

Research Article

Effects of pH on the Shape of Alginate Particles and Its Release Behavior

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A vast majority of alginate particles exist as spheres in most practical uses, and both the particle shape and size are the key factors dominating the applications and performance of alginate gels. Therefore, it becomes an issue of great interest to investigate the aspheric alginate particles. As the first step, various shaped alginate particles were formed due to various pH values in gelation solutions. It was experimentally demonstrated that a low pH brought about an oblate shape, and particularly lower concentrations of both alginate and divalent cations resulted in a flattened oblate shape. Ba²⁺ acting as a cross-linker had a less impact on the particle shape than Ca²⁺ due to a higher affinity in alginate intermolecular cross-linking. With a larger surface area, an oblate particle offered a higher release rate than a spheric one.

1. Introduction

Over the past few decades, utilization of natural polymers for the development of drug delivery systems remained a subject of great interest [1–3]. Natural polymers remain attractive mostly due to their easy availability, cost effectiveness, biodegradability, and biocompatibility [4]. There is a large amount of alginate in our environment [5] and is discovered as the structural components of brown marine algae [6]. Alginate is a linear copolymer consisting of guluronic (G) and mannuronic (M) acid forming regions of M- and G-blocks and alternating structure (MG-blocks) [7]. Divalent cations such as Ca²⁺ and Ba²⁺ bind to the G-blocks of alginate in a highly cooperative manner, such that a gel is formed [8, 9]. Commonly used in a wide variety of fields, alginate is so far mainly processed into capsules, beads, and fibres [10] and has been frequently used for encapsulation of biologically active agents [11].

A vast majority of alginate particles exist as spheres in either internal [12, 13] or external gelation [14–17], but

this geometry has some disadvantages when in use [18]. Applications and performance of alginate particles depend upon their shape and size. It becomes an issue of great interest to investigate the effects of aspheric particles in a wide range of scientific fields [19, 20], such as tail-shaped [21], mushroom-like [22], and hemispheric [23] particles. Disk-shaped particles showed a better half-life in circulation and precise targeting in mice [24]. Particles of rod shape performed higher adhesion than spheric particles in endothelium [25]. Elliptical disk-shaped particles are not phagocytosed by macrophages [26]. Hence, an issue of aspheric alginate particles deserves to be studied.

In most cases, spheric alginate particles are available by use of a typical droplet formation approach in an attempt to minimize the interfacial free energy between particles and gelation solution [22, 27]. Scientists look forward to finding a simple way for the synthesis of aspheric alginate particles, and it is indicated that the shape of particles can be easily tuned by the interfacial tension [28]. Accordingly, various oil/water interfaces and polyelectrolyte complexation were

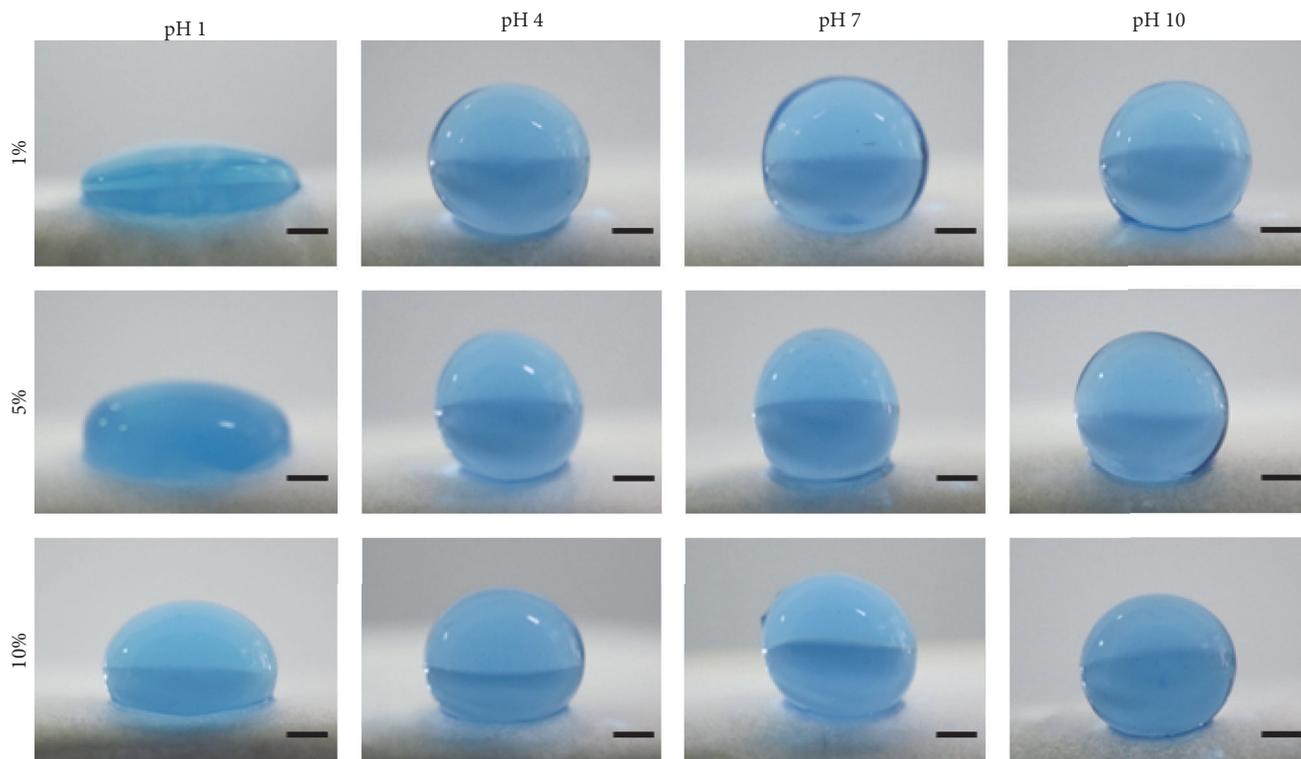


FIGURE 1: Comparison of alginate particle profile versus combinations of Ca^{2+} concentration and pH value in a 0.8% alginate solution. Scale bar = 1 mm.

used to change the interfacial tension so as to obtain aspheric particles [18, 23]. The pH value is found as another tunable parameter in alginate gelation process and is directly related to the interfacial tension [26, 29]. Surprisingly, there are few publications on the shape tailoring of aspheric alginate particles by tuning pH-induced interfacial tension. In this study, variation in interfacial tension is obtained by simply changing pH. Various shaped aspheric alginate particles are made by only tuning the pH values of gelation solutions, and their release behaviors are investigated as well.

2. Materials and Methods

2.1. Materials. Sodium alginate powder was purchased from Sigma Aldrich Chemical Co., Ltd. (Louis, USA), and Coomassie brilliant blue G-250 was obtained from One Star Biotechnology Co., Ltd. (Taipei, Taiwan). Barium chloride dihydrate and anhydrous calcium chloride were supplied from Eco Chemical Co., Ltd. (Taichung, Taiwan). Artificial gastric juice (pH 1.2) and intestinal juice (pH 7.5) were bought from Sigma Aldrich Chemical Co., Ltd.

2.2. Preparation of Alginate Particles. Respective alginate solutions were prepared in distilled water at concentrations of 0.8%, 1.6%, and 2% (w/v). The Coomassie brilliant blue G-250 was added separately into these three alginate solutions at a concentration of 0.05% (w/v) for comparative observation purposes. After homogenization, the alginate solution was filled into a disposable syringe (TERUMOR® Syringe, 3 mL)

and extruded by a KDS230 syringe pump (KD Scientific Inc., Holliston, MA, USA). The alginate droplets were cross-linked with BaCl_2 or CaCl_2 solutions to form alginate particles for 5 minutes. There are four predetermined pH conditions (pH 1, 4, 7, and 10) to address the pH effects on alginate particle shapes. Moreover, BaCl_2 and CaCl_2 cross-linkers were presented for three concentrations (1%, 5%, and 10% w/v). A digital camera (DP70, Olympus, Taiwan) was employed for imaging to estimate the morphology of alginate particles.

2.3. Release Behavior. Coomassie brilliant blue G-250 was used as a model drug to evaluate the drug release behaviors of the spheric and oblate alginate particles, and subsequently a comparison on the release behaviors is made between the gastric and intestinal cases. At specified time intervals, solutions were drawn to determine the released amount of Coomassie brilliant blue G-250 by a UV/VIS Spectrophotometer (HITACHI U1800, Japan) at a wavelength of 590 nm.

3. Results and Discussion

3.1. Effects of pH on Alginate Particle Shapes. To gain a clear understanding, pH effects on particle shapes for various alginate solution concentrations and various cross-linker types were made. Presented in Figures 1–3 are the photos of the particles, made from 0.8, 1.6, and 2% alginate solutions, cross-linked with respective concentrations of CaCl_2 . In Figure 1, higher aspect ratios (ratio of long radius to short radius) are seen in the pH 1 case than others, particularly

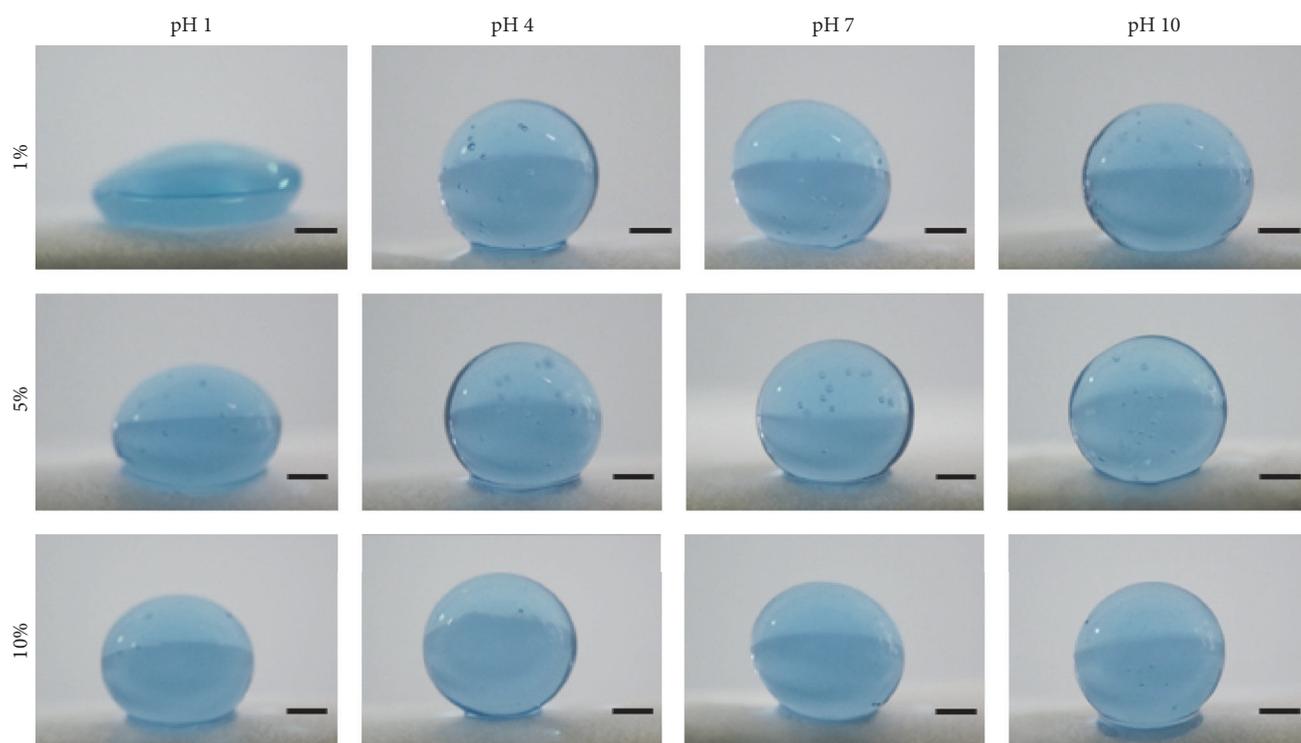


FIGURE 2: Comparison of alginate particle profile versus combinations of Ca^{2+} concentration and pH value in a 1.6% alginate solution. Scale bar = 1 mm.

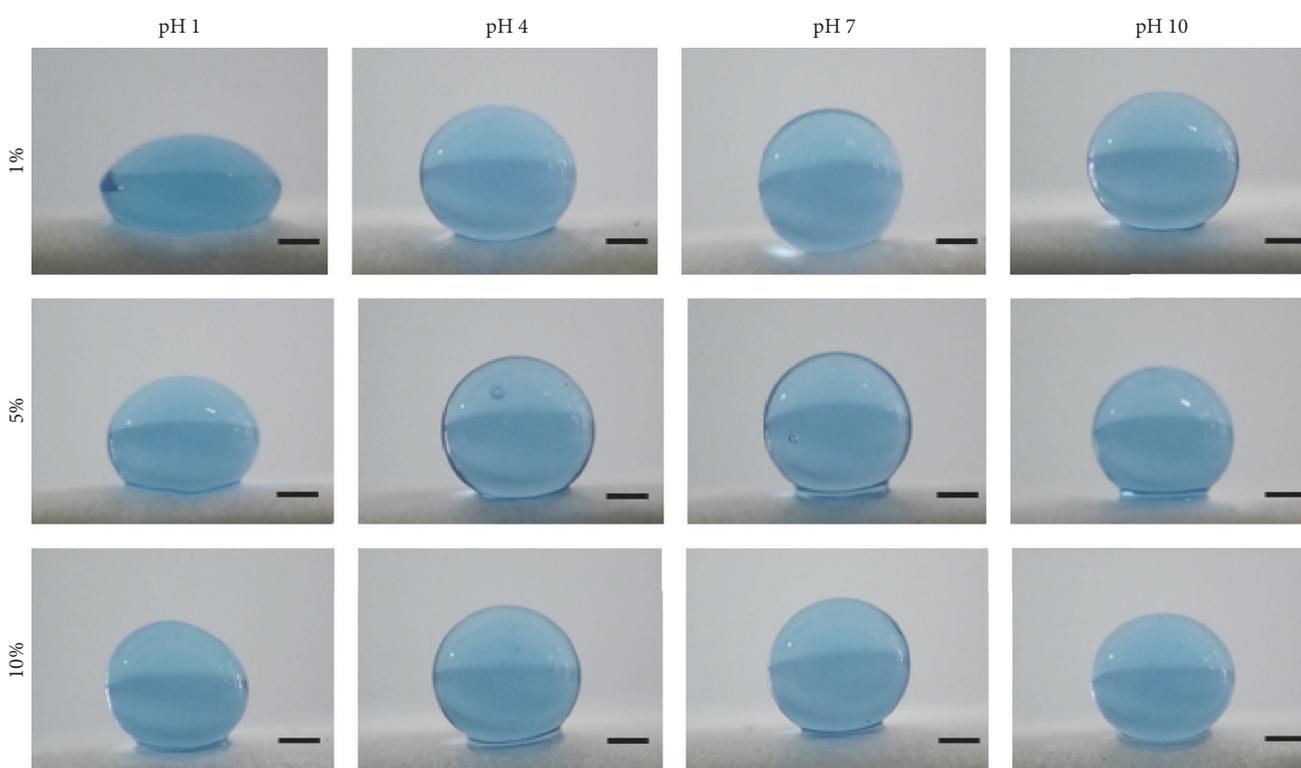


FIGURE 3: Comparison of alginate particle profile versus combinations of Ca^{2+} concentration and pH value in a 2% alginate solution. Scale bar = 1 mm.

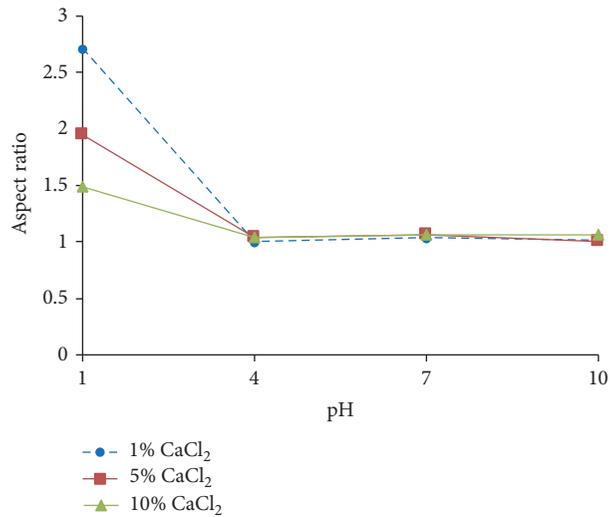


FIGURE 4: Comparison of aspect ratio (ratio of long radius to short radius) in different preparation parameters of 0.8% alginate particles.

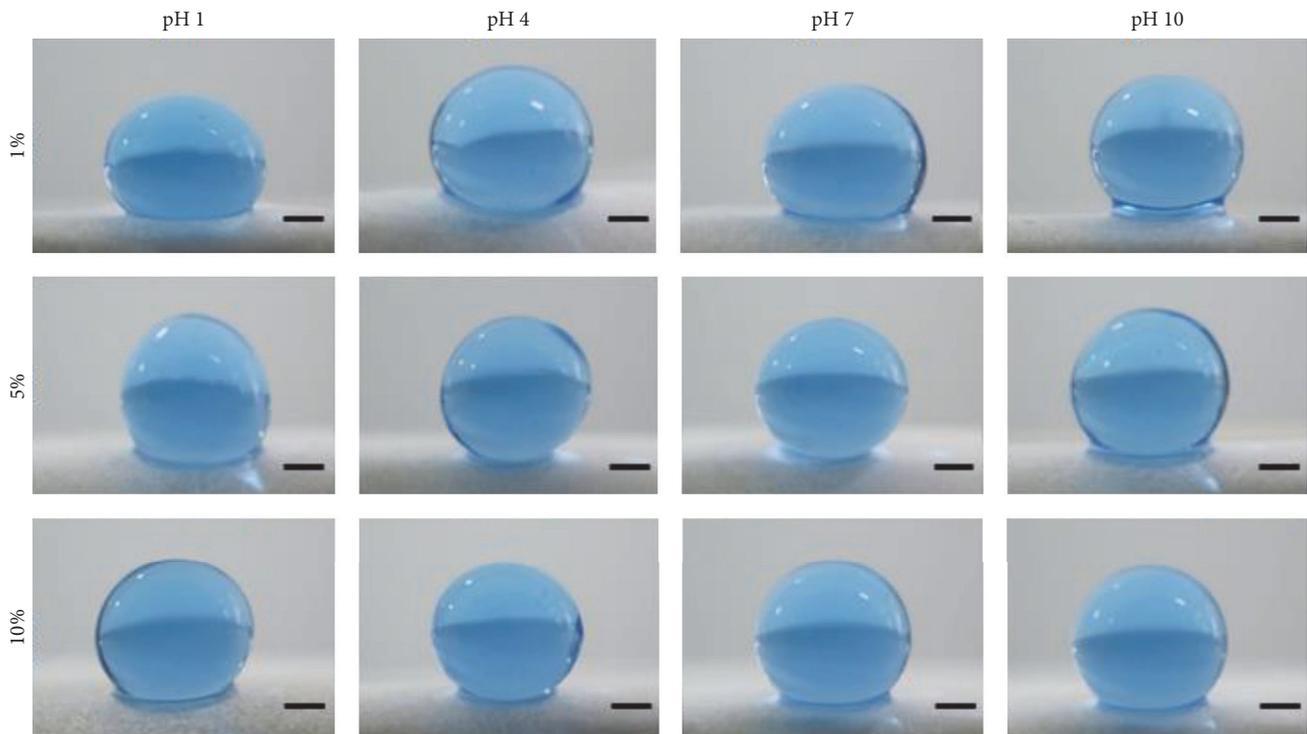


FIGURE 5: Comparison of alginate particle profile versus combinations of Ba²⁺ concentration and pH value in a 0.8% alginate solution. Scale bar = 1 mm.

for a low Ca²⁺ concentration. Figure 4 shows that the aspect ratio in Figure 1 increases with the decrease of pH value and the Ca²⁺ concentration, and the same observation applies to Figures 2 and 3. However, a low pH value demonstrates more significant effect on the particle shape at a low alginate and Ca²⁺ concentration. The alginate particle shape is tailored by the balance between the polymer viscosity and the interfacial

tension [26]. A lower pH gelation solution gives rise to a lower interfacial tension [30] and further results in an oblate particle with a higher aspect ratio.

Since gel-forming ability of alginate relies on the G-blocks binding divalent cations [31], there are an inadequate number of divalent cations in a low concentration of cross-linker to bind G-blocks and inadequate number of G-blocks in a low

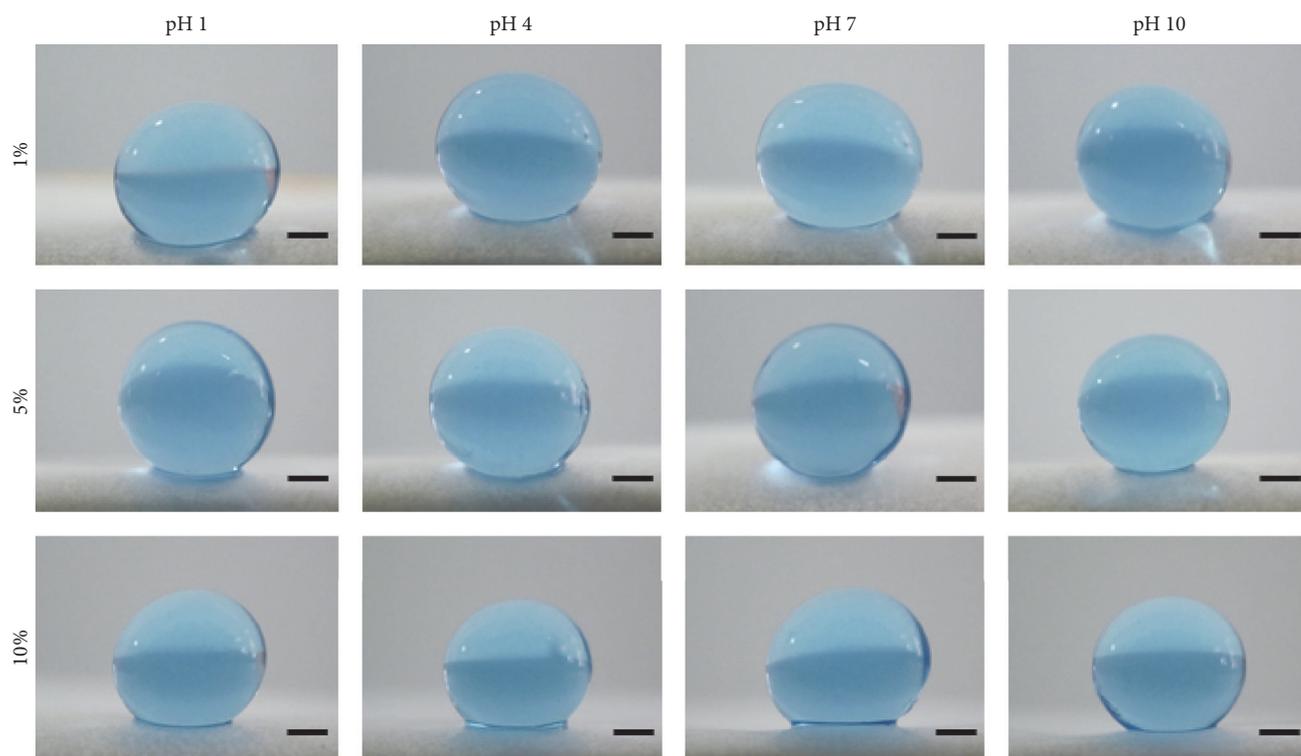


FIGURE 6: Comparison of alginate particle profile versus combinations of Ba²⁺ concentration and pH value in a 1.6% alginate solution. Scale bar = 1 mm.

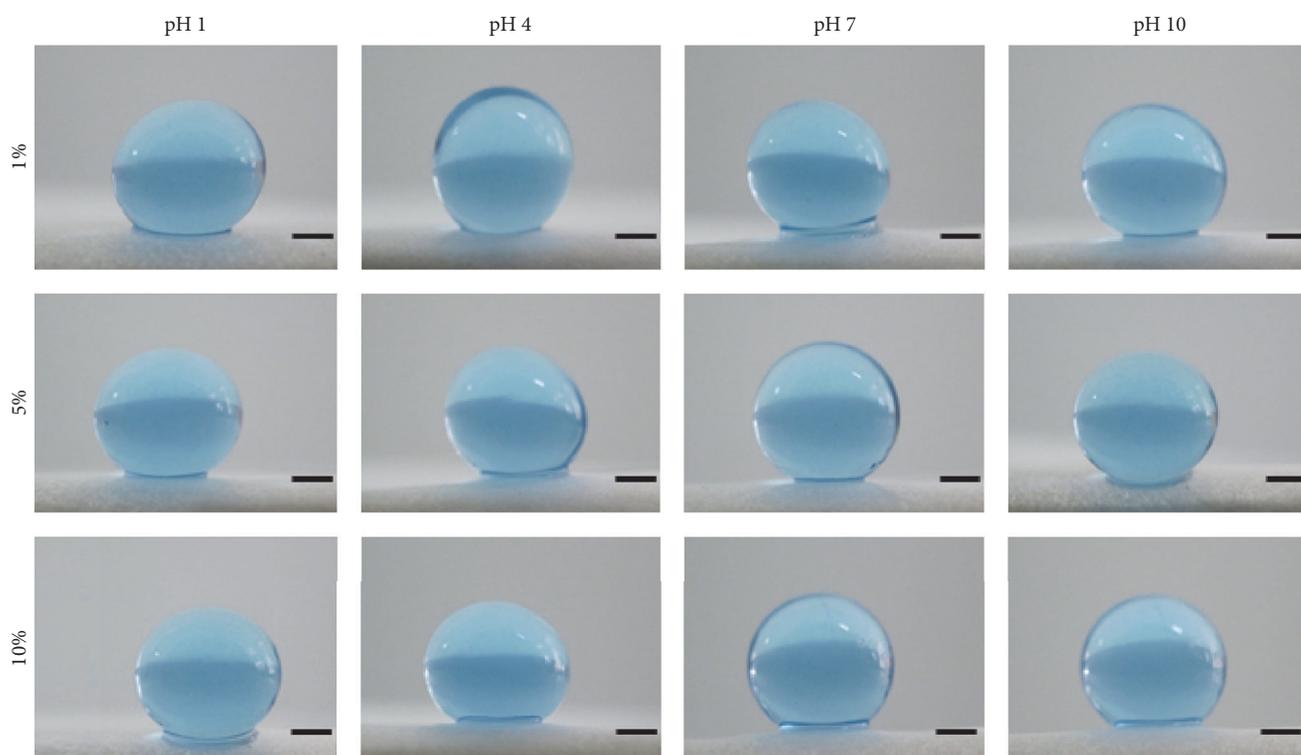


FIGURE 7: Comparison of alginate particle profile versus combinations of Ba²⁺ concentration and pH value in a 2% alginate solution. Scale bar = 1 mm.

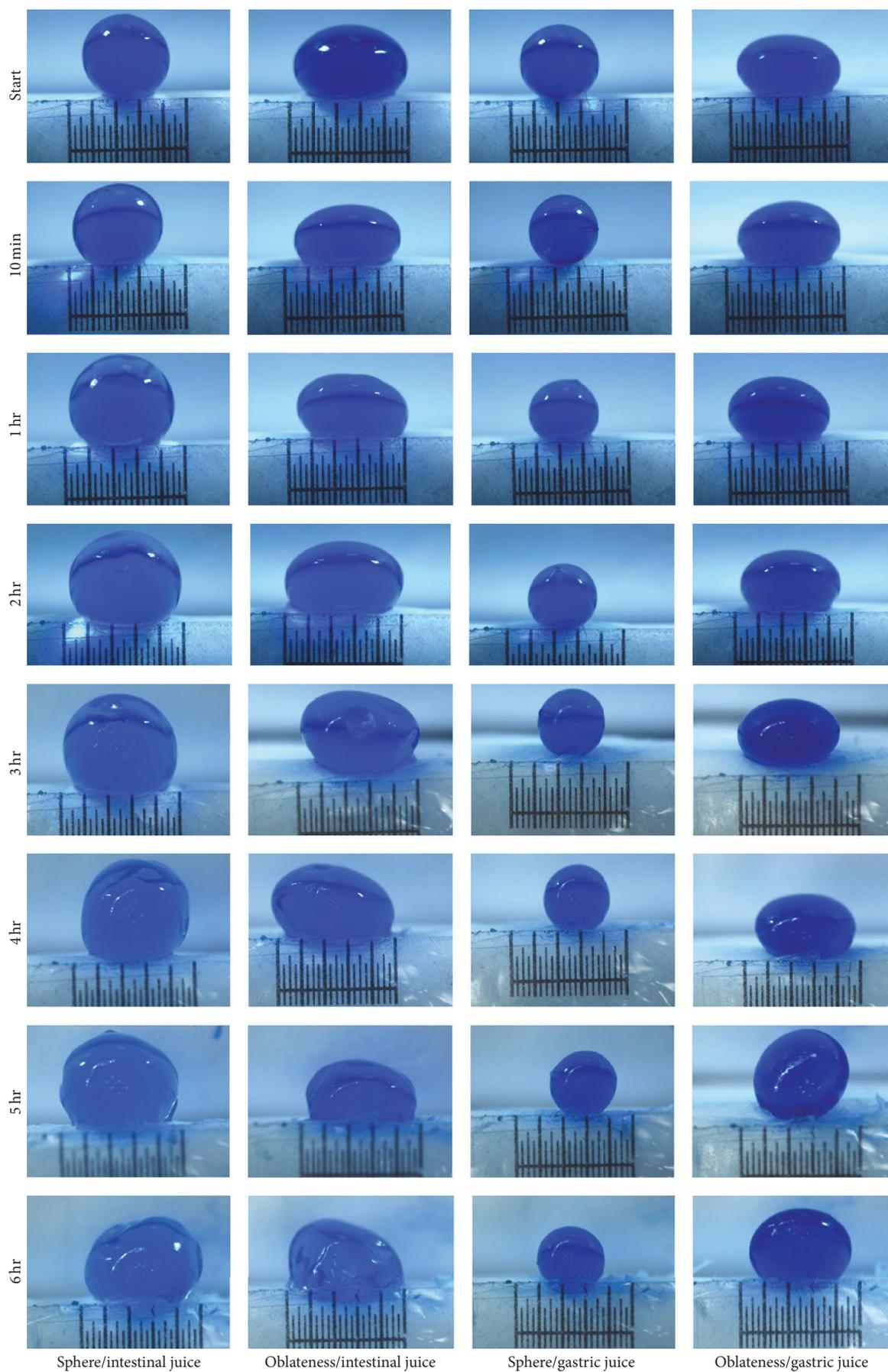


FIGURE 8: Dynamic shape changes of spheric and oblate alginate particles in intestinal and gastric juices.

alginate concentration for binding. Besides, there is a low viscosity for the dilute alginate solution. Hence, there exist oblate particles in low concentrations of alginate and cross-linker. This phenomenon is obvious at a low pH value, but not significant when pH is greater than 4.

Demonstrated in Figures 5–7 are the photos of the particles cross-linked with Ba^{2+} under the aforementioned alginate conditions, namely, 0.8, 1.6, and 2%. In contrast to those cross-linked with Ca^{2+} in Figures 1–3, there are no significant shape changes in Figures 5–7. This is simply due to the reason that alginate has a higher affinity with Ba^{2+} than with Ca^{2+} [32], and stable shaped particles are formed when cross-linked regardless of the change in the pH-induced interfacial tension.

3.2. Dynamic Shape Changes and Swelling Ratio in Gastric and Intestinal Juices. Figure 8 shows the dynamic shapes of oblate and spheric alginate particles immersed in gastric and intestinal juices for as long as 6 hours. Results indicate that the particles in the intestinal juice became easily collapsed, while in contrast there was no significant shape change in the gastric juice. Both spheric and oblate particles in intestinal juice swelled gradually over time and began to collapse 4 hours later. In a good agreement with previous reports, alginate particles disintegrated in alkaline, but not in acidic, media [33]. The pKa values of mannuronic and guluronic acid residues of alginate are measured to be 3.38 and 3.65, respectively [34]. With a pH value below the pKa of the uronic acid, alginate gels are stabilized by an intermolecular hydrogen bonding network [35]. Therefore, alginate particles are broken down easily in intestinal, but stable in gastric, juice.

Figure 9 reveals a swelling ratio comparison between alginate particles in gastric and intestinal juices. A higher swelling ratio of the particles is seen in the intestinal than in the gastric cases. In a good agreement with a previous study, there is a higher swelling degree in a simulated enteric than in a gastric environment [36]. The higher swelling ratio can be attributed to a chain expansion from the ionic carboxylate groups of alginate at a higher pH value [35].

3.3. Drug Release Behavior. Exhibited in Figure 10 are the release behaviors of Coomassie brilliant blue G-250 encapsulated in oblate and spheric alginate particles. The 1.6% alginate and 5% Ca^{2+} were used, and pH 1 and 7 were applied to obtain oblate and spheric particles, respectively. It is experimentally demonstrated that both oblate and spheric particles have a higher drug release rate in the intestinal than in the gastric juice. It is due to the change in the alginate microstructure in solutions under different pH conditions. As a pH-responsive polymer [37], alginate will shrink in a low pH medium but swells or further dissolves in a high pH one. Therefore, a relatively intact microstructure of alginate in the gastric juice yields a relatively slow release rate. In this study, a higher drug release rate is seen from oblate than from spheric particles, since an oblate particle is of a larger surface area than a spheric one [38]. As a consequence, the alginate particle shape is characterized as a significant parameter through drug release profile in pharmaceutical kinetics.

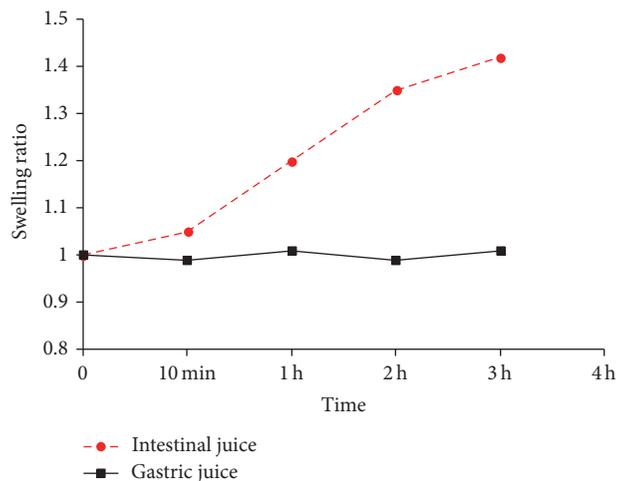


FIGURE 9: Swelling ratio comparison for spheric alginate particles between the intestinal and gastric cases.

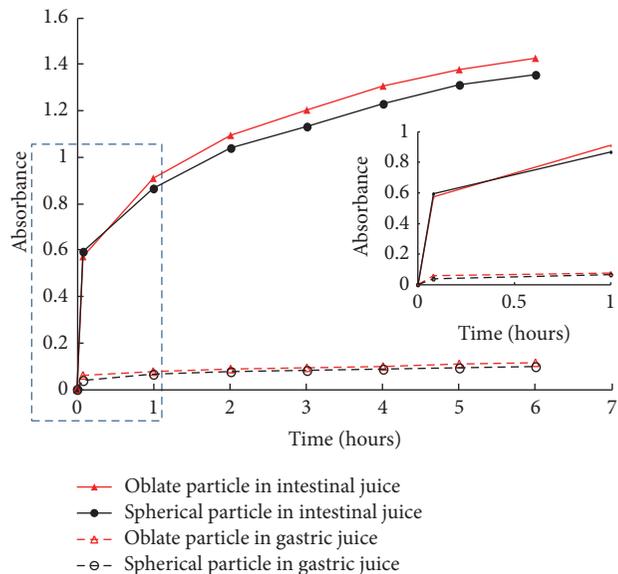


FIGURE 10: Release profile comparison of Coomassie brilliant blue G-250 encapsulated in spheric and oblate alginate particles immersed in the intestinal and gastric juices.

4. Conclusion

This study demonstrates pH-tuned shape tailoring of alginate particles and analysis on release behaviors. Alginate droplets contained in low pH gelation solutions tend to turn into oblate alginate particles. Oblate particles with a higher aspect ratio are experimentally demonstrated in lower concentrations of both alginate and Ca^{2+} cross-linker. In contrast, the Ba^{2+} cross-linker demonstrates little influence on the alginate particles. In intestinal juice, spheric and oblate particles swelled gradually over time and began to collapse 4 hours later. However, there was no significant shape change in the gastric juice cases. A higher release rate is seen in the intestinal than in the gastric juice. Additionally, a higher

release rate is demonstrated in the oblate case than in the spheric counterpart since a sphere has a smaller surface area than other shaped particles. This research finding could be seen as informative when applied to successive studies on the related alginate issue.

Competing Interests

The authors declare no conflict of interests.

Acknowledgments

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