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TECHNICAL NOTE

A SIMPLE PROCEDURE FOR THE PREPARATION OF STIRRED YOGHURT FOR SCANNING ELECTRON MICROSCOPY

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Abstract

Stirred yoghurt is aspirated into agar gel tubes having 1.2 mm inner diameter, fixed in glutaralddehyde, dehydrated in ethanol, freeze-fractured under liquid nitrogen, and critical-point dried. Agar gel encapsulation protects the sample and prevents it from disintegration during the preparative steps. Scanning electron microscopy of the mounted fragments reveals the corpuscular microstructure of this type of yoghurt which develops due to stirring and pumping of the product during manufacture.

Introduction

Development of microstructure in set-style yoghurt was studied by electron microscopy (3-6, 8). Scanning electron microscopy (SEM) in particular has been found to be useful to show the porosity of the protein matrix (5), distribution of lactic acid bacteria (8), and the presence of fat globules in yoghurt made from whole milk (1). Preparation of stirred yoghurt for SEM is difficult due to disruption of the rigid gel matrix during manufacture caused by mechanical agitation to produce a smooth flowing viscous product. If a sample of stirred yoghurt is placed in an aqueous fixative, the broken gel part of it was aspirated into the agar gel tube as 3-5 ml of the yoghurt was placed on a glass plate and a consistent were used in this study. A volume of approx. 59°C for 24 h, dehydrated in a graded ethanol series, frozen in Freon 12 at -150°C, freeze-fractured under liquid nitrogen, melted in absolute ethanol, and critical-point dried from carbon dioxide. The fragments were mounted on aluminum SEM stubs using silver cement, sputter-coated with gold, and examined in a Cambridge Stereoscan Mark II scanning electron microscope operated at 20 kV.

Results and Discussion

Two steps are essential to this procedure: immobilization of the liquid sample in the agar gel tube and freeze-fracturing to obtain smooth fracture planes suitable for SEM examination. The sample to be examined by SEM is considerably larger than that destined for TEM. Also, because stirred yoghurt is a dense and viscous suspension of casein micelle clusters, it is easier to aspirate it into agar gel tubes of a diameter larger than that used for TEM.

During all the preparatory steps, the agar gel tube remained to be part of the sample. There was no separation of the yoghurt sample from the agar gel (Fig. 2) and the gel appeared to be relatively dense (Fig. 3). The sample fragments remained cohesive and were easy to mount on metal stubs using silver cement. Sputter coating provided sufficient conductivity to examine the sample at 20 kV without encountering charging artefacts. Freeze-fracturing revealed the corpuscular microstructure of the sample (Fig. 4). However, a detail of casein micelle chains and clusters in Fig. 5 is in agreement with images obtained with set-style yoghurt (5, 8).

Thus, this simple procedure makes it possible to prepare stirred yoghurt for SEM and to study the effects of manufacturing conditions on the dimensions and distribution of protein particles. It is probable that other similar foods such as cultured buttermilk can be prepared for SEM using this procedure.

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KEY WORDS: Agar gel; Encapsulation; Freeze-fracturing; Scanning electron microscopy; Stirred yoghurt.
Fig. 1. Aspiration of stirred yoghurt into an agar gel tube (A) and its sealing (B and C).

References


