RINGKASAN

Bioleaching Logam Berat Timbal Dari Sedimen Tercemar Oleh *Pseudomonas fluorescens, Thiobacillus ferrooxidans, Escherichia coli* dan *Bacillus sp*

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Bioleaching merupakan suatu proses pelarutan/pelepasan logam atau pengambilan (ekstraksi) logam dari sedimen atau mineral sukar larut menjadi bentuk yang larut dengan menggunakan bakteri. Bakteri yang digunakan adalah bakteri *Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans* dan *Bacillus sp* sebagai bakteri *leaching* yang mampu melarutkan senyawa timbal sulfida sukar larut menjadi senyawa timbal sulfat yang dapat larut melalui proses biokimia.

Proses *Bioleaching* merupakan teknologi alternatif yang dapat dikembangkan sebagai salah satu teknologi untuk memperoleh (*recovery*) logam di masa mendatang. Salah satu penerapan proses ini adalah untuk melepaskan dan mengekstraksi logam berat yang ada dalam sedimen, sehingga sedimen tersebut bebas logam berat dan aman terhadap lingkungan. Disamping itu proses *bioleaching* (*bacterial leaching*) dapat menimbulkan dampak negatif terhadap lingkungan. Selama proses *bioleaching* berlangsung dihasilkan asam sulfat, ion H^+, dan senyawa khelat yang dapat menyebabkan logam berat seperti Pb dalam sedimen akan melarut sehingga ion-ion logam ini menimbulkan efek toksisitas terhadap lingkungan. Oleh sebab itu untuk mecegah pencemaran badan air lebih lanjut maka perlu mengendalikan faktor yang dapat menghambat

proses *bioleaching* dan atau menambahkan basa (kapur) agar ion logam berat yang dihasilkan (dileaching) mengendap kembali sehingga air yang menuju ke saluran pembuangan badan air aman terhadap cemaran logam berat tersebut.

Penelitian ini bertujuan untuk melihat pengaruh bakteri *Escherichia coli, Pseudomonas fluorescens, Thiobacillus ferrooxidans* dan *Bacillus sp* tehadap peningkatan kadar Pb pada *bioleaching* sedimen limbah. Variabel terikat dalam penelitian ini adalah kadar Pb terlarut dan pH medium setelah mengalami perlakuan tertentu. Variabel bebas terdiri jenis bakteri, waktu inkubasi (5 hari, 10 hari, 15 hari) dan jenis limbah (limbah baterei dan elektroplating). Proses *bioleaching* dilakukan dengan memasukkan sedimen yang mengandung logam Pb ke dalam wadah (botol) dan diinokulasi dengan 10% (v/v) bakteri *Escherichia coli, Pseudomonas fluorescens, Thiobacillus ferrooxidans* dan *Bacillus sp.* Pengambilan sampel dilakukan setiap 5 hari sekali selama 15 hari, sampel disentrifugasi dan supernatan digunakan untuk mengukur pH dan menentukan kadar Pb yang terlarut dengan menggunakan *Flame Atomic Absorption Spectroscopy* (FAAS).

Penelitian ini merupakan penelitian eksperimental laboratorik dengan Rancangan Acak Lengkap pola faktorial. Untuk menguji pengaruh jenis bakteri (*P. fluorescens, E. coli, T. ferrooxidans,* dan *Bacillus sp*) dan waktu inkubasi (5 hari, 10 hari, 15 hari) terhadap kadar Pb pada *bioleaching* limbah (baterei dan elektroplating) digunakan analisis varians satu jalur (One-Way ANOVA). Untuk menguji pengaruh jenis limbah

terhadap kadar Pb pada proses *bioleaching* digunakan uji t test. Sementara untuk menguji interaksi jenis bakteri dan waktu inkubasi terhadap kadar Pb dan pH pada *bioleaching* limbah digunakan analisis varians dua jalur (Two-Way ANOVA). Uji lanjut analisis varians adalah uji beda nyata terkecil (LSD). Taraf kemaknaan uji statistik yang digunakan adalah 5% ($\alpha = 0,05$).

Hasil penelitian menunjukkan bahwa bakteri *P. fluoresncens, E. coli, T. ferrooxidans* dan *Bacillus sp* berpengaruh terhadap kadar Pb pada *bioleaching* limbah baterei dan elektroplating. Demikian pula waktu inkubasi berpengaruh terhadap kadar Pb pada *bioleaching* limbah baterei dan elektroplating. Jenis limbah baterei dan elektroplating berpengaruh pada proses *bioleaching* oleh bakteri. Interaksi bakteri dan waktu inkubasi berpengaruh terhadap kadar Pb pada *bioleaching* limbah baterei dan elektroplating. Jenis bakteri berpengaruh terhadap pH, sedangkan waktu inkubasi dan Interaksi jenis bakteri dan waktu inkubasi tidak berpengaruh terhadap pH *bioleaching.*

Berdasarkan hasil penelitian membuktikan bahwa bakteri *P. fluorescens, E. coli, T. ferrooxidans,* dan *Bacillus sp* mempunyai kemampuan berbeda dalam melakukan proses *bioleaching* senyawa Pb yang terdapat dalam sedimen limbah, sehingga kadar Pb dalam sedimen limbah industri menurun atau Pb terlarut dalam medium meningkat. Kadar Pb terlarut tertinggi (0,667 ppm dan 0,512 ppm) dicapai pada perlakuan bakteri *Bacillus sp.* Pada waktu inkubasi 15 hari kadar Pb terlarut diperoleh 0,567 ppm (limbah baterei) dan 0,404 ppm (limbah

elektroplating). Peningkatan waktu inkubasi dapat berpengaruh pada pertumbuhan bakteri, semakin lama bakteri teradaptasi dengan kondisi yang ada maka jumlah sel bakteri yang dihasilkan makin bertambah. Meningkatnya jumlah sel bakteri seiring dengan lamanya waktu bakteri melakukan aktivitas metabolismenya, mengakibatkan jumlah Pb yang di *leaching* makin meningkat pula. Dapat dijelaskan bahwa peningkatan waktu inkubasi akan menyebabkan kontak bakteri dengan permukaan partikel sampel yang di *leaching* berjalan lama sehingga makin banyak waktu yang dibutuhkan bakteri untuk melakukan aktivitas *leaching*.

Berdasarkan temuan disertasi ini, maka dalam upaya mencegah toksisitas terhadap lingkungan karena melarutnya Pb dalam sedimen limbah industri perlu dihindari kehadiran atau keberadaan bakteri khususnya bakteri yang dapat melakukan aktivitas *leaching* terhadap limbah.

SUMMARY

Bioleaching of Heavy Metal from Contaminated Sediment by *Pseudomonas fluorescens, Thiobacillus ferrooxidans, Escherichia coli, and Bacillus sp*

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Bioleaching is a process of metal dissolving or releasing or extracting from sediment or insoluble mineral to soluble form by using bacteria. The bacteria used here are *Pseudomonas fluorescens, Thiobacillus ferrooxidans, Escherichia coli,* and *Bacillus sp* as the leaching bacteria being able to dissolve insoluble lead sulfide to lead sulfate that can dissolve through biochemical process.

The bioleaching process is an alternative technology that can be developed as one of technologies for recovery of heavy metals in the future. One of its applications is to release and extract heavy metals present in the sediment so that this sediment is free of heavy metals and safe for the environment. In addition, bioleaching process (bacterial leaching) can induce negative impacts for the environment. The bioleaching process produces sulfate acids, H^+ ions and chelate compounds that can generate heavy metal such as Pb in the sediment that may dissolve so that its ions result in toxicity in the environment. Therefore, to prevent further pollution in body of water, there should be the control of factor that can inhibit bioleaching process or the base (lime) addition making ions of the heavy metal precipitate back so that water flowing into the drain will be safe from pollutant of the heavy metal.

Objective of this research is to see the effect of *Escherichia coli, Pseudomonas flurorescens, Thiobacillus ferrooxidans* and *Bacillus sp* microbes on the waste bioleaching. The dependent variables are including the dissolved Pb levels and pH of medium after given treatment. The independent variables are consisting of incubation time (5, 10 and 15 days) and type of wastes (battery and electroplating wastes). The bioleaching process is done by putting the Pb-consisting sediment into a bottle and inoculated with 10% (v.v) *Eschericia coli*, *Pseudomonas fluorescencs, Thiobacillus ferrooxidance, Bacillus sp*. The sample is taken each 5 day for 15 days. The sample is centrifuged and supernatant is used to measure pH and determine the dissolved Pb levels by making the use of *Flame Atomic Absorption Spectroscopy* (AAS).

This research is a laboratory based experimental research with factorial pattern of complete random design. To examine the effect of types of bacteria (*Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans, Bacillus sp*) and incubation time (5, 10, 15 days), the one-way ANOVA analysis is used. To examine the effect of waste on the Pb levels in the bioleaching process, the t test is employed. Furthermore, to know an effect of the bacteria and incubation time upon the Pb levels and waste bioleaching, the two-way ANOVA is used. Other variance analysis used is Least Significant Difference (LSD). Significance level of statistical test is 5% ($\alpha = 0.05$).

The results showed that the *Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans* and *Bacillus sp* possessed effects on Pb

levels in the bioleaching of battery and electroplating wastes. Moreover, incubation time also exerted impact on the Pb levels in the bioleaching. Types of the waste had impact on the bioleaching process by bacteria. Interaction of bacteria and incubation time influenced Pb levels in the bioleaching. Type of bacteria influenced pH, while incubation time and interaction of bacteria and incubation time had no influence on the bioleaching pH.

These results prove that *Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans* and *Bacillus sp* have different abilities in bioleaching of Pb compounds existing within the waste sediment, so that Pb levels in the industrial waste sediment will decrease or the dissolved Pb in the medium will increase. The highest dissolved Pb levels (0.667 ppm and 0.512 ppm) are achieved in *Bacillus sp*. Treatment. In incubation time of 15 days, the dissolved Pb levels are 0.567 ppm (battery waste) and 0.404 ppm (electroplating waste). The increased incubation time can influence bacteria growth. When bacteria are adapted to the available condition in longer time, then number of bacteria cells will be greater. The increased bacteria cells along with duration of bacteria's metabolism activities will also induce increase in Pb levels. Thus, it can be stated that expansion of incubation time will make the contact of bacteria with particle surfaces of the sample to be leached takes place more longer. Thus, time needed by bacteria to undertake leaching activity also increases.

Based on these findings, to prevent toxicity in environment because of the dissolved Pb in the industrial waste sediment, then presence or

ABSTRACT

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Bioleaching is a process of metal dissolving or releasing or extracting from sediment or insoluble mineral to a soluble form by using bacteria. The bacteria used here are *Pseudomonas fluorescens, Thiobacillus ferrooxidans, Escherichia coli,* and *Bacillus sp* as the leaching bacteria being able to dissolve the insoluble lead sulfide to the lead sulfate that can dissolve through biochemical processes.

Objective of this research is to see the effect of types of bacteria (*Escherichia coli, Pseudomonas flurorescens, Thiobacillus ferrooxidans* and *Bacillus.sp*), incubation time (5, 10, 15 days) and types of waste (battery and electroplating) on the Pb levels in the bioleaching process. The variables observed are the dissolved Pb and pH of medium. The dissolved Pb is analysed by using the *Flame Atomic Absorption Spectroscopy* (FAAS).

The results showed that the *Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans* and *Bacillus sp* possessed effects on Pb levels in the bioleaching of battery and electroplating wastes. Moreover, incubation time also exerted impact on the Pb levels in the bioleaching. Interaction of bacteria and incubation time influenced Pb levels in the bioleaching. Type of bacteria influenced pH, while incubation time and interaction of bacteria and incubation time had no any influence on the bioleaching pH.

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Key Words: Bioleaching, lead, incubation time, *Pseudomonas fluorescens, Escherichia coli, Thiobacillus ferrooxidans, Bacillus sp.*