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Stability of alginate encapsulation beads for microalgae cultivation

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Abstract

This research investigated the preparation of calcium alginate beads by encapsulation technique for algae cultivation for biofuel. The effects of calcium chloride ($CaCl_2$) to sodium alginate ratios of 3:0.5, 4:0.5, and 5:0.5 were tested for physical, chemical and biological stabilities. Solutions at pH of 2, 4, 7, 9, and, 11 were tested. The effects of different medium including tap water, fertilizer water (FER), effluent anaerobic digestor 5% dilution (EAD5) and 10% dilution (EAD10) with tap water, a raw domestic sewage (RSW) were evaluated. The results show that the bead can with stand most tested conditions except for biological stability test that use effluent from anaerobic digestor (60-80% stable). Further study on algae growth in those conditions ($CaCl_2$: SA ratio) is required.

Keywords: Algae, Wastewater from anaerobic digester, Growth rate, Calcium alginate, Cell encapsulation technique

1. Introduction

Fuel production from algae oil has become interested to public during the past decades as it is one of alternatives of renewable and sustainable energy. Oil from algae has advantages over terrestrial crops because the yield per unit area (95,000 L/ha) is higher than other terrestrial crops such as palm oil (5950 L/ha) [1]. Also it does not affect the crops that are made for food supply.

Algae has requires solar energy in photosynthesis and help decrease carbon dioxide as a greenhouse gas to create new algae cell. Another factor that affects the algae grow this the nutrient that use synthetic chemicals will increase the costs. Algae harvesting is one of the important aspect that also affects the overall algae biofuel production. Since the size of algae is relatively small and not easily settled, separating them could add the cost as well [2].

Cell immobilization and cell encapsulation are cell entrapment techniques that can be used to help with algae separation. Polymer substances (such as calcium alginate) and hardening agents (such as $CaCl_2$) are used to form the hydrogel bead where algae can grow inside [2]. As a result, the algae beads can be easily separated from water and later extracted for lipid. The conceptual design of algae beads cultivation in the reactor is shown in Figure 1. To prepare the alginate beads for algae cultivation for biofuel, the ratio of chemicals used to create the beads must be determined for stability in the conditions supporting the growth of algae. The strength of the bead itself should be tested without algae cell prior to cell addition. Therefore, this research aimed to determine the ratio of mixing chemicals to form encapsulated alginate beads. Also, testing the physical, chemical and biological stability of alginate beads formed under different ratio of chemicals.

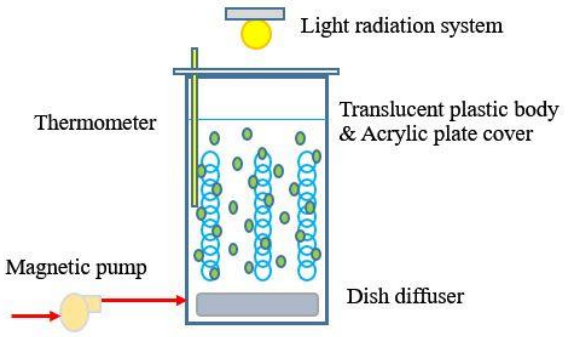

Figure 1 Conceptual design of encapsulated algae reactor

2. Materials and methods

2.1 Chemicals for alginate bead encapsulation

Alginate capsules (beads) are prepared from sodium alginate (SA) (CAS No. 9005-38-3, Sigma-Aldrich, MO, USA) solution (0.5 %w/v) and calcium chloride (Sigma-Aldrich, MO, USA) solution. To form alginate beads, $CaCl_2$solution was dropped into stirred sodium alginate solution (Figure 2).Once the beads were formed, they were strained and soak in 2%w/v $CaCl_2$ for 2 hours for hardening.

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Three concentrations of $CaCl_2$solution (3, 4, and 5% w/v) were used to prepare alginate beads.

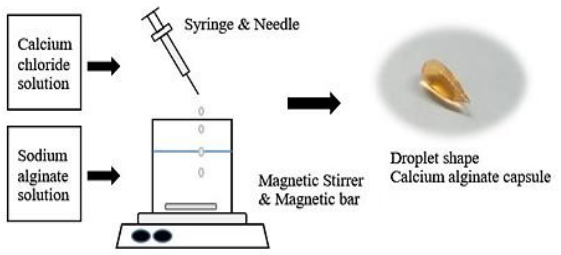

Figure 2 Alginate bead encapsulation process

2.2 Experimental setup and design

The experiments for testing alginate bead stability consist of 3 tests including physical, chemical, and biological stabilities. The tests were performed to determine which bead preparation condition could with stand the environmental condition that might be exposed during the algae cultivation. It was noted that this experiment is only preliminary test for the alginate beads preparation. Therefore, no algae cell was introduced in all of these tests. The stability of the beads with algae cell will be further investigated (not in this study) after the best condition is obtained.

2.2.1 Physical stability test

This test was performed to determine the effect of shear and lateral force between calcium alginate beads in the reactor. Twenty alginate beads and 15 of glass beads were placed in 50 mL of deionized (DI) water in 250-mL Erlenmeyer flask. The flask was then shaken at 100 rpm for 7 days. The alginate beads were observed daily to determine the breakage or damage. The experiments were triplicated.

2.2.2 Chemical stability test

Chemical stability test consist of two parts; 1) the effect of pH and 2) the effect of medium. For the effect of pH, solution pH of 2, 4, 7, 9, and 11 were investigated. Solution pH 2 solution was prepared by dissolving 7 g $KHC_4H_4O_6$ in 1 L of DI water and adjusted to pH 2 using 1 N H_2SO_4. Solution of pH4 was prepared from 10.12 g $KHC_8H_4O_4$ in 1 L of DI water. Solution of pH 7was the DI water and adjusted pH by1 N H_2SO_4 or 1 N NaOH. Solution of pH 9 was prepared from 3.80 g $Na_2B_4O_7.10H_2O$ in 1 L of DI water. Solution of pH 11 was made of 2.092g $NaHCO_3$ + 2.640gNa_2CO_3in 1 L of DI water. Then pH was adjusted by 1 N NaOH. The alginate beads were soaked in solutions for 1 week. The alginate beads were observed daily to determine the breakage or damage. The experiments were triplicated. For the effect of medium, tap water (TW), fertilizer water (FER), effluent anaerobic digestor 5% dilution (EAD5) and 10% dilution (EAD10) with tap water, a raw domestic sewage (RSW) were used since these media could be the potential substrate sources of algae cultivation. FER was prepared from commercial fertilizers: 46% urea and N-P_2O_5-K_2O (8-24-24) [3]. One liter of prepared medium contains 56 mg N, 12.4 mg P and 14.8 mg K.

2.2.3 Biological stability test

The effect of biological degradation potential of bacteria in an aqueous medium was tested. A series of experiments were conducted by two separate sets of sterile and non-sterile EAD10 (will be used to culture algae). EAD10 medium was sterilized by autoclaving at 121°C for 105 min. The beads were shaken incubation for 1 week and then observed for breakage or damaged. The experiments were duplicated.

3. Results

3.1 Physical stability

From the period of 1 week incubation shaking, it was found that the impact from shear force model was very little. No capsule alginate bead was damaged in all ratios of $CaCl_2$: SA (Figure 3a).

3.2 pH stability

The calcium alginate beads were immersed in solution pH of 24 7 9 and 11 for a period of one week. During this time, no damage or breakage was observed except at pH 11 (Figure 3b).

3.3 Effect of medium

The effect of medium on stability of the calcium alginate beads is shown in Figure 4. Results showed that the effect of medium to alginate capsules was low for TW, FER and RSW. The medium that caused beads damage were EAD5 and EAD10. Bead stability in EAD5 and EAD10 ranged from 60-80%.

Figure 3 Physical (a) and pH (b) stability of calcium alginate beads

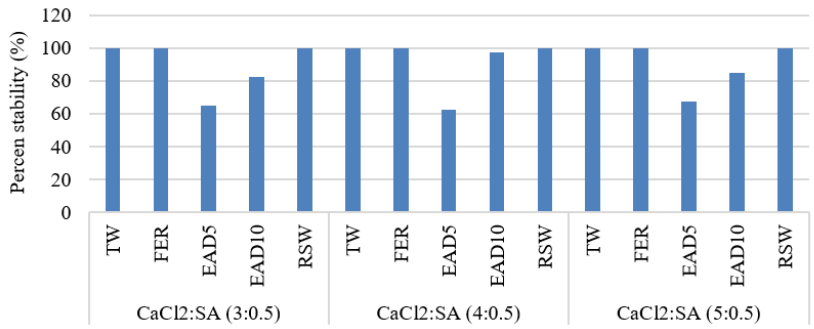

Figure 4 Effect of medium on stability of calcium alginate beads

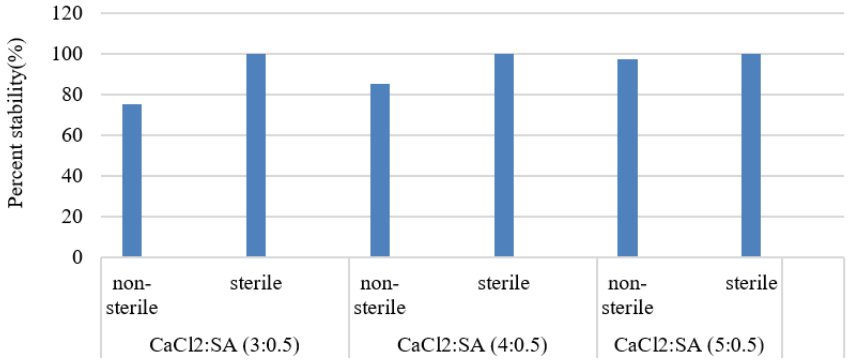

Figure 5 Biological stability of calcium alginate bead

3.4 Effect of biological degradation

From results in section 3.3, it has shown that EAD5 and EAD10 media affected calcium alginate capsules (Figure 5). It was speculated that biological might be play role in beads stability. The results show that the non-sterile EAD10 has microorganism that can damage the beads while the bead soaked in sterile EAD10 for 7 days has no damage.

4. Discussion

From the physical, chemical and biological stability tests, it was found that the calcium alginate beads were quite stable in all tested conditions except for EAD medium. The biological stability test proved that the damage of the beads could be from biological activity of microorganism in effluent of anaerobic digester. Higher $CaCl_2$ concentration tended to provide better stability under EAD (Figure 5). This suggested that one must be careful to use capsule algae using EAD as source of nutrients.

5. Conclusions

From this study, it was found that the calcium alginate beads prepared by an encapsulation technique have ability to withstand environmental condition for algae cultivation. Biological activity of indigenous organism could play a vital role in bead stability. The ratio of $CaCl_2$ to SA is important to biological stability as it increases more crosslink structure inside the bead. To determine which condition is suitable for algae cultivation and harvesting, further study of the algae growth in the beads needed to be conducted.

6. Acknowledgements

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7. References

[1] Gao Y, Gregor C, Liang Y, Tang D, Tweed C. Algae biodiesel - a feasibility report. Chemistry Central Journal 2012;6(S1):1-16.

[2] Lam MK, Lee KT. Immobilization as a feasible method to simplify the separation of microalgae from water for biodiesel production. Chemical Engineering Journal 2012;191:263-268.

[3] Thiansathit W, Keener TC, Khang KJ, Ratpukdi T, Hovichitr P. The kinetics of *Scenedesmus obliquus* microalgae growth utilizing carbon dioxide gas from biogas. Biomass and bioenergy 2015;76:79-85.