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Optimization of heterotrophic cultivation of *Chlorella* sp. for oil production

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HIGHLIGHTS

- ▶ A novel microalga *Chlorella* sp. LAM-H had a great potential to product lipid.
- ▶ Central composite design was used to optimize cultivation factors for bio-oil.
- ▶ The interactions between temperature and medium components were investigated.
- ▶ The model was found to be excellent in predicting the lipid productivity.
- ▶ The lipid productivity reached 247.16 mg l^{-1} d^{-1}, higher than other reports.

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ABSTRACT

The oleaginous microalga *Chlorella* sp. LAM-H screened from freshwater was proven to be a prospective feedstock for oil production according to its fatty acid composition. In order to enhance lipid production, response surface methodology (RSM) was used with central composite design (CCD) to optimize the heterotrophic cultivation of microalgae. The experiment results showed that a satisfactory second-order polynomial regression equation was achieved with a high coefficient of determination (R^2 = 0.9911) in analysis of variance. The effects of individual factors and their interactions on lipid productivity were successfully revealed. The greatest lipid productivity reached 247.16 mg l^{-1} d^{-1} under the optimal conditions of glucose concentration 26.2 g l^{-1}, sodium nitrate concentration 2.06 g l^{-1} and temperature 28.18 °C. Moreover, validation tests were performed and the results were very close to the predicted values. It was demonstrated that the obtained model was effective for predicting lipid productivity of the isolated microalga.

1. Introduction

Nowadays, the environmental problem is increasingly serious because of the overuse of non-renewable fossil fuels (such as petroleum, coal and natural gas) (Balat, 2010). To alleviate energy crisis and reduce environmental pollution, great efforts have been made to exploit alternative fuels. In this context, microalgae, as one of the biomass energy resources, have attracted considerable attention with advantages of fast growth, short production cycle, efficient oil production and low environmental demand (GuanHua et al., 2010; Hong et al., 2011). In particular, *Chlorella* sp. was reported to be potentially used as a candidate for biodiesel production (Hsieh and Wu, 2009; Miao and Wu, 2006). However, microalgal cultivation, especially in large scale, is discouraged by low cell density and high light requirement. Hence, the search for the approach to overcome these drawbacks is well expected for more efficient microalgal cultivation.

In comparison with photoautotrophy, heterotrophic cultivation allows microalgae to accumulate higher proportion of lipid within less time, and the scale-up process is much easier (Bumbak et al., 2011; Mohamed et al., 2011). Thus, it offers a potential pathway to produce lipid for large scale biodiesel production. Nevertheless, the production cost of biodiesel produced from microalgae is much higher than that of diesel derived from petroleum, due to the low efficiency of cultivation process (Cheng et al., 2009). During the process, the lipid production rate is significantly affected by several factors, such as temperature, concentrations of carbon and nitrogen (Brennan and Owende, 2010; Dean et al., 2010; Queiroz et al., 2011). Therefore, it is necessary to study the optimal conditions of heterotrophic cultivation to achieve high lipid productivity.

To develop the process technology in microalgal cultivation, single-factor experiment was applied to optimize the cultivation conditions of microalgae (Ruangsomboon, 2012). However, the traditional method is time-consuming and has a poor performance for optimizing a large number of parameters. Besides, it might lead to erroneous conclusions, since interactions between factors are

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