Water Lettuce (Pistia stratiotes, Linn) Potency as One of Eco-friendly Phytoextraction Absorbers of Zinc Heavy Metal to Solve Industrial Waste Problem in Indonesia

Pebri Nurhayati¹⁺, Sapta Abimanyu², Siti Kaswati³ and Iqbal Raditya Fajr ⁴

¹Student at Faculty of Social Sciences, Yogyakarta State University

²Student at Faculty of Languages and Arts, Yogyakarta State University

³Student at Faculty of Mathematics and Natural Sciences, Yogyakarta State University

⁴ Student at Faculty of Social Sciences, Yogyakarta State University

Abstract. Generally, environmental condition in Indonesia is very critical due to the rapid development of industry. This fact is diminished by many industrial wastes which are in the form of heavy metals that are dangerous for human and environment. Recently, there are a lot of researches about environmental problem solution; the mostprominent is development of phytoextraction as bioremediation by utilizing plant. This method uses regeneration (replacement) of plants to obtain an optimal absorption to decrease Zinc (Zn) heavy metal concentration. In this study, we use water lettuce (*Pistia stratiotes*, Linn) as bioremediation to absorb Zinc heavy metaling rowing media solution. In regeneration method, Zinc metal concentration was left at 6.6965%, while in non-regeneration method it was left at 10.0147% in growing media solution of total initial amount 20 ppm. Test of released Zinc metal of water lettuce into distilled water shows that released Zinc metal percentage in distilled water was 0.0059 ppm. Quantitative analysis of Zinc metal which was left in the mediawas measured by Atomic Absorption Spectrophotometer (AAS). In the future, we hope water lettuce an be one of eco-friendly phytoextraction absorbers of Zinc heavy metal to solve industrial waste problem in Indonesia.

Keyword: Zinc (Zn) heavy metal absorber, Phytoextraction, Water lettuce, Bioremediation, Plant regeneration

1. Introduction

Generally, environmental condition in Indonesia is very critical due to the rapid development of industry. This fact is diminished by many industrial wastes which are in the form of heavy metals that are dangerous for human and environment. The presence of heavy metals in the environment comes from two sources. First, heavy metals come naturally from the environment. Heavy metals level from environment in biosphere is relatively small so heavy metals from that source are not dangerous for the environment. Second, heavy metals come from anthropogenic activity. The presence of heavy metals in this case is caused by human activity, such as metal plating industry waste; paints mining; motor vehicle exhaust gas; and also used goods such as batteries, cans, etc. Heavy metals which come from anthropogenic activity are dangerous for the environment. The presence of heavy metals from anthropogenic activity in the environment will causechronic effects of bio-accumulation. Lubis (2002) said that for human, the bio-accumulation is necessary tobe noticed because human is a creature with the highest trophic level in the food chain. Heavy metals pollution can be in the form of Mercury (Hg), Cadmium (Cd), Copper (Cu), Chromium (Cr), Lead (Pb), Zinc (Zn), and so on. Based on Environmental Impact Management Agency of Indonesia (Bapedal) decision letter, standard of total Zinc in industrial waste water that is allowed is 0.921 ppm. Waste water management gets an

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⁺ Corresponding author. Tel.: +6285643872986. *E-mail address*: pebrinurhayati@ymail.com.

intensive care from all parts of the society. The right management of waste water without side effect is expected to reduce pollutants, especially heavy metals that potentially damage the environment.

Palar (1994) said that environment has a system of defense to prevent heavy metals from reaching the sea; itis mangrove ecosystem which generally dominates the beach area. One of thisecosystem functions is to absorb or bind heavy metal.Raskin (1996) said that basically all types of aquatic plant can absorb heavy metals by their root tissue and can be used as a biological filter or just used as an indicator of pollution. There are some methods to manage the waste water. They are physical, chemical, biological, and microbiological method. From this natural phenomenon of mangrove ecosystem, biological method through bio-filtration process by utilizing plant to manage waste water is mostly used. Ulfin (2001) said that bio-filtration process has some advantages. Those advantages are; it is effective, it has low cost bio-filtration pool making, the plant used in this process is fast-growing and easy to maintain, it does not need an operator who has a special skill, and it's eco-friendly. In this study, we use two methods. They are non-regeneration and regeneration. In nonregeneration method, water lettuces were directly planted in four days and after that they were separated from the solution, while in regeneration method water lettuces were planted and replaced everyday or at least once every two days and we measured amount of absorbed and left Zinc concentration. We did the water lettuce replacement (regeneration)so that the plants grown in growing media could be always fresh and could absorb the heavy metal well, so an optimal absorption could be done in a short time. In this study, we used 100 grams of water lettuce in two liters solution for both methods. In non-regeneration method we directly used those 100 grams of water lettuce, while in regeneration method we used 25 grams of water lettuce a day in four consecutive days. We will compare the absorption capability of water lettucebynon-regeneration method to the regeneration method.

2. Experiment Methodology

2.1. Equipment and Materials Used

The equipment used in this study were an analytical scale, measuring cups, beaker glass, a measuring flask, a pH meter, an oven, a funnel, a watch glass, plastic containers, hot plate, a vial and an Atomic Absorption Spectrophotometer (AAS).

The materials used in this study were distilled water, concentrated HNO₃ solution(65%), Zinc solids, concentrated NH₃ solution (25%), Whatman 42 papers and water lettuces.

2.2. Study Procedures

2.2.1 Preparation for Water Lettuce (*Pistia stratiotes*, Linn)

Water lettuces used in this study came from Way Jepara, East Lampung, Indonesia. We selected plants in the same size, and then they were washed. After that, they were acclimatized for 10 days with distilled water and used as culture stocks which are ready to be used for further experiments.

2.2.2. Analysis of Zinc Concentration by not Replacing Plants (Non-Regeneration Method)

Water lettuces taken from the culture stocks were weighed 100 grams and planted in two liters solution containing 20 ppm Zinc concentration. Water lettuces were left for four days. After that, we separated the plants from the solution. We got filtrate and the plants. Next, the water lettuces were processed again to find out released Zinc amount from water lettuces into the distilled water (procedure 2.4). Then we analyzed left Zinc concentration from the filtrate by SSA.

2.2.3. Analysis of Zinc Concentration by Replacing Plants (Regeneration Method)

Water lettuces taken from the culture stocks were weighed 25 grams and planted in two liters solution containing 20 ppm Zinc concentration (media day-1). The plants were left for a day, after that we separated the plants from the solution. We got filtrate and the plants day-1. We measured day-1 filtrate volume, and then we analyzed the left Zinc amount by SSA. The rest of the day-1 filtrate was used as a new growing media of the water lettuce (media day-2) with a fixed amount of new water lettuce 25 grams. The plants were left for a day, after that we separated the plants from the solution. We got filtrate and the plants day-2. Filtrate day-2 got the same treatment as the filtrate day-1. The analysis was continued until day-4.

2.2.4. Analysis of Released Zinc from Water Lettuce into Distilled Water

Water lettuces which were gotten from procedure 2.2were planted in two liters distilled water for fourdays. After that, we separated the plants from the solution. We got filtrate and the plants. Then we analyzed the released Zinc concentration from the filtrate by SSA.

3. Results and Discussion

3.1. Preparation of Plant

In this study we used water lettuces as a bio-filter. The plants came from Way Jepara, East Lampung, Indonesia. The advantages of this plant are it has alot ofrootsand it can be easilybred. Preparation was done to make the plants have an optimal condition in bio-filtration process. The plants were acclimatized for 10 days in order to make the plant can adjust to then ewen vironment and to reduce metals and impurities in the root tissue so that the roots of the plant can absorbinan optimal condition. We selected plants in the same size, especially the roots, so that the replication which is performed will have the absorption which is not much different. Largestabsorption was found in the roots and it was just a little in the leaves.

3.2. Analysis Result of Non-Regeneration Method

In non-regeneration method, water lettuces were weighed 100 grams and they were directly planted for four days. In non-regeneration method the Zinc concentration was left 2.0029 ppm after four days(Table 1.)

Left Zinc Absorbed Zinc % ofLeft Initial Left Initial Zinc concentration concentration Zinc Day of Volume Volume concentration amount amount growing (ml) (ml) (ppm) media (ppm) (ppm) 2000 1717 20 2.0029 17.9971 10.0147

Table 1: Zinc Concentration Data by Non-Regeneration Method

Plant weight 100 grams; total planting time 4 days

3.3. Analysis Result of Regeneration Method

Regenerationmethodin this studyis a methodby replacing theplantseverydayin the same growing media(solution). In thismethod, we usedwater lettuces weighed 25grams, twoliterssolutioncontaining 20 ppm Zinc concentration, and four days planting time. Growing mediawasanalyzedeverydaytofind outthe left Zinc amount. The results are presented in Table 2.andImage 1.below

Day of	Initial volume (ml)	Left volume (ml)	Initial Zinc concentration (ppm)	Left Zinc concentration amount/day (ppm)	% ofLeft Zinc in growing media	Absorbed Zinc concentration amount/ day(ppm)	% of Absorbed Zinc in the plants/day
1	2000	1920	20.0000	12.0580	60.2900	7.9420	39.7100
2	1820	1708	12.0580	6.6701	55.3168	5.3879	44.6832
3	1608	1526	6.6701	3.7688	56.5029	2.9013	43.4971
4	1426	1345	3.7688	1.3393	35.5365	2.4295	64.4635

Table 2: Zinc Concentration Data by Regeneration Method

Plant weight 25 grams a day; total planting time four days

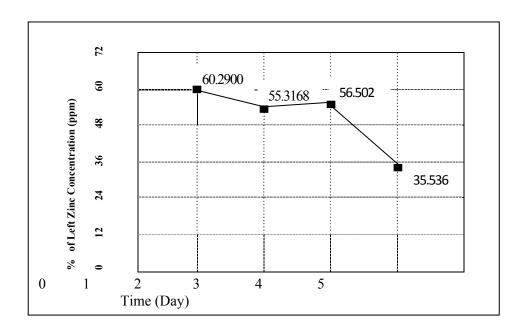


Fig. 1: Left Zinc Concentration Percentage by Regeneration Method

Table 1. and Fig. 1. shows percentage of left Zinc concentration from day-1 until day-4. The percentage of left Zinc concentration is decreasing significantly. Until the day- 4, Zinc concentration couldbe lowered to 6.6965% of the concentration of 20 ppm or to 1.3393 ppm. Daily plants replacement is efficient to do because the concentration could be significantly decreased. By replacing the plants everyday, the plants could absorb in an optimal condition. Water lettuces regeneration and acclimatization made the root tissue did not have stiff Zinc concentration, saturation disappeared and damaged cells were repaired. On the day-4, the amount of absorbed Zinc concentration wasthe smallest because the media which was used in day-4 was alsothe smallestcompared to previous days (3.7688 ppm). But, the percentage of absorbed Zinc concentration was the biggest (64.4635%). From table 1, we can see that the biggest amount of absorbed Zinc concentration was in the day-1, where the water lettuces could absorb 7.9420 ppm or 39.71% of the initial concentration 20 ppm. This is because the initial amount used was the biggest amount, so the Zinc absorbed amount day-1 was the biggest compare to Zinc absorbed amount day-2 until day-4. This absorption process is appropriate with diffusion law. Diffusion law is the movement of Zinc metal ion from a stiff concentration into a more dilute concentration in the plant cell membrane. Plants could accumulate that metal by converting it into a liquid phase into the cell metabolism with different stages. Decrease of the absorbed Zinc concentration which was absorbed by water lettuce on day-2 to day-4 happened because we used media with the left Zinc concentration from the previous days. It also means the Zinc metal ion in the media that goes into the root cell membrane will have a small diffusion.

3.4. Analysis Result of Comparison between Non-Regeneration Method and Regeneration Method

In this study we compare non-regeneration and regeneration method in order to prove the hypothesis that regeneration method is more effective than non-regeneration method. We can compare both methods because they have the same concentration amount, planting time, and plant weight. They just have a different treatment or method used. In non-regeneration method, 100 grams of plant were directly planted for four days; while in regeneration method 25 grams of plant were planted everyday in four consecutivedays. At the end the total planted plants number were the same with non-regeneration method. In non-regeneration method the Zinc concentration was left 2,0029 ppm (Table 2) after four days, while in regeneration method was left 1,3393 ppm (Table 1).

From t test for two different methods, we know that $t_{count} > t_{table}$ in 95% confidence interval of the difference, so Ho is rejected. It means regeneration method is better than non-regeneration method in the case of Zinc concentration decrease. By using regeneration method, left Zinc amount can be smaller than

non-regeneration method. This different result is caused by the accumulation of heavy metals into the root tissue. A factor that influences plant absorption power is the plant freshness. In regeneration method, the plants were different plants everyday, so they were still fresh and they could still absorb optimally, while in non-regeneration method the plants were the same until the day-4, so they had been aturated in the root tissue. Other factors that also influence are the nature of the plants, such as growth speed, size and depth of roots, evaporation rate, and the need to metabolize food. In regeneration method, plants could do better nature growth because the small number of plants (25 grams a day) did not make the media becomes full, so it could do optimal evaporation. Sunlight also affects the plant metabolism to absorb optimally.

3.5. Analysis Result of Released Zinc Concentration of Water Lettuce into Distilled Water

Water lettuces used in this analysis was the water lettuces which were used in non-regeneration method. We analyzed them to find out the result of bio-filtration process after they absorbed Zinc concentration in four days. Analysis result shows that the Zinc concentration amount which was absorbed by the water lettuces after four days is 17.9971 ppm. The early hypothesis is released Zinc concentration amount into the distilled water will be equal to absorbed Zinc metal concentration from growing media. Analysis result shows that the Zinc concentration which was released is only 0.0059 ppm. Amount of released concentration is so small compared to the amount of absorbed concentration. It could happen because the absorbed Zinc metal which was converted by plants into a liquid phase in the root tissues had a metal hyper accumulation, so part (little or much) would be transferred to the leaves. Physically the plants leaves looked yellowing and they were finally separated from their branches. Stiff Zinc concentration in the plants had damaged leaves cells and made some leaves separated from the branches so part of Zinc concentration could not be released into the distilled water when the releasing time. The presence of released Zinc concentration shows that there was a physical absorption. Released Zinc concentration was not as big as absorbed Zinc concentration because there were chemical and physical absorption. Metal ion which was bind by fitokelatin made complex ion of metal brace, so metal ion could not easily be released. Root tissue releasedZinc concentration in large amount because Zinc concentration in the root tissue membrane was stiffer than in the distilled water as the media. Hyper accumulated metal ion could not make metal brace perfectly because the plants had been saturated. Another factor that caused the amount of released Zinc was small is the releasing time was too short. It should be at least the same with acclimatizing time that was 10 days. This metal would be saved in vacuole. Water lettuce as an easy bred plant will do cell regeneration so hyper accumulated cells will die. Application that must be done to the environment is the plants that have been used as bio-filtration do not have to be destroyed. We just have to place them to a neutral growing media.

3.6. Analysis of Comparison between Non-Regeneration Method and Regeneration Method

Analysis of comparison was done by comparing plant absorption power during the planting time. We will know which method is more effective to get an optimal result by analyzing the amount of absorption Zinc concentration.

From analysis result of Table 1. and Table 2., we know that plant absorption power in regeneration method is bigger than in non-regeneration method. By total initial amount 20 ppm in both methods, in regeneration method Zinc metal concentration amount was absorbed 18,6607 or 93,3035%, while in non-regeneration method Zinc metal concentration amount was absorbed 17,9971 ppm or 89,9855%. From analysis of t test in both methods in 95% confidence interval of the difference with degrees of freedom 4, we got t_{count} is bigger than t_{table} . From that analysis, we can assume that Ho is rejected; it means that in regeneration method water lettuce is better and Zinc metal absorption rate is higher than in non-regeneration method.

4. Conclusions and Recommendations

4.1. Conclusion

The result of study analysis that has been done gives several conclusions:

 By using water lettuce regeneration method, the Zinc heavy metal concentration can be lowered to 1.3393 ppm from the initial concentration amount 20 ppm or can be absorbed 18.6607 ppm. Percentage of left Zinc from day-1 (60.2900%) is significantly different with percentage of left Zinc day-4 (6.6965%). It shows that water lettuce absorption to Zinc heavy metals is effective and has a big influence to the environment if we apply it.

- Water lettucecan be used as an eco-friendly phytoextraction absorber of Zinc heavy metal.
- Regeneration methodis abetter methodin the absorption of Zinc heavymetal than others because its absorption power is higher than others.
- If we use water lettuce as Zinc heavy metal absorber, we do not need much cost.

4.2. Suggestion

The industrial factories should utilize water lettuce to solve Zinc heavy metal waste problem, because from theresults of this study;if we use water lettuce as Zinc heavy metal absorber, we do not need much cost and it can be used as an eco-friendly phytoextraction absorber of Zinc heavy metal to solve industrial waste problem in Indonesia. To know further about water lettuce benefits to solve environmental problem, we must do more experiments.

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