Medbiotech Journal

MBTJ, 4(3): 118-120, 2020

Research Paper

Preparation of Probiotic Fruits through Encapsulation of Bacteria

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Received: 02 July 2020 Accepted: 15 August 2020 Published: 01 September 2020

Abstract

Lactobacillus casei cells were immobilized on fruit (apple and pear) pieces and the immobilized biocatalysts were used separately as adjuncts in probiotic cheese, whey and another product making. In the present review, the use of probiotic microorganisms for the production of novel foods is disc used, while the benefits and criteria of using probiotic cultures are analyzed. Subsequently, immobilization/ encapsulation application sin the food industry aim in gat the prolongation of cell viability are described together with an evaluation of their potential future impact, which is also highlighted and assessed. In parallel, cheese with free L.casei cell sand cheese only from renneted milk were prepared. The produced cheeses were ripened at 4 to 6°C and the effect of salting and ripening time on lactose, lactic acid, ethanol concentration, pH, and lactic acid bacteria viable counts were investigated.

Keywords: Cheese; Fruit; L.C Cell; Whey

How to cite the article:
E. Al Aziz, L. Temiat, Preparation of Probiotic Fruits through Encapsulation of Bacteria, Medbiotech J. 2020; 4(3): 118-120, DOI: 10.22034/mbt.2020.120936.

Introduction

Immobilization/encapsulation of probiotic is an exciting field of food technology that has emerged and developed rapidly in the past decade. The most excellent application of probiotic immobilization technology is the controlled and continuous delivery of cells in the gut [1]. The potential benefit of this therapeutic strategyi to maintain greater cell viability despite the acidity of the stomach. In their viable state, probiotics exert a health benefit on the host [2].

Fruits contain non-digestible carbo- hydrates, which constitute the base for cell immobilization. Apple and quince pieces proved to be suitable supports for immobilization of Lactobacillus casei cells The Immobilized biocatalysts were used in lactic acid and probiotic additive fermented milk production, while the immobilized bacterial cells were abletore activate after storage for 129days at 4°C. In the fermented milk, a fruity, distinctive aroma was predominant during all the storage period. Immobilized L. casei cells on fruit pieces have also been successfully used in probiotic cheese production.

Lactobacillus casei cells have been immobilized in some supports for lactic acid production. Agar was more effective than polyacrylamide for L. casei entrapment for lactic acid production from whey. Also, calcium pectate gel and chemically modified chitosan beads were used as sup- ports for L. casei cell immobilization. Alginate has so far been a popular matrix for immobilization of lactic acid bacteria. Other supports used for immobilization include porous foam glass particles, ceramic beads or porous glass, poraver beads, and gluten pellets [3].

In order to increase probiotic viability, production of apple and quince pieces supported L. casei is necessary due to the suitability of the supports as

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food ingredients. The produced biocatalysts have to be also examined for their suitability for lactic acid production. Therefore, the aims of t investigation were the increase of probiotic viability through fruit supported probiotic organism and use of the produced biocatalysts for lactic acid production [3].

Cell Immobilization for Probiotic Cheese Production

Lactobacillus casei ATCC393 was used. It was grown on MRS broth. Cell immobilization on apple and pear pieces (~0.5-cm3cubes) was carried out as described previousl. In brief, fruit (apples and pears of Stark in and Conference varieties, respectively) pieces (~500g) were in troduced in to 1L of L. casei liquid culture (~109cfu/mL) ,and allowed to ferment over night at 37°C wit out agitation [4]. When immobilization was complete (glucose in the liquid culture was <1g/L; concentration of glucose was determined by HPLC using the same method described for lactose determination below), the fermented liquid was decanted and the supported biocatalysts were washed twice with pasteurized milk. The biocatalysts were the nusedin cheese production Ewes 'milk was used for cheese production. It was he at ed at 65°Cfor30minandthencooled at 37°C.Cm m ercial rennet(0.01%)was added and the whole was left undisturbed for 2h for curd formation [3].

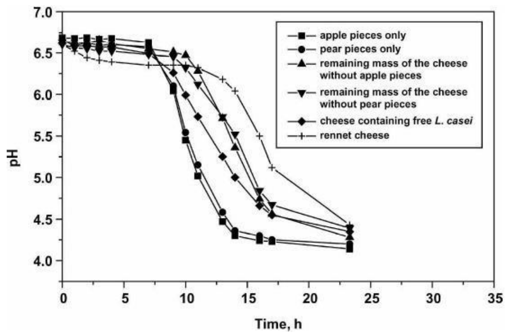

Figure 1. Kinetics of pH during lactic acid fermentation of whey by: a) *Lactobacillus casei* cells immobilized on apple pieces only, withdrawn from salted cheese (_), b) *L. casei* cells immobilized on pear pieces only, withdrawn from salted cheese (●), c) remaining mass of salted cheese produced by *L. casei* cells immobilized on apple pieces (▲), d) remaining mass of salted cheese produced by *L. casei* cells immobilized on pear pieces (▼), e) salted cheese containing free *L. casei* cells (◆), and f) salted rennet cheese (+), after ripening for 7 mo. Preliminary sensory evaluation revealed the fruit ytaste of the cheeses containing immobilized L.casei cells on fruit pieces. Commercial Feta cheese was characterized by a more sourtaste, where a snosignificant differences concerning cheese flavor were reported by the panel bet ween cheese containing free L.casei and rennet cheese. Salted cheeses scored similar values to commercial Feta cheese, where a sun salted cheese scores were significantly lower,but still acceptable to the sensory pane lists [5].

Subsequently, the curd was cut in squares (~ 1cmdiameter), left undisturbed for 10 min, and then clothfiltered. Immobilized L.casei on apple and pear pieces were added separately (50gof immobilized biocatalyst /L of milk used) during cloth filtration, which lasted overnight at room temperature (18to22°C) for complete whey removal. Cheese produced from milk containing ~ 109cfu/m L off ree L. casei , and cheese without L.casei cells(called rennet cheese)were produced for comparison. The effect of salt addition on chees equality characteristics was studied by rubbing 10g of salt/100gof cheese on the surface. Ripening of the produced cheeses was monitored at 4 to 6°C for 71 d [3].

Cell immobilization for Successive fermentation batches of whey

Apple and quince pieces were used as supports for immobilization. For the immobilization of cells, pieces of apple or quince (~500g) were introduced in 1 l of liquid culture of L.casei and allowed over night at 37∘Cwith out agitation. When immobilization was complet, the fermented liquid was decanted and the supported biocatalysts were washed twice with250 ml of whey. The biocatalysts were then used for lactic fermentation [3].

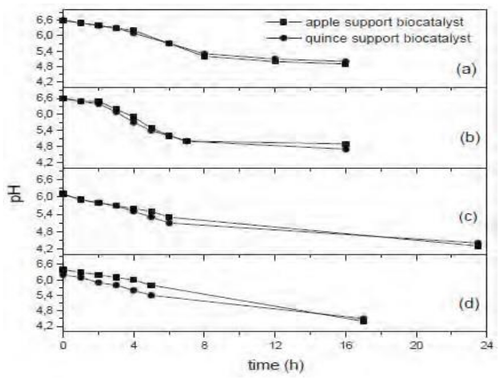

Figure 2. Kinetics during milk fermentation using immobilized *L. casei* cells on apple and quince pieces before (a) and after storage at 4 ∘C for 15 days (b), for 98 days (c), and for 129 days (d).

One hundred and seventy grams of apple or quince supported biocatalyst, prepared as described above, were introduced in to 400 ml of whey and 15 successive fermentation batches were carried out at different temperatures:30, 37, and 45∘C. All fermentations were performed under stationary conditions. During fermentation the pH was adjusted in the range 5.5–6.0 by addition of a saturated solution of Na2CO3. When the fermentation was completed, the liquid was decanted and the supports were washed twice with 250 ml of whey. At the end of every batch, samples were collected and an alyzed for lactic acid, ethanol, and residual sugar. Six successive fermentation batches of whey using initially 12.5g/l wet weight free cells were carried out at 30,37, and 45 ∘ C. The exact quantity of immobilized cells was impossible to estimate and an dicative quantity of free cells was used [3].

Cell immobilization on apple pieces for various microorganisms

Immobilization of S. cerevisiae AXAZ-1, K .marxianus IMB3 and L. casei on apple pieces was carried out as described previously. In brief, the proper amount of apple pieces was introduced ina liquid cell culture and allowed to ferment overnight. Then the fermented liquid was decanted, the immobilized biocatalyst was washed twice with the liquid that was used for the next fermentation and apple-supported biocatalyst was ready for use [1].

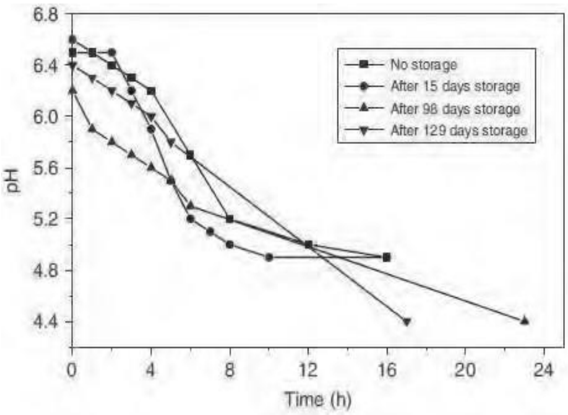

Figure 3. Fermentation kinetics during milk fermentation using immobilized L. casei cells on apple pieces after storage at 4 1C for 15, 98 and 129 days.

Lacto baccillus casei cells immobilized on apple pieces were used for probiotic, fermented milk and lactic acid production. Apple supported L.casei used for successive fermentation batches of whey proved to be very effective and suitable for food-grade lactic acid production. The immobilized biocatalyst was also used for milk fermentation and was able to ferment after storage for 15, 98 and 129 days at 41C, while no infection was reported during storage periods [3,5].

Conclusions

Pieces could be manufactured with some additions to the traditional cheese-making practice. The production of probiotic cheese in which the probiotic culture would survive and develop during manufacture and throughout its shelf life could lead to a major economic advantage. Apple pieces were found suitable support for yeast and bacterium cell immobilization. Immobilization method was cheap, simple and easy. The immobilized biocatalysts were success fully used in wine-making in both batch and continuous process and in lactic acid fermentation of whey and milk, while preliminary evaluation of the quality of the fermentation products revealed their improved aroma and fine taste [1].

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