

The Biosorption of Copper Metal Ion by Tropical Microalgae Beads Biosorbent

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The Biosorption of Copper Metal Ion by Tropical Microalgae Beads Biosorbent

Tiara Wilan, Nanda Astuti Lieswito, Amelia Suwardi, Rositayanti Hadisoebroto, Melati Feranita Fachrul, Astri Rinanti

Abstract: This study aims to remove copper (Cu²⁺) from wastewater with a biotechnological approach using microalgae beads mixed culture of *Chlorella vulgaris*, *Chlorococcum* sp., and *Scenedesmus obliquus*. The effect of contact time and pH was examined to achieve the greatest efficiency in the Cu²⁺ metal removal. The exponential phase of mixed microalgae cultures occurred on the 7th day. In the exponential phase, microalgae can be harvested and made as biosorbent. Biosorbent is made into beads by mixing 0.5 g of dead biomass/ gr (Na-alginate polymer). These microalgae beads are contacted with a solution containing Cu²⁺ in the pH ranges and contact time 0-180 minutes. The highest absorption efficiency of Cu²⁺ is 98.56% obtained at pH 5, 180 minutes of contact time, at 25 °C, and with an initial waste concentration of 25 mg/L. The biosorption process in this study follows Langmuir Isotherm model with an R² value of 0.9994. It is proven that the biosorbent beads of mixed culture microalgae can be used as an environmentally friendly method to control heavy Cu²⁺ metals in the waters.

Index Terms: Beads, Biosorption, Biosorbent, Copper (Cu²⁺), Heavy Metal, Microalgae, Removal

1. INTRODUCTION

Heavy metals with high concentration are harmful to living the aquatic environment because it can disrupt aquatic creatures. They are generally found in the waters because of ecosystems due to the inhibition of sun penetration as a raw their mobility on its surface [1]. Some heavy metals often material for producers in photosynthesis so that it will cause an found in the waters are Cu, Zn, Cr, Cd, Pb, Ni, and Ag. Copper imbalance of aquatic ecosystems [10]. The abundance of Cu²⁺ ion naturally enters the environment through various microalgae can be used as a heavy metal absorbing biomaterial natural phenomena such as from the weathering result of [11]. However, a microbe with the most significant ability in dissolve minerals contained in rocks, and unnaturally through absorbing heavy metals is microalga, which has a functional a variety of human activities, for instance, the entry of group capable of binding metal ions, especially carboxyl, amine, electroplating industry wastewater [2]. According to [3], metal sulphate, and sulphonate functional group located in the cell Cu²⁺ tends to be toxic assuming it enters an organism's body wall inside the cytoplasm [4]. The advantages associated with in large amount or exceeds their tolerance limit, thereby, using microalgae are due to its environmentally friendly disrupting the brain, skin, liver, pancreas, and myocardium. biological materials which are recyclable, and its cheap Furthermore, It also becomes a poison when the concentration maintenance cost [12]. According to [13], the adsorption of is above 20 mg/kg [4] and tends to cause diseases such as heavy metal ions using microorganisms is reversible and the neurotoxicity, acute toxicity, dizziness, and diarrhea [5]. process occurs quickly. The biosorption process utilizes living or Various methods have been conducted to separate heavy dead biomass because it occurs on the surface of the microalga metals from water, for instance, the physical adsorption cell wall. However, using dead biomass is more advantageous method, chemical sedimentation, mechanical filtration, and ion because it does not need nutrient supply regularly for growth exchange. However, some of those methods have [14].

disadvantages, such as causing secondary pollution as a Many factors affect the biosorption process towards heavy result of using chemicals and its cost-effectiveness [6]. One of metals some of these are the biomass characteristics, the environmental friendly alternative methods used to temperature, pH, initial waste and biosorbent concentration, overcome the pollution problem is biomass or microorganism, contact time and width of the surface area. The immobilization also known as biosorption [7]. The advantages of using this of microalgae biomass is used to avoid the reactor clogging method are the high efficiency in decontaminating wastewater [15]. Immobilization is the physical absorption of biomass or cell localization into the polymer [16]. Alginate chitosan, cellulose, polyurethane, polyvinyl alcohol, acrylic, and gelatin are some polymers used to immobilize biomass [17]. However, in this research, it is used as a polymer. This research aims to obtain the highest biosorption efficiency of heavy metal Cu using immobilized microalga by optimizing the environment condition during the process.

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Furthermore, the biomasses used for biosorption are microalgae, bacteria, fungi, and plants [9]. Microalgae is one of

the biomass which can also cause water problems such as blooming algae. Algae blooming can have a negative impact on ecosystems due to the inhibition of sun penetration as a raw their mobility on its surface [1]. Some heavy metals often material for producers in photosynthesis so that it will cause an found in the waters are Cu, Zn, Cr, Cd, Pb, Ni, and Ag. Copper imbalance of aquatic ecosystems [10]. The abundance of Cu²⁺ ion naturally enters the environment through various microalgae can be used as a heavy metal absorbing biomaterial natural phenomena such as from the weathering result of [11]. However, a microbe with the most significant ability in dissolve minerals contained in rocks, and unnaturally through absorbing heavy metals is microalga, which has a functional a variety of human activities, for instance, the entry of group capable of binding metal ions, especially carboxyl, amine, electroplating industry wastewater [2]. According to [3], metal sulphate, and sulphonate functional group located in the cell Cu²⁺ tends to be toxic assuming it enters an organism's body wall inside the cytoplasm [4]. The advantages associated with in large amount or exceeds their tolerance limit, thereby, using microalgae are due to its environmentally friendly disrupting the brain, skin, liver, pancreas, and myocardium. biological materials which are recyclable, and its cheap Furthermore, It also becomes a poison when the concentration maintenance cost [12]. According to [13], the adsorption of is above 20 mg/kg [4] and tends to cause diseases such as heavy metal ions using microorganisms is reversible and the neurotoxicity, acute toxicity, dizziness, and diarrhea [5]. process occurs quickly. The biosorption process utilizes living or Various methods have been conducted to separate heavy dead biomass because it occurs on the surface of the microalga metals from water, for instance, the physical adsorption cell wall. However, using dead biomass is more advantageous method, chemical sedimentation, mechanical filtration, and ion because it does not need nutrient supply regularly for growth exchange. However, some of those methods have [14].

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2 MATERIAL AND METHODS

2.1 Materials

Materials that used for this research are: *Chlorella vulgaris*, *Scenedesmus obliquus*, dan *Chlorococcum* sp., aquades, Na-

Alginate, $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, alcohol, HCl and Microalgae Growth materials: KNO_3 , KH_2PO_4 , $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, EDTA, $\text{FeCl}_2 \cdot 6\text{H}_2\text{O}$, ZnCl_2 , H_3BO_3 , $\text{CaCl}_2 \cdot 6\text{H}_2\text{O}$, $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, and $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$

2.2 Microalgae Cultivation

Microalgae *Chlorella Vulgaris*, *Chlorococcum* sp., and *Scenedesmus obliquus* were grown in the batch culture using an artificial growth media PHM (Provasoli Haematococcus Media). Its cultivation was carried out on the 30 L photobioreactor and 1 L Erlenmeyer flask, with a microalgae and PHM media ratio of 1:9. The environment condition was set at room temperature of $25 \pm 2^\circ\text{C}$, pH 6, lighting intensity of 3500 lux, and an airflow rate of 0.3 Liter/second. The growth of this mixed culture microalgae was determined using the optical density method, with the absorbance value measured using UV-Vis spectrophotometer using a wavelength of 680 nm. After obtaining the absorbance value, the growth curve could be used to obtain the exponential phase.

2.3 Microalgae as Biosorbent Preparation

The cultivated microalgae was then harvested and separated

using the centrifugation method at the speed of 4000 rpm for 20 minutes. The separated biomass was then dried in an oven for 24 hours at a temperature of 80°C till a microalgae powder was obtained and ready to be immobilized. The resulted microalgae powder was then weighed and mixed with sodium alginate (2% w/v) dissolved in 200 ml of demineralized aqua using a magnetic stirrer. After mixing it completely, the solution was dropped using a syringe into $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ 0.1 M solution and stirred concurrently, using a magnetic stirrer. The beads were then moved into CaCl_2 5 mM solution and stored in a refrigerator for 24 hours to perfect its formation. After 24 hours, the beads were washed with distilled water, stirred using a magnetic stirrer for 30 minutes and filtered on a cloth.

2.4 Artificial Cu^{2+} Waste Preparation

The artificial wastewater made of sulphate copper ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) was dissolved in distilled water till the concentration reached 1000 ppm as the main solution. Furthermore, the determined concentration of Cu^{2+} was dissolved based on the concentration needed with the pH value set by adding HCl 0.1 N or NaOH 0.1 N solution.

2.5 Biosorption with pH and Contact Time Optimization

The pH was set using HCl and NaOH solution until it recorded values of 4, 5, and 6. Furthermore, the concentration of Cu^{2+} solution in the waste was 25 mg/L, while the weight of the biosorbent beads was 10 grams and the temperature $25 \pm 2^\circ\text{C}$. The biosorbent beads and waste were contacted in the 1500 ml photobioreactor, with aeration given from the bottom using a hose to make contact between the biosorbent beads and waste solution. This process was carried out within the contact times of 60, 120, and 180 minutes. After the biosorption process, metal Cu^{2+} was analyzed using AAS (Atomic Absorption Spectrophotometry).

2.6 Removal and Desorption of Cu^{2+}

After AAS analysis, the removal efficiency of Cu^{2+} can be counted using the equation below:

$$\% \text{Removal Efficiency} = \left[\frac{C_{1a} - C_{1b}}{C_{1a}} \right] \times 100\% \quad (1)$$

C_{1a} = Initial Waste Concentration of Cu^{2+} ; C_{1b} = Final Waste Concentration of Cu^{2+}

$$\log q_e = \log k + \left(\frac{1}{n} \right) \log C$$

2.1 Isotherm Adsorption

The adsorption isotherm study showed the capacity of the biosorbent beads required to carry out the adsorption. This is, however, explained by the isotherm of equilibrium sorption which is indicated by a certain constant which expresses the surface and affinity characteristics of the biosorbent. This research utilizes two models of isotherm, namely Langmuir and Freundlich.

In Langmuir, when C is plotted against C/QE, constant b and QM are obtained. The value of 1/b indicates the slope, while 1/qm.b is the intercept. The maximum capacities of adsorption and qm are obtained from the intercept and slope [18]. This model of this equation is:

$$C/q_e = (1/b \cdot q_m) + (C/q_m) \quad (2)$$

C is the concentration of the waste after the biosorption process (mg/L), q_e is the amount of adsorbed copper ion (mg/g), b is the coefficient of Langmuir, and q_m is the adsorption capacity of the biosorbent beads (mg/g).

The Freundlich isotherm model uses the assumption that adsorption runs in physics. The equation required to obtain its

constants:

$$q_e = k \cdot C^{1/n} \quad (3)$$

and n are Freundlich adsorption capacity (mg/g) and adsorption intensity.

3. Results and Discussion

3.1 Cultivation of Microalgae

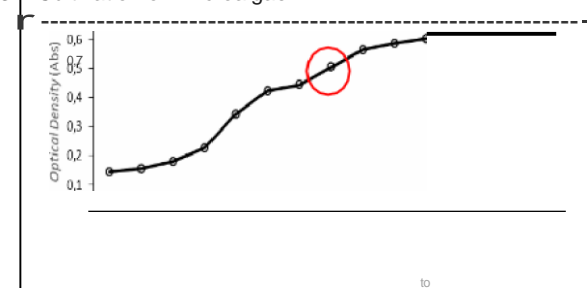


Fig. 1. The Microalgae Cultivation Graph

In the exponential phase, the photosynthesis activity occurred high to form protein during the growth period (19). The growth curve illustrated in Figure 1, shows that the mixed culture microalgae reached the exponential phase from the 3rd to 11th day. The harvesting of the microalgae was conducted on the 7th day, which was in the middle of the exponential phase, using centrifugation and biofloculation methods. This was carried out during this period because the greatest increase in biomass occurred on that day. The harvesting of the microalgae was in the middle of the exponential phase with the aim of obtaining the most biomass and the assumption that the walls of microalgae have been fully formed during growth, so that they are ready and can be used as biosorbents when absorbing heavy metals. The results were then dried and made as beads to be used in the biosorption process.

3.2 pH and Contact Time Optimization

The adsorption of Cu^{2+} ion in the biosorption process as a pH and time function is shown in Figure 2. The pH value and contact time are some of the factors that influence the biosorption process.

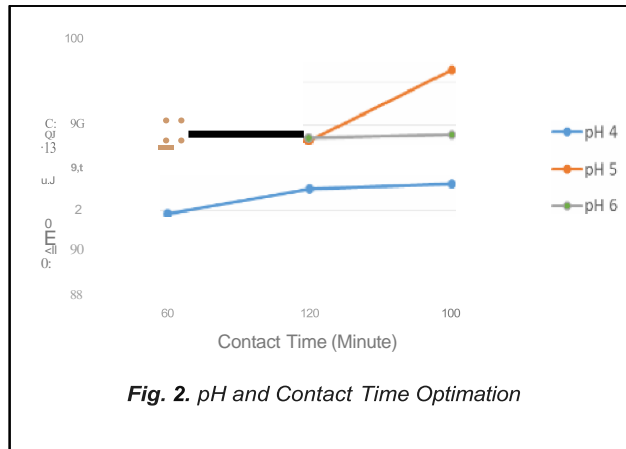


Fig. 2. pH and Contact Time Optimization

The removal efficiency of Cu^{2+} at pH 4 tended to be higher, at the contact times of 120 and 180 minutes (Fig 2). On the other hand, at pH 5 the removal efficiency at contact time of 60 minutes and 120 minutes tended to show similar number and when it reached contact time of 180 minutes, the efficiency increased significantly. However, pH 4,5, and 6 shows great removal efficiency of Cu^{2+} with value above 90%. This happens because at pH above 3, electrostatic attraction between copper ions and algal with negative charged cells surface would occur which will enhance the adsorption (20). The maximum absorption value of Cu^{2+} occurred at pH 5 with a contact time of 180 minutes, and elimination efficiency above 98.56%. When compared with the research result of a researcher (21) who used a mixed culture of *S. cerevisiae* and *Chlorella* sp. immobilized with pH 4 and contact time of 120 minutes to absorb Cu^{2+} with the elimination efficiency of 81.05%, this research result is better and more efficient in absorbing Cu^{2+} . The optimization of pH in this research showed the most effective value for a biosorbent to absorb Cu^{2+} ion of copper. According to (22), pH affects the protonation of the metal-binding site exposed by the cell surface. As seen in Figure 2, the beads of mixed microalga appeared to provide less efficient elimination at pH 4. This was possible because its three types failed to work optimally at a lower pH value. At pH 4, the removal efficiency increases continually on the elimination efficiency. This means that from 60 to 120 minutes, it increased up to 1.23%, thereby, gaining up to 0.24% at the contact time of 180 minutes. This proves that the longer the biosorption process conducted, the better

the elimination efficiency obtained.

3.3 Adsorption Isotherm

According to the calculation using the equation (2), the R^2

value obtained for the Langmuir isotherm (Figure 3) in this research was 0,9994. Figure 4 shows the Freundlich constant

and the coefficient of correlation regression from the copper biosorption process in this research. The R^2 values for the Freundlich isotherm were 0,9054.

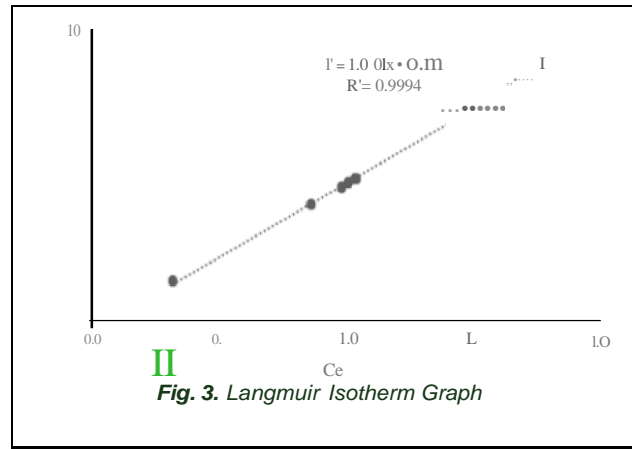


Fig. 3. Langmuir Isotherm Graph

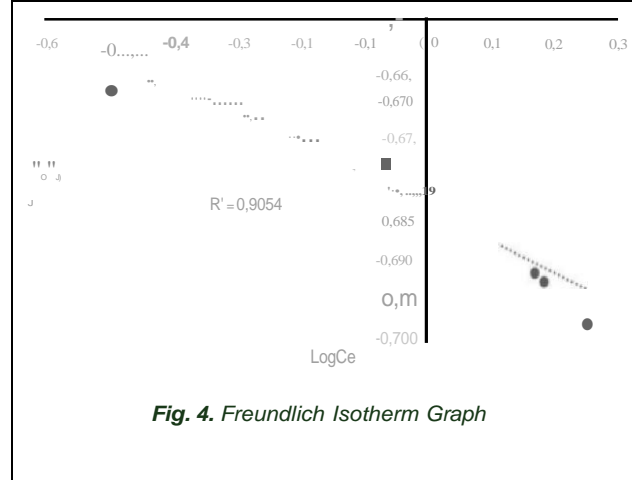


Fig. 4. Freundlich Isotherm Graph

The constant value of both isotherm models are shown in Table 1. Langmuir isotherm shows the adsorption capacity of this process is 0,1972 mg/g and b indicates Langmuir constant with the value of 21,797. For Freundlich isotherm model, k indicates the adsorption capacity with value of 0,2073 mg/g and n value of 25,380.

TABLE 1
LANGMUIR AND FREUNDLICH CONSTANT VALUE

Isotherm	Constant	Value
Langmuir	qm	0,1972
	b	21,797
Freundlich	K	0.2073
	n	25,380

The R^2 value is used as the criteria for both models of isotherm (23). Its value for Langmuir isotherm was bigger than Freundlich. Therefore, it indicated that the biosorption process following Langmuir isotherm was proof that it stops at one monolayer, and consistent with the functional binding (24).

4 CONCLUSION

The biosorbent beads of mixed culture microalga *Chlorella*

Vulgaris, *Chlorococcum* sp., and *Scenedesmus obliquus* were capable of acting as an effective biosorbent to reduce Cu^{2+} in water. The microalgae reached the exponential phase on 7th day, which shows that the mixed microalgae is capable of living synergistically. The maximum percentage of Cu^{2+} elimination using microalgae beads occurred at pH 5 with a contact time of 180 minutes. The biosorption process is properly explained well using the Langmuir isotherm model with the R value of 0.9994, q_m of 0.1972 mg/g and b constant of 21,797.

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