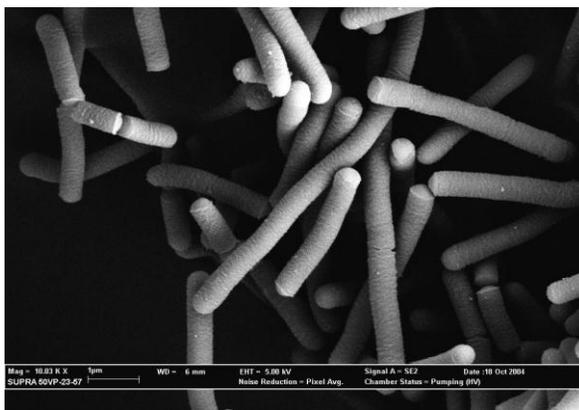


Figure 2: The SEM micrograph of *Lactobacillus acidophilus*