



Kinetic Model of *Ulva sp* Protein Extraction using Ultrasound-Assisted Osmotic Shock Method

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Abstract. *Ulva Sp* is a type of macroalgae that contains 13-26% protein. This works is to study kinetic of *Ulva sp* protein extraction using Ultrasound-Assisted Osmotic Shock Method. The *ulva sp* powder and Na-Sulfate-ethanol solution in a ratio of 1:10 (g/mL) were fed into a set of extractor unit in a presence of ultrasound wave for 30 minutes. The variables extraction was particle size (60, 80, 100 mesh) and extraction time (10, 15, 30 minutes). Supernatans thus was separated and purified from precipitates to analyze protein content using Bradford method. The maximum protein yield obtained is 61.98 % with an operating condition of particle size of 100 mesh and extraction time of 15 minutes. The diffusion coefficient empirically was formulated as $De = 3 \times 10^{-15} \times (\text{particle size})^{0.6}$

Keywords: Ultrasound-assisted extraction, protein yield optimization, diffusion coefficient analysis

INTRODUCTION

Ulva Sp. have a considerable protein content of 10-43% w / w, while the protein content from traditional sources is (10-30 % w / w). Commonly, *Ulva Sp* proteins comprise amino acids such as asparagine, glutamate, and aspartate. These proteins and amino acids have significant benefits for human health [1,2].

Physical and chemical extraction was the most commonly used method during this time. Physical extraction requires a long time and high energy to break down the cell wall. Meanwhile, chemical methods require high temperatures to break down cell walls. High temperatures and chemical compounds eventually cause damage to proteins. Consequently, an effective extraction method is needed to obtain protein from algae and does not damage its proteins and amino acids. The ultrasound-assisted osmotic shock extraction is a method that applies ultrasonic and osmotic shock technology [2-7]. This extraction method is faster and more efficient because ultra-sonic waves facilitate the osmotic shock extraction process, which produces enormous energy that hits the cell wall. The collision causes cell disruption, thus facilitating and speeding up the diffusion process [1-3].

This work aims to determine the extraction conditions that provide maximum protein yield and study the extraction kinetics based on diffusion models.

METHOD

Materials

The *Ulva Sp* was obtained from the farmers on the north coast of Situbondo. Before being used, the *Ulva sp* was washed, dried and milled to a specified size. The solvent used was ethanol obtained from Sigma Aldrich Indonesia. The Aspartate, glutamate, and arginine were also purchased from Sigma Aldrich Indonesia.

Ultrasound-Assisted Osmotic Shock Extraction

The *Ulva Sp* was obtained from the farmers on the north coast of Situbondo. Before being used, the *Ulva sp* was washed, dried and milled to a specified size. The solvent used was ethanol obtained from Sigma Aldrich Indonesia. The Aspartate, glutamate, and arginine were also purchased from Sigma Aldrich Indonesia.

Variables Extraction

The variables for extraction are particle size (60, 80, 100 mesh) and extraction time (10,15, and 30 minutes).

Protein Analysis

Protein analysis uses the Lowry and Bradford methods. The calibration curve is prepared using a standard bovine albumin solution in the 0-1500 µg/mL concentration range. After that, the sample was added 100 µL of Folin-Ciocalteu reagent and incubated for 30 minutes. The quantification process will be carried out using UV-Vis spectrophotometers at 260 nm and 280 nm wavelengths.

Kinetic Model

The extraction kinetic employs diffusion-controlled model.

$$\frac{1}{De} \frac{dy}{dx} = \frac{d^2C}{dr^2} + \frac{2}{r} \frac{dC}{dr} \quad (1)$$

in which C is the concentration of oil at the position of r and time of t.

RESULT AND DISCUSSION

Effect of Variable Extraction on Protein Yield

The effect of variable extraction on protein yield was studied in range of 10-30 minutes of extraction times and particle diameter of 60-100 mesh. Based on FIGURE 1 and FIGURE 2, it can be seen that extraction time shows a predominant effect on yield. As extraction increased, the contact time between solvent and cell matrix is getting longer. The solvent diffuse into cell matrix and push out the protein from cell matrix. Thus, yield also increases. Meanwhile, the particle diameter of 100 mesh provide maximum yield than 60 and 80 mesh. It is obviously that smaller particle size facilitates large contact area. Thus, rate of diffusion run faster. Furthermore, the optimum condition, was established at extraction time of 15 minutes and particle diameter of 100 mesh, gained protein yield of 61.98 wt%.

Extraction Kinetic

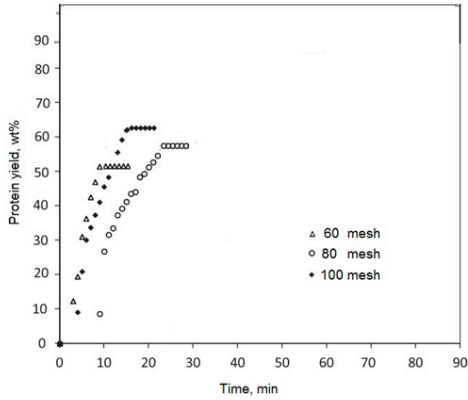


FIGURE 1. The relationship between protein yield and extraction time on various particle diameter

This work employs diffusion controlled model to study extraction kinetic. As seen on FIGURE 3, the kinetic study showed that the diffusion coefficient (D_e) using diameter particle of 100 mesh is larger than 60 and 80 mesh. It indicates that mass transfer using smaller particle is higher than the extraction using bigger particle size. Eventually, based on the experimental data, the diffusion coefficient empirically was formulated as $D_e = 3 \times 10^{-15} \times (\text{particle size})^{0.6}$.

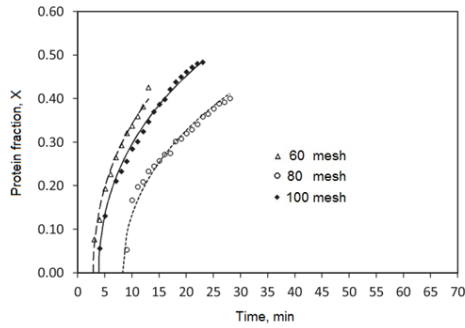


FIGURE 2. The relationship between protein fraction and extraction time on various particle diameter

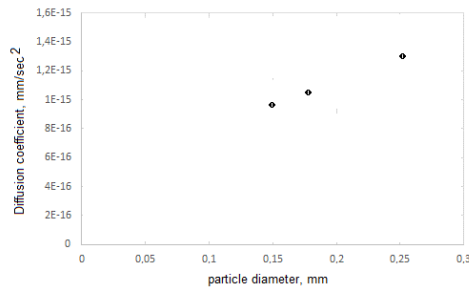


FIGURE 3. The relationship between diffusion coefficient and particle diameter

Particles with a smaller size have larger diffusion constant while a larger size have smaller diffusion constant. This implicitly informs the smaller particle perform a faster diffusion rate than a bigger particle.

CONCLUSION

The extraction of *Ulva sp* protein using ultrasound-assisted osmotic shock method gained protein yield of 61.98 % at optimum condition of 15 minutes and particle diameter of 100 mesh. The kinetic study empirically indicates that the diffusion coefficient (D_e) is strongly influenced by particle diameter. Mathematically is expressed as $D_e = 3 \times 10^{-15} \times (\text{particle size})^{0.6}$.

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