Biosorption of Copper (II) Ions using Living *Chlorella sp.* from Aqueous Solution

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Abstract. Liquid waste from the electroplating industry is classified as hazardous and toxic waste. It contains a lot of heavy metal ions which are toxic and even carcinogenic so that this waste has to be treated first. In this present study, heavy metal waste treatment using biosorption method was investigated. Microalgae *Chlorella sp.* was used as biosorbent. The experimental was conducted using copper (II) (Cu²⁺) ions which the initial concentration was varied at 20 to 80 ppm. pH scale was also varied at 2 to 5. After several hours, sample was taken and analyzed with UV–vis spectrophotometer. The experimental results showed that the best percentage removal of Cu²⁺ ions was 96.10% which was achieved at 40 ppm and pH 5.

1. Introduction

Metal coating industries, usually called electroplating industries, are developing along with the increasing demand for metal–coated products and metal alloys. However, the rise of this industry not only has a positive impact but also causes a negative impact which generates liquid waste during the electroplating process. Although the amount of electroplating waste is small, this waste is classified as hazardous and toxic waste because it contains heavy metal ions, such as copper (Cu), nickel (Ni), chromium (Cr), iron (Fe) and manganese [1]. The content of this heavy metal is toxic; even some elements are carcinogenic. It means this waste has to be processed first until the environmental quality standards before it discharges into the environment.

On the industrial scale, the heavy metal liquid waste treatment can be carried out physically, chemically or biologically. Physical and chemical waste treatment, such as adsorption or coagulation–flocculation has been widely applied in chemical industries. However, these treatments have several disadvantages when compared with biological treatment, especially in terms of high operational cost, using less environmentally friendly chemicals and inefficient in low concentrations of liquid waste.

Heavy metal liquid waste is biologically treated using biosorption methods and involves microorganisms, such as bacteria, fungi, or (micro)algae as biosorbent [2]. The use of microalgae in biosorption process is fascinating to be further developed. Microalgae are cheap, easy to handle, non-pathogenic and effective biosorbents in removing heavy metals in liquid waste. The effectiveness and efficiency of microalgae in biosorption process can be observed from the biosorption mechanisms that can occur in active uptake (using living microalgae) and passive uptake (using dead microalgae) [3]. In addition, microalgae residues can be reused as raw material for producing biodiesel [4–6].

Several studies related to biosorption process using microalgae have been carried out. Previous studies can provide percentage removal of heavy metal ions more than 70% depending on the operating conditions and the type of microalgae [7–13]. However, most studies have been conducted...
using dry/dead microalgae (biomass) as biosorbents. In this present study, the biosorption process of using living microalgae was investigated. Copper (II) (Cu$^{2+}$) ions and microalgae *Chlorella sp.* were chosen as adsorbed metal ions and biosorbent, respectively.

2. Materials and methods

In this present study, biosorption process was conducted using copper (II) sulfate pentahydrate (CuSO$_4$.5H$_2$O) as mother liquor (source of Cu$^{2+}$ ions) and microalgae *Chlorella sp.* from Ugo Plankton, Central Java as biosorbent. The initial concentration of Cu$^{2+}$ ions was varied at 20, 40, 60 and 80 ppm whereas pH scale was varied at 2, 3, 4 and 5.

900 ml Cu$^{2+}$ ions solution at a certain concentration was prepared and poured into the erlenmeyer flask, then 100 ml microalgae *Chlorella sp.* with cell density of 3,800 cells/ml were also put into that flask. pH scale was adjusted using 1 M hydrochloric acid (HCl) solution until a certain pH scale was reached. When pH scale was reached, biosorption process was started. During the biosorption process, the flask was closed and aerated using air pump. Instead of the source of oxygen, the aeration also acted as agitator in the biosorption system. LED lights with a light intensity of 9,000 lux were used as light sources.

Samples were taken at 0, 1, 2, 3, 4, 5, 6, 24 and 48 hours, then were analyzed the cell density using haemocytometer and the content of remaining Cu$^{2+}$ ions using UV–vis spectrophotometer at 615 nm. During the analysis, the ammonia (NH$_3$) solution was added as complexing agent so that dark blue Cu(NH$_3$)$_4$$^{2+}$ ligand complex compounds were formed. That measured concentration data were calculated as a percentage removal of Cu$^{2+}$ ions using the following equation:

$$\text{Percentage removal of Cu}^{2+}\text{ions} = \frac{C_o - C_t}{C_o} \times 100\%$$

with $C_o$ is an initial concentration of Cu$^{2+}$ ions; $C_t$ is concentration of Cu$^{2+}$ ions at t hours.

3. Results and discussion

3.1. The effect of pH scale in the biosorption process

The pH scale is one of important factor in biosorption process using microalgae because this parameter also affects the metabolism of microalgae growth. This biosorption process was conducted by varying pH scale at 2–5 while initial concentration of Cu$^{2+}$ ions was 40 ppm. The maximum pH scale at 5 was chosen because in basic conditions (above 5), copper hydroxide [Cu(OH)$_2$] will be formed and this compound will be insoluble in water. The formation of these compounds was avoided because the reduction of Cu$^{2+}$ ions was not only caused by biosorption process but also because of precipitation. The effect of pH scale on this biosorption process is shown in Figure 1.

Based on Figure 1, the higher the pH value used, the better the biosorption process that occurs. It can be characterized by the figure that a higher percentage removal can be reached at higher pH scale at each sampling time. The best percentage removal of Cu$^{2+}$ ions was 96.10% and was achieved at 48 hours and pH 5.

*Chlorella sp.* can grow optimally at a pH of around 4.9 – 7.3. Although it does not grow optimally, *Chlorella sp.* still grows at pH 2.1 – 4.9 because it has thick cell walls so it can defend itself in acidic pH conditions. In this study, during biosorption process, cell density was also counted to prove that statement, and the experimental data is presented in Figure 2.

Figure 2 showed that cell density increased throughout the sampling time and high pH scale. The more acidic condition caused toxicity level of Cu$^{2+}$ ions to increase and disrupted the metabolism of *Chlorella sp.*. The better growth of *Chlorella sp.* is directly related to the biosorption process. If there are more microalgae in the system, Cu$^{2+}$ ions will be adsorbed well, both actively and passively.
3.2. The effect of initial concentration Cu$^{2+}$ ions in the biosorption process
In this study, the initial concentration Cu$^{2+}$ ions were varied at 20–80 ppm while pH scale was kept constant at 5. The experimental results can be observed in Figure 3. Figure 3 showed that at a concentration of 40 ppm, the biosorption process run at optimum condition while the performance of biosorption process decreased at concentration of 20 ppm. It occurs because the growth of microalgae was not running optimally. However, the performance of the biosorption process also decreased dramatically at concentration 60 and 80 ppm. In that condition, high Cu$^{2+}$ ions would be “toxic” to microalgae, and the growth of Chlorella sp. could not tolerate that condition. It could be proven from the growth of microalgae during the biosorption process and be presented in Figure 4.
Figure 3. The effect of initial concentration Cu$^{2+}$ ions in the biosorption process

Figure 4. Growth of microalgae *Chlorella sp.* at the various initial concentration of Cu$^{2+}$ ions during biosorption process.

Figure 4 showed that the density cell microalgae decreased at an initial concentration of 60 ppm (after 4 hours) and 80 ppm (after 3 hours). This phenomenon would cause the active uptake mechanism would not occur because microalgae had perished. However, the biosorption could still occur passively but this mechanism ineffectively. It could be seen in Figure 3 were at initial concentration of 60 and 80 ppm, the percentage removal of Cu$^{2+}$ ions changed insignificantly after 6 hours of the biosorption process.

4. Conclusion
The biosorption process of Cu$^{2+}$ ions using microalgae *Chlorella sp.* is influenced by initial concentration Cu$^{2+}$ ions and pH solution. Both parameters are also related to the growth performance of microalgae and affect the biosorption process directly. In this study, the best percentage removal of
Cu$^{2+}$ ions was 96.10% and was achieved at initial concentration of 40 ppm and pH 5 within 48 hours of the biosorption process. At higher initial concentration, microalgae growth was inhibited because high Cu$^{2+}$ ions were toxic for microalgae so that the microalgae will perish and the biosorption process was not optimal.

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References
[1] Nurhasni, Salimin Z and Nurifitriyani I 2013 Pengolahan Limbah Industri Elektroplating dengan Proses Koagulasi Flokulasi Valensi 3(1) pp 41–47.
[2] Wang J and Chen C 2009 Biosorbents for heavy metals removal and their future Biotechnology Advances 27 pp 195–226.
[3] Suhendrayatnna 2001 Heavy Metal Bioremoval by Microorganisms: A Literature Study Seminar on–Air Bioteknologi untuk Indonesia Abad 21, Sinergy Forum–PPI Tokyo Institute of Technology 1–14 February 2001.
[4] Chisti Y 2007 Biodiesel from microalgae Biotechnology Advances 25 pp 294–306.
[5] Mata T M, Martins A A and Caetano N S 2010 Microalgae for biodiesel production and other applications: A review Renewable and Sustainable Energy Reviews 14 pp 217–232.
[6] Huang G, Chen F, Wei D, Zhang X and Chen G 2010 Biodiesel production by microalgal biotechnology Applied Energy 87 pp 38–46.
[7] Aung W L, Hlaing N N and Aye K N 2013 Biosorption of Lead (Pb$^{2+}$) by using Chlorella vulgaris International Journal of Chemical, Environmental & Biological Sciences 1(2) pp 408–412.
[8] Purnamawati F S, Soeprobowati T R, Izzati M 2015 Potensi Chlorella vulgaris Beijerinck dalam Remediasi Logam Berat Cd dan Pb Skala Laboratorium BIOMA 16(2) pp 102–113.
[9] Pinem O R B, Sani T F, and Juliastuti S R 2013 Pemisahan Logam Berat Cu dan Cd dari Larutan Logam Sintetis dan Air Limbah Industri dengan Menggunakan Biomassa Chlorella vulgaris dan Biomassa Chlorella vulgaris yang Terimobilisasi sebagai Adsorben Jurnal Teknik Pomits 2(1) pp 1–5.
[10] Kumar Y P, King P and Prasad V S R K 2006 Removal of copper from aqueous solution using Ulva fasciata sp. – A marine green algae Journal of Hazardous Materials B137 pp 367–373.
[11] Chong A M Y, Wong Y S, Tam N F Y 2000 Performance of different microalgal species in removing nickel and zinc from industrial wastewater Chemosphere 41 pp 251–257.
[12] Kalyani S, Rao P S and Krishnaiah A 2004 Removal of nickel (II) from aqueous solutions using marine macroalgae as the sorbing biomass Chemosphere 57 pp 1225–1229.
[13] Gong R, Ding Y, Liu H, Chen Q, Liu Z 2005 Lead biosorption and desorption by intact and pretreated Spirulina maxima biomass Chemosphere 58 pp 125–130.