



## Spray drying *Lactobacillus casei* 01 in lychee juice varied carrier materials



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Sodium carbonate (PubChem CID: 10340)  
Gallic acid (PubChem CID: 370)  
Oxalic acid (PubChem CID: 971)  
L-Ascorbic acid (PubChem CID: 54670067)  
Bile salt (PubChem CID: 439520)  
2,6-Dichlorophenolindophenol (PubChem  
CID: 13726)  
Ethanol (PubChem CID: 702)  
Phosphate buffer (PubChem CID: 24203)  
Hydrochloric acid (PubChem CID: 313)

### ABSTRACT

The objective of this study was to find out an optimal condition of encapsulated *Lactobacillus casei* 01 in lychee juice by spray drying. Accordingly, four outlet temperatures, i.e. 60, 70, 80 and 90 °C and two carrier materials, i.e. maltodextrin and gum arabic were conducted; subsequently, the physical, chemical and microbiological qualities of all spray-dried powders were investigated. The results showed that an optimal outlet temperature for spray drying *L. casei* 01 in lychee juice could be achieved at 80 °C, since it enabled product powder with satisfactory number of survival cells, good solubility, low moisture content and low water activity. In addition, 15% (w/v) maltodextrin plus 5% (w/v) inulin was considered as an optimal carrier to protect the cells from being damaged by an adverse environment of simulated gastrointestinal tract also by storage condition.

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## 1. Introduction

Most probiotics are usually associated with dairy products. Thus, an application of the probiotic cultures in non-dairy products like beverages or dehydrated fruit-juice could be an optional diversity for the consumers who are aware of functional soft-drinks. Spray dry is one of the feasible methods used in dairy industries to immobilize or encapsulate viable cells (Riveros, Ferrer, & Bórquez, 2009). An optimal drying condition can maintain the activity and viability of the probiotic throughout the storage period as well as in the human digestive tract. However, their survivability would

totally depend on the operating technique such as drying temperature, carrier materials and other storage conditions (Peighambaroust, Tafti, & Hesari, 2011). Lian, Hsiao, and Chou (2002) found that 10% (w/w) gelatin, gum arabic or soluble starch have given rise to a high survival of spray-dried *Bifidobacterium* cultures and the viability of these cultures was greatly dependent on the type and concentration of carriers. In addition, the stability of spray-dried lactobacilli usually decreased upon storage, but low storage temperatures enabled higher survival rates (Corcoran, Ross, Fitzgerald, & Stanton, 2004).

Lychee fruit (*Litchi chinensis* Sonn.) is an excellent source of vitamin C (40–90 mg/100 g) (Menzel, 2002). It also contains 21.6 g/100 g total sugar contents (Haq & Rab, 2012), 0.37 g/100 g total titratable acidity (as citric acid) (Sun, Liang, Xie, Lei, & Mo, 2010), 43 aroma volatile compounds (Wu, Pan, Qu, & Duan, 2009) and

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0.8 mg/g total phenolic compounds (Dajanta, Apichartsrangkoon, & Somsang, 2012). Fortification of this fruit juice with probiotics in the powdery form could produce an interesting product to tackle the global markets. Only a few studies to date have examined this type of functional soft-drink. Thus, this study had attempted to search for an optimal spray-drying temperature and an appropriate carrier material to encapsulate *Lactobacillus casei* 01 in lychee juice.

## 2. Materials and methods

### 2.1. Probiotic strain and growth condition

*Lactobacillus casei* 01 was purchased from Chr. Hansen (Hørsholm, Denmark). A weight of 10 g dried probiotic cells were rehydrated in 100 ml de Man Rogosa and Sharpe (MRS) broth (Hi-Media, Mumbai, India) for 10 min at room temperature and was incubated anaerobically at 37 °C for 24 h using Anaerocult® C system (Merck, Munich, Germany). Then, 1% (v/v) culture was inoculated in MRS broth and incubated anaerobically at 37 °C for 14 h to achieve its early stationary stage (Chaikhram, Apichartsrangkoon, Jirattananarangsri, & Van de Wiele, 2012; Chaikhram, Apichartsrangkoon, George, & Jirattananarangsri, 2013). After the incubation, probiotic cells were harvested and washed twice with 0.85% (w/v) sterile saline water by centrifugation at  $4000 \times g$  for 15 min at 4 °C. The cell pellet was diluted to provide a bacterial concentration of  $10^{11}$  CFU/ml by saline water.

### 2.2. Preparation of lychee juice

The lychee fruit (*Litchi chinensis* Sonn., cv. Hong Huay) from an orchard in Chiang Mai, Thailand was peeled, removed seeds and cleaned. The fruit was extracted with drinking water at a ratio 1:1 (w/w) using a juice extractor. The extract was then pasteurized at 90 °C for 1 min in a double jacket kettle with consistent stirring, subsequently it was cooled down to 25 °C. The pasteurized juice has pH  $3.62 \pm 0.34$  and total soluble solids  $8.75 \pm 0.26$  °Brix.

### 2.3. Spray drying

A spray dryer (JCM Engineering concept, Bangkok, Thailand) was equipped with a fluid atomizer (inside diameter of 5 mm) operated in a co-current manner. Drying condition for the entire experiment was adjusted as follows: 25 °C feeding temperature, 0.6–1 l/h feeding rate, 15 psi atomizing pressure and 150–170 °C hot-air-inlet temperature to generate 60–90 °C outlet temperatures. Pasteurized lychee juice was separately blended with either 20% (w/v) maltodextrin (10.5 dextrose equivalent, DE) or mixture of 15% (w/w) maltodextrin plus 5% (w/w) inulin or mixture of 15% (w/w) gum arabic plus 5% (w/w) inulin or 20% (w/v) gum arabic. Subsequently, the probiotic culture was inoculated into the mixed juice to obtain a bacterial concentration of roughly  $10^{10}$  CFU/ml. Since the outlet temperature had significant effect on the survival cells, various outlet temperatures i.e.  $60 \pm 2$ ,  $70 \pm 2$ ,  $80 \pm 2$  and  $90 \pm 2$  °C were adjusted. The collected powder was vacuum sealed in a laminated bag (polyethylene terephthalate/polypropylene/aluminum) and kept in a refrigerator for further study.

### 2.4. Quantification of the encapsulated cells

For the purpose, 1 g powder was mixed with 9 ml of 0.1 M sterile phosphate buffer (pH 7) (Merck, Munich, Germany) in a stomacher (IUL Instruments, Barcelona, Spain) for 10 min at room temperature. After releasing the probiotic cells, various dilutions were made with 0.1% (w/v) sterile peptone water (Hi-Media, Mumbai, India), subsequently they were plated on MRS agar (Hi-Media, Mumbai,

India) and incubated anaerobically at 37 °C for 48–72 h prior to performing the viable count.

### 2.5. Physical measurement

#### 2.5.1. Microstructure of probiotic-lychee-juice powder

To examine the microstructure of probiotic-lychee-juice powder, the powder samples were coated with gold for 10 nm using E1010 ion Sputter (Hitachi Science Systems, Ltd., Tokyo, Japan). The gold-coated samples were then determined by a Scanning Electron Microscope (Model S-3000 N, Hitachi High-Technologies Co., Ltd., Tokyo, Japan) at an accelerating voltage of 15 kV.

#### 2.5.2. Glass-transition temperature ( $T_g$ )

Differential scanning calorimeter (Diamond DSC, Perkin–Elmer, Inc., Waltham, MA, USA) was used to study the glass-transition temperature of powder samples. The instrument was calibrated by using indium. A weight of  $5 \pm 2$  mg sample was placed in an aluminum pan and sealed. The  $T_g$  of the sample was scanned with 10 °C/min scanning rate over the temperature range of 30–140 °C using an empty pan as a reference. The glass transition value was taken as the midpoint of the glass transition range. At least 10 runs per sample were performed.

#### 2.5.3. Bulk density

Bulk density of the powder was determined following Goula and Adamopoulos (2005) with some modifications. Accordingly, 5 g probiotic-lychee-juice powder was filled into a 10 ml graduated cylinder and held on a vortex vibrator for 1 min. The ratio of mass of the powder and the volume occupied in the cylinder determines the bulk density value (g/ml).

#### 2.5.4. Solubility

Solubility was determined as described by Cano-Chauca, Stringheta, Ramos, and Cal-Vidal (2005) with some modifications. Briefly, 1 g sample powder was added into 100 ml distilled water and agitated with a magnetic stirrer at a medium speed for 5 min. The mixture was then centrifuged at  $3000 \times g$  for 5 min. After that, an aliquot of 25 ml supernatant was carefully transferred to pre-weighed Petri dishes and immediately oven-dried at 100 °C overnight. The solubility (%) was calculated as the weight difference.

#### 2.5.5. Hygroscopicity

Hygroscopicity of spray dried powder was determined following the method described by Tonon, Brabet, and Hubinger (2008) with some modifications. A weight of 1 g powder sample was placed in an airtight container containing a saturated NaCl solution (75.29% RH) at 30 °C. After one month, the sample was weighed and hygroscopicity was expressed as gram of adsorbed moisture per 100 g sample (g/100 g).

#### 2.5.6. Water activity

A water activity meter (AquaLab Series 3, Decagon Devices, Inc., Pullman, WA, USA) was used to measure water activity ( $a_w$ ) of the spray-dried powders.

### 2.6. Chemical analysis

#### 2.6.1. Moisture contents

Moisture of the powders was carried out using 5 g sample, and dried in an oven at 100 °C until reaching a constant weight (AOAC, 2000).

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