



The influence of solution viscosities and surface tension on calcium-alginate microbead formation using dripping technique



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ABSTRACT

Currently, there is a growing interest for encapsulation of bioactive ingredients using calcium alginate microspheres or beads. Diffusion of the Ca in the alginate droplets provokes an ionic gelation and their conversion to hydrogel beads. The main objective of this study is to investigate the influence of physical properties of alginate and CaCl₂ solutions on alginate droplet formation and penetration into gelling bath, in size and shape of Ca-alginate beads. The droplet formation and penetration in the calcium bath was investigated using a high-speed video recording. Viscosity of alginate solution was modified by changing the alginate concentration (from 10 to 30 g/L) and the viscosity of the bath was modified by using different water/glycerol mixes (0–90% of glycerol) to prepare the CaCl₂ solution. Surface tension of CaCl₂ solution is reduced by adding different concentrations of surfactants (Tween 20) in a range of 0.01–1 g/L. Droplets detach from the tip with a tear shape. In all conditions tested, the droplets reach a spherical shape in less than 15 ms after detachment and less than 25 mm from the tip. Spherical beads are obtained when the kinetic energy is high enough to break the surface resistance of the calcium bath and droplet viscosity high enough to avoid deformations. Penetration depth of alginate droplets were mainly affected by viscosity and surface tension of CaCl₂ solution. When viscosity and surface tension of CaCl₂ solution increases, sphericity decrease and shape deformations are observed. The surfactant addition enhanced penetration and prevented shape deformations of Ca-alginate beads.

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1. Introduction

Bioencapsulation is defined as the process of confining bioactive compounds (e.g. microbial cells, enzymes, animal cells, plant cells, antibiotics, etc.) within a matrix in particulate form (i.e. bead or capsule) in order to achieve one or more desirable effects: immobilization, protection, stabilization, controlled-release and/or affect the physical properties of the bioactive compounds (Chan, Lee, Ravindra, & Poncelet, 2009; Doherty et al., 2011). Nowadays, functional foods are new trend for consumers towards a healthier life. Encapsulation technologies are promising alternatives to overcome problems in food industry such as degradation or oxidation of bioactive compound incorporated into food (Akhtar,

Murray, Afeisume, & Khew, 2014; Li et al. 2015; Schrooyen, van der Meer, & De Kruif, 2001; Ubbink & Krüger, 2006). The application of encapsulation in food industry includes the entrapment of bioactive compounds such as antioxidants, vitamins and minerals, essential oils, polyunsaturated fatty acids (PUFAs) mainly n-3 polyunsaturated fatty acids, flavors, enzymes, probiotics, etc., into small capsules (Gibbs et al., 1999; Koupantsis, Pavlidou, & Paraskevopoulou, 2014; Rajabi, Ghorbani, Jafari, Mahoonak, & Rajabzadeh, 2015; Yang & McClements, 2013).

The alginate, a natural polysaccharide, is extensively used as wall material in encapsulation processes due to their non-toxicity, biocompatibility and mild gelling properties (Knezevic et al., 2002). Moreover, sodium alginate is preferentially used as a gelling agent in the food industry mainly due to its high water-solubility (Nakauma et al., 2016). The alginate gel beads are generally produced by extrusion drop-by-drop (dripping) of an alginate solution in gelling calcium bath (Kierstan & Bucke, 2000; Lević, Lijaković, Đorđević, Rac, Rakić, Knudsen, et al., 2015; Zeeb, Saberi, Weiss, & McClements, 2015). Physical characteristics such

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as shape and size are important in the consideration of alginate beads properties (Belščak-Cvitanović, Bušić, Barišić, Vrsaljko, Karlović, Špoljarić, et al., 2016; Chan, Lim, Ravindra, Mansa, & Islam, 2012; Nussinovitch, 2010). However, there is limited information on shape deformations of alginate beads, which occurred, in different steps of encapsulation process. Recently, a master shape diagram has been developed to reveal the relationship between the alginate solution variables and the shape of the Ca-alginate large beads formed by the simple extrusion dripping method (Chan et al., 2009).

The main objective of this study is to investigate the alginate droplet formation at nozzle tip, its evolution during falling and penetration in the gelling calcium bath, in function of physical properties such as viscosity and surface tension of alginate and receiving CaCl_2 solution using a high-speed camera. Images were captured and analyzed to define bead diameter, sphericity and shape deformations during the different steps of the bead production.

2. Materials and methods

2.1. Materials

Sodium alginate powder ALGOGEL 3001 (molecular weight: 151,200 Da) was obtained from Cargill (France). Algogel 3001 has a high guluronic acid content (M/G ratio) as 0.64. Calcium chloride dihydrate powder ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) was purchased from Panreac (Spain). Glycerol used in the study was 99, 5% pure and obtained from Labogros (France). Surfactant (Tween 20) and all the other chemicals used in the study were obtained from Sigma-Aldrich (St. Louis, MO USA).

2.2. Methods

2.2.1. Preparation of alginate solutions

A defined quantity of alginate was spread in a beaker containing 1 L of distilled water under high speed mixing. Then, the mixing speed was reduced and stirring was maintained for 1 h at low speed. Finally, the solutions were left standing for 24 h to remove bubbles before use. To evaluate the impact of the alginate solution viscosity, solutions of 5–35 g/L were prepared.

2.2.2. Preparation of gelling CaCl_2 solutions

Twenty g calcium chloride ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) was introduced in a beaker containing 1 L of an aqueous solution. In order to evaluate the impact of the viscosity of gelling calcium solution, the aqueous solution was prepared by mixing glycerol and water. The concentration of glycerol was varied from 0 to 90% (v/v). In order to observe the influence of surface tension on penetration process, up to 1 g/L Tween 20 was added into CaCl_2 solution. In solution series, the calcium concentration was kept constant.

2.2.3. Viscosity measurement of solutions

The viscosities of alginate and CaCl_2 solutions were determined by using viscometer (Haake VT 550, USA) using Rheowin Job Manager Software. Fifty ml of solution inserted in the cell. The sample placed into the chamber was subjected to an increase of shear rate from 0 to 300 s^{-1} followed by a decreasing shear rate from 300 to 0 s^{-1} , over 60 s. The reading is done at 200 s^{-1} . During the viscosity measurements, the temperature was kept at $25 \text{ }^\circ\text{C}$. All measurements were done as three replicates.

2.2.4. Surface tension measurement of solutions

The surface tensions of solutions were determined by using tensiometer (KRÜSS K-12, France) using du Nouy ring method and

LabDesk™ software. For the surface tension measurement, 70 ml of each solution was used for the analysis and the temperature was kept at $25 \text{ }^\circ\text{C}$ (Lee, Ravindra, & Chan, 2008). All measurements were done as three replicates.

2.2.5. Experimental set-up and production of the alginate beads

Fig. 1 shows the experimental set-up for producing the beads. Alginate solution was filled into a hypodermic syringe (Terumo, Tokyo) which was connected to a syringe pump (KD Scientific, USA). Solution was extruded drop-wise through conical tip of 0.6/0.9 mm internal/external diameter (Norton EFD, Dosage 2000; France) into the CaCl_2 bath. A gentle mixing was applied to CaCl_2 solution to prevent sticking of the beads.

First, alginate flow rate were changed from 50 to 300 ml/h in order to study the influence of it on bead diameter and sphericity. Then, alginate flow rate was set to 100 ml/h in order to see the influence of viscosity and surface tension of solution. The distance between the nozzle tip and collecting bath was 20 cm. After droplet penetration, beads were let to harden for 30 min to ensure complete gelation. Afterwards, the beads were filtered using a sieve and rinsed with distilled water. Then, beads were hold in CaCl_2 solution until size and shape analysis. In order to understand if the falling drop has enough energy to break the surface, kinetic energy was measured and the liquid properties of the gelling bath were not varied this time. The collecting distance between the dripping tip

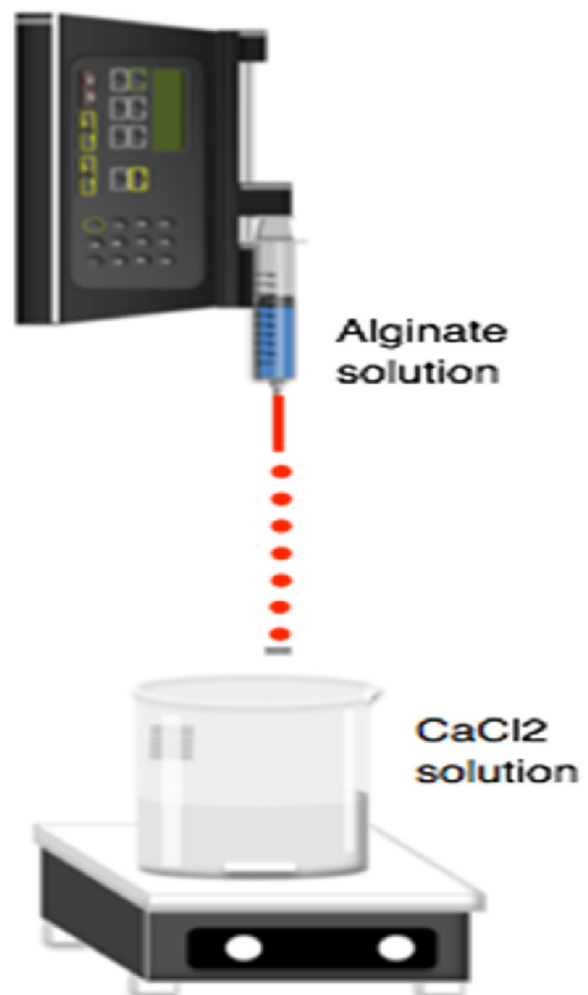


Fig. 1. Schema of the alginate bead production set-up.

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