Materials Science and Engineering C 46 (2015) 132–139

Contents lists available at ScienceDirect

Materials Science and Engineering C

journal homepage: www.elsevier.com/locate/msec

Bioinspired preparation of alginate nanoparticles using microbubble bursting

Mohamed Elsayed, Jie Huang, Mohan Edirisinghe *

Department of Mechanical Engineering, University College London, Torrington Place, London WC1E 7JE, UK

ARTICLE INFO

Article history:
Received 7 July 2014
Received in revised form 18 August 2014
Accepted 26 September 2014
Available online 28 September 2014

Keywords:
Nanoparticles
Microbubbles
Microfluidic
T-junction
Alginate

ABSTRACT

Nanoparticles are considered to be one of the most advanced tools for drug delivery applications. In this research, alginate (a model hydrophilic polymer) nanoparticles 80 to 200 nm in diameter were obtained using microbubble bursting. The natural process of bubble bursting occurs through a number of stages, which consequently produce nano- and microsized droplets via two main production mechanisms, bubble shell disintegration and a jetting process. In this study, nano-sized droplets/particles were obtained by promoting the disintegrating mechanism and suppressing (limiting) the formation of larger microparticles resulting from the jetting mechanism. A T-junction microfluidic device was used to prepare alginate microbubbles with different sizes in a well-controlled manner. The size of the bubbles was varied by controlling two processing parameters, the solution flow rate and the bubbling pressure. Crucially, the bubble size was found to be the determining factor for inducing (or limiting) the bubble shell disintegration mechanism and the size needed to promote this process was influenced by the properties of the solution used for preparing the bubbles, particularly the viscosity. The size of alginate nanoparticles produced via the disintegration mechanism was found to be directly proportional to the viscosity of the alginate solution.

1. Introduction

Nanoparticles have made a remarkable impact in biomedical applications as drug delivery carriers, non-viral vectors for gene therapy, labelling and tracking agents for imaging and diagnosis etc. [1]. They are characterised by having a higher cellular uptake compared to microparticles, which can be beneficial in decreasing the required dosage of the administrated drug [2]. Nanoparticles also have a better chance to fulfil their therapeutic purpose as they undergo a lower intensity of opsonisation reactions caused by the accumulation of proteins on the surface of the particles, thus they are able to avoid the fast immune response leading to clearance from the body [3]. Nanoparticles with sizes between 70 and 200 nm are found to have the longest circulation time in the body in comparison with larger particles [4]. A longer circulation time for a drug carrier in the blood flow can cause better absorbance and sustained release for the administrated drug, which is an important contributor for the efficiency and success of a drug carrier [5]. Therefore, nanoparticles are able to improve the treatment of a wide range of diseases, such as cancer, cardiovascular problems, viral and bacterial infections [6].

Both synthetic and natural polymer nanoparticles have been developed for drug delivery, such as poly(D,L-lactide), poly(lactic acid), poly(D,L-glycolide), poly(lactide-co-glycolide) acid, gelatin, chitosan, collagen and alginate [7]. Compared to hydrophobic polymers, hydrophilic nanoparticles are accompanied by lower intensity of opsonisation reactions [3]. They are capable of intracapillary and transcapillary penetrations, and reducing the immunological opsonisation reactions that cause the fast clearance of drugs before reaching their targeted tissues [8].

Alginate nanoparticles, a class of natural polymers constituted by sugar monomers, were found to significantly enhance the bioavailability of encapsulated drugs [9]. Alginate is known for its mucoadhesive nature caused by its anionic nature which enables it to selectively adhere to mucosal membranes, a beneficial feature for developing drug delivery systems for transmucosal applications [10]. Furthermore, alginate-based materials are known to be pH-sensitive and this property can be used for the 'smart' control of the release mechanism of the encapsulated biomolecules from the alginate drug delivery vehicle.

Many technologies have been used to prepare nanoparticles, such as ionic gelation [11], nanoprecipitation [12], emulsion cross-linking [13] and spray-drying [14]. Using the microemulsion method, 350 nm alginate nanoparticles have been produced with 40% encapsulation efficiency [15]. Despite their success in the preparation of nanoparticles, conventional polymer processing methods often face obstacles in obtaining monodispersed nanoparticles, such as being energy intensive and dependant on the need of organic solvents.

Bubble bursting occurring in the marine environment is one of the powerful phenomena in nature and is able to produce nano-sized and micron-sized droplets and particles [16]. The natural process of bubble formation in the marine environment is caused by wave breaking. The

* Corresponding author.

http://dx.doi.org/10.1016/j.msec.2014.09.036
0928-4931/

burst of foam bubbles produced can project a large collection of drops into the air [17]. The process of bubble bursting is known to produce droplets with various sizes in the nano- and the micrometre range using two formation mechanisms, i.e. bubble film disintegration and jetting [18]. The former mechanism depends on the disintegration of the bubble cap or film, which separates the inside of the bubble from the surrounding air, and results in the production of very fine droplets, down to the nanometre range. In contrast, the latter produces droplets in the micrometre range by the partitioning of a liquid jet that emerges after the collapsing of the bubble [19].

The composition of particles and droplets produced by bubble bursting was found to be dependent on the composition of the coating of the parent bubbles, which is closely related to the solution (the environment) bubbles originate from [20]. The process of bubble bursting in the marine environment is governed by various factors, particularly the bubble size [18]. The domination of film drop production is correlated with larger bubbles, in the millimetre range, while jet drops are generally accompanied by the bursting of smaller micrometre sized bubbles [17]. Therefore, it is hypothesised that the phenomenon of bubble bursting can be utilized to obtain alginate nanoparticles solely from alginate microbubbles by controlling the size of bubbles and preventing the jetting process.

A number of microfluidic devices have been successfully used for microbubble preparation [21]. In particular, gas pressure-driven T-junction devices have the ability to prepare microbubbles with controllable sizes and can also be combined to other methods such as electrohydrodynamics to control the bubble size [22]. Using T-junctions, monodispersed microbubbles can be obtained, which can significantly narrow the bubble size distribution from other bubbling methods such as sonication [23]. Microbubbles obtained using a T-junction are produced by a 'pinch-off" process that occurs at the gas–liquid interface [24]. The process of bubble formation and their size prepared using a T-junction with circular cross section can be controlled using capillary size, liquid viscosity and the ratio of liquid to gas flow rate [25]. To achieve a stable microbubbling process, the bubbling pressure has to be high enough to overcome the surface tension of the solution [21] and the output bubble consists of a volume of gas, encapsulated in a shell of a stabilizing liquid.

In addition to the simplicity of design, the T-shaped device is known to have high efficiency in preparing droplets and bubbles in a highly controllable manner [25]. Recently, hydrophobic polymethylsilsesquioxane (PMSQ) nanoparticles have been obtained from microbubbles using a T-junction [24]. The mechanism of nanoparticle formation is dependent on the evaporation of a volatile solvent (i.e. acetone and ethanol) from the coating (shell) in the microbubbles.

This study, the process of obtaining alginate nanoparticles was carried out by controlling bubble bursting, without the aid of solvent evaporation and this can extend the selection of biomolecules to be delivered. Furthermore, the development of a nanoparticle preparation method for hydrophilic natural polymers that does not depend on treatments such as sonication or organic solvents can enhance their utilization in biomedical applications. This is due to the highly organized structure (at both the molecular and macroscopic levels) of natural polymers that, if preserved, can offer useful features particularly within biomedical applications [26].

2. Materials and Methods

2.1. Materials and preparation of alginate solutions

A series of alginate solutions with concentration of 0.1, 0.25, 0.4 and 0.7 wt.% were prepared from sodium alginate powder (Sigma-Aldrich, Poole, UK). Alginate powder was dissolved in deionized water followed by the addition of the surfactant polyethyleneglycol-40-stearate (PEG-40S, Sigma-Aldrich, Poole, UK, density; 1300 kg/m^3) and the two contents were further stirred until a homogenous solution was obtained. The concentration of the surfactant was 0.25 wt.% in all solutions.

2.2. Characterisation of alginate solutions

A Brookfield DV-11 Ultra programmable Rheometer (Brookfield Engineering Laboratory Inc., Middleboro, MA, USA) was used to measure the viscosity of the alginate solutions. The surface tension of the solutions was measured using a Kruss Tensiometer (Model-K9, Kruss GmbH, Germany). The different measurements were performed at ambient temperature (22 °C) and the average of seven readings was taken to ensure accuracy.

2.3. Preparation of alginate microbubbles

The T-junction microbubbling set-up is illustrated in Fig. 1. The T-junction microfluidic device is fabricated using poly(methylmethacrylate) via CNC machining. It consists of two perpendicular Teflon capillaries with a constant inner diameter (200 μm). The vertical capillary is connected to a gas cylinder, connected to a digital manometer for controlling the pressure supplied to the junction. The horizontal capillary provides alginate solution flow and is connected to a 20-ml stainless steel syringe (KD Scientific, Holliston, MA, USA), which is connected to a syringe pump (Harvard, PHD 4400). To produce microbubbles, the gas pressure was gradually increased until it overcame the surface tension of the supplied solution. The liquid and gas meet at the intersection zone of the two capillaries and bubbles are produced at the gas–liquid interface.

The produced microbubbles were collected from the outlet capillary on a glass slide coated with deionized water. The size and uniformity of the bubbles were examined using an optical microscope (Zeiss Axiotech) fitted with a Nikon Eclipse ME 600 camera. The influences of the gas (air) pressure and the flow rate of alginate solutions on the size of the alginate microbubbles were investigated firstly. The effect of the size of microbubbles on the formation of nanoparticles from microbubble bursting was then studied. Finally, the effect of the viscosity of the alginate solution on the size of the nanoparticles obtained was investigated. The alginate nanoparticles obtained from the bursting of bubbles prepared from the different alginate solutions were examined using a Jeol JSM-6301F field emission scanning electron microscope (FE-SEM, JEOL Ltd., Herts, UK). Before SEM examination, the samples were sputter coated with gold for 3 min (Edwards Sputter coater S150B). Nanoparticles were observed under SEM at an accelerating voltage of 3–5 kV, with a working distance of 15–35 mm. The size distribution of microbubbles and the nanoparticles at various processing conditions was measured using the UTHSCSA Image Tool software program (Version 3.00).

3. Results and Discussion

3.1. Alginate solutions

The viscosity and surface tension of the alginate-PEG solutions, which are crucial for microbubbling, are shown in Table 1. With increasing alginate concentration (0.1 to 0.7 wt.%) the viscosity of alginate-PEG-40S solutions increased from 1.5 to 19 mPa s and surface tension varied between 43 and 47 mN/m.

3.2. Alginate microbubbles

The formation of bubbles depends on the meeting of an air column with the solution inside the T-junction, where a neck starts to form which gradually narrows and eventually forms a "slug" that detaches and the resulting bubbles are collected from the end of the capillary. One of the main advantages of using the T-junction is the ability to prepare monodispersed bubbles. To obtain monodispersed microbubbles, the flow rate of the solution and the gas pressure have to be adjusted simultaneously, otherwise microbubbles with various sizes and large polydispersity are formed. In addition to monodispersity, the size of