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Phytoremediation of Zinc, Copper, and Lead Using Ipomoea Aquatica in Water Contaminants

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ABSTRACT

Lake Tempe in the Wajo Regency, South Sulawesi (Indonesia) is highly toxic due to metal pollution from industrial activities and the activities of residents living around the region. Zinc-contaminated water poses a potential threat to biotic communities. This research aims to develop phytoremediation technology to effectively remove toxic zinc from contaminated lake Tempe. The use of plants as phytoremediation agents to accumulate metals in polluted water is considered adequate because the method is environmentally friendly and presents economic value. This study was therefore designed to assess the phytoremediation potential of water spinach against zinc (Zn), copper (Cu), and lead (Pb). Water spinach was planted in Tempe lake contaminated with zinc (Zn), copper (Cu), and lead (Pb) metals, and the study was conducted for 30 days under natural conditions. Subsequently, the Tempe lake physicochemical properties, including pH, TDS, TSS, total nitrogen, total phosphate as P, and Zn content, were measured, before and after the phytoremediation process. The ability of plants to absorb zinc (Zn), copper (Cu), and lead (Pb) were assessed by the bioconcentration factor (BCF). The results showed that there was a correlation between the BCF value and the phytoremediation time. The longer the phytoremediation time, the higher the BCF value are obtained. Infra-Red (IR) data shows the presence of metal binding in plants with the functional groups C=S, C=N, and OH. Water spinach has the potential as a phytoremediation agent in remediating zinc (Zn), copper (Cu), and lead (Pb) metals in polluted lake Tempe water.

1. INTRODUCTION

Lake Tempe covers an area of 14,406 hectares, located in three districts, namely Wajo (8.510 ha), Soppeng (3.000 ha), and Sidrap (2.896 ha). An increase in the number of industries and residential areas is usually followed by an increase in the amount of solid, liquid, and gaseous waste [1]. One of the wastes that should be observed is metals because they are widely used as raw materials and auxiliary media in various types of industries. Dumping this waste into the water can reduce quality and cause environmental pollution. In addition to changing the quality of water, the metals deposited with sediments can also cause the transfer of toxic chemicals from the sediments to organisms [2]. Zn metal is a type that has the potential to contaminate the temple lake.

Potential anthropogenic sources of metallic zinc pollution in lake Tempe can come from the disposal of PLTG waste (Gas Power Plant) and the activities of fishermen. In this area, waste is discharged directly or indirectly into the lake, and sources of Zn metal can be resident waste (such as battery waste, pipe waste, burning garbage, vehicle exhaust, and household industry activities), agriculture, and livestock, both in the form of solid, liquid, and gas. Furthermore, there are boats as a means of transportation for fishermen, dozens of floating houses whose daily activities such as cooking, bathing, and others are carried. Many people live on the land around the lake and most of the water that goes into the lake is household waste [3, 4].

The increase in metal levels in the water is followed by an increase in the contents of these substances in aquatic organisms such as shellfish, seaweed, and another marine biota. The use of these organisms as food ingredients endanger human health [5, 6]. When Zn penetrates the human body, the metal accumulates in body tissue can no longer be excreted outside the body. According to the US National Library of Medicine's Toxnet database, the oral LD50 for zinc is about 3 g/kg of body weight. This value is over 10 times higher, compared to cadmium and 50 times higher, compared to mercury [7]. At already high concentrations in the human body, this will have serious negative consequences, namely; inhibits enzyme activity to disrupt metabolic processes, causing chromosomal abnormalities (genes), inhibiting fetal development, reducing female fertility, inhibiting spermatogenesis, reducing peripheral nerve conduction, inhibiting hemoglobin formation, causing kidney damage, causing blood deficiency or (anemia), swelling of the head (encephalopathy), and causes emotional and behavioral disorders [8].

Several attempts have been made to remove metals, such as nanofiltration [9-11], ion exchange resins [12], electrocoagulation [13], and adsorption [14]. However, this method is relatively expensive, less effective, and detrimental to health [15]. Another alternative that can be used to repair metal contaminants is certain factories that can ingest and accumulate high levels of metals known as phytoremediation. Furthermore, it is a sanitary method developed in science and technology to purify contaminated soil, water, or air [16], relying on the role of plants to absorb, degrade, transform, and mobilize metal pollutants from the environment. Phytoremediation is also cost-effective and durable for the repair of contaminated sites [17]. The most important factor in the method is the selection of suitable plants, the absorption potential in absorbing high levels of pollutants, and the ability to survive in polluted waters [18].

Water spinach is suitable for phytoremediation, and has a high potential due to the fast growth, high potential for metal accumulation, and high tolerance to toxic substances. Kale easily absorbs nutrients, especially Zn and Se [19, 20], however, the water spinach plant is also used because of its high growth rate and the ability to absorb water rapidly. The roots provide a site for filtration and adsorption of suspended solids, as well as microbial growth. A study by Hapsari et al. [21] described water spinach as a plant with fairly wide adaptability to climatic and soil conditions in the tropics, and therefore planted in various regions. Meanwhile, Suchaida et al. [22] explained water kale plants have tolerance for heavy metal absorption. The optimum temperature for water spinach growth is 25-33℃ because the plant does not grow properly at temperatures below 23.9℃. The plant (Ipomoea Aquatic Forsk) has the potential to become a phytoremediation agent for heavy metals Zn, Cu, as well as Pb, and several studies have reported water spinach's effectiveness in remediating Furthermore types of waste, including chromium [23, 24], cadmium [25], mercury [26], and household waste [27]. In addition to reducing various pollutant contents, water spinach reared in polluted media is also indicated to have adequate growth ability [28]. The functional groups contained in the plant can bind with metal ions, for instance, carbonyl, amino, thiol, hydroxyl, phosphate, and hydroxy-carboxyl, on the cell walls [29, 30]. Therefore, this study aims to determine water spinach plants' ability as a phytoremediation agent for Zn, Cu, and Pb metal in lake Tempe, contaminated with Zn Cu, and Pb metals.

2. MATERIALS AND METHODS

2.1 Plant sampling and water

This study used water spinach which functions as a phytoremediation plant to reduce Zn in Tempe lake contaminated by $ZnCl_2$. The plants were collected from the Tempe lake area, South Sulawesi, Indonesia, while water samples were obtained from the lake. Subsequently, 50 ppm of $ZnCl_2$ pollutants were added to the water samples. The addition of $ZnCl_2$ was caused by the level of Zn in the Tempe lake<0.01 ppm. This value did not have a big effect and was not suitable for use as wastewater. The same treatment was also carried out for Cu and Pb metals with the addition of $CuCl_2$ and $PbCl_2$.

2.2 Experimental model

The plants were planted in a rectangular basin, with length, width, and height of 20 cm, 30 cm, and 20 cm, respectively, and containing lake Tempe water contaminated with $ZnCl_2$, at a level of about 5-10 cm. Furthermore, the acclimatization process was carried out for about a week, to enable the plants to adapt to the new environment. The water spinach was planted for 1 month (September to October 2007), with plant and water samples collected at a 10-day interval. The same treatment was also carried out for Cu and Pb metals with the addition of $CuCl_2$ and $PbCl_2$. The experimental model is shown in Figure 1.

(a) (b)

Figure 1. (a) The plants were planted in a rectangular basin and (b) a Rectangular basin containing lake Tempe contaminated with $ZnCl_2$ at a level of about 5-10 cm

2.3 Metals levels in plants and water with Atomic Absorption Spectroscopy (AAS)

Plant water spinach was grown using growth media contaminated with Zn (II) 50 ppm for 30 days, and the absorption concentration of Zn (II) was measured four to 4 times at 10-day intervals. Every 10th day, Zn (II) levels were tested on water spinach plants, and lake Tempe water used to grow water spinach was analyzed using an Atomic Absorption Spectrophotometer. Subsequently, the samples were analyzed, using the SNI 06-6989.8-2004 method. The determination process starts from manufacturing standard solutions and calibration curves to obtain a standard regression equation, to determine the sample's metal content. The same treatment was also carried out for Cu and Pb.

2.4 Physicochemical analysis of contaminated Tempe lake

Physicochemical analysis of the lake was carried out in an accredited laboratory with two replications to obtain valid data. Tests were carried out before and after phytoremediation which included pH, total nitrogen, total phosphate tests as P, TDS, and TSS. The values at zero (0) days are recorded as initial values, while the values recorded after phytoremediation are indicated with the final grade. Physicochemical analysis of water samples was analyzed using, test methods based on SNI 06-6989.11-2004 for pH, AOAC Official Method 973.48.18th Ed, 2005 for total nitrogen, SNI 06-6989.31-2005 for total phosphate as P, SNI 06-6989.27-2005 for TDS, and SNI 06-6989.3-2004, for TSS.

2.5 Bio-concentration Factor (BCF)

BCF indicates an index of the plant's ability to accumulate metals relating to metal concentrations in the substrate. The BCF was calculated as follows [31]. Furthermore, it is a useful parameter for evaluating the potential for plants to collect metals and the value is calculated based on the dry weight [32, 33].

$$\mathrm{BCF}(L/kg) = \frac{\mathrm{MCP\ (mg/kg)}}{\mathrm{MCW\ (mg/L)}} \tag{1}$$

where, MCP=Metal concentration in plants; MCW=Metal concentration in water.

2.6 Identification of functional groundwater spinach before and after phytoremediation process using infrared spectroscopy

This study identifies with IR to see the presence of the Zn, Cu, and Pb metal ion binding process in kale plants. Thus, researchers can review the ability of water spinach plants to reduce Zn in Lake Tempe. A total of ± 1 mg of finely groundwater spinach dry powder was mixed with approximately 100 mg of dry and finely ground KBr powder in a mortar. Subsequently, the mixture was pressed with a special hydraulic press (KBr pellet disc) at a pressure of 10,000-15,000 pounds/inch, to form a round, thin, translucent plate. These KBr plates or pellets were formed in a vacuum (while pressed by a hydraulic tool) to avoid inclusions, then installed in the cell and placed in the beam's path, to form the spectrum.

3. RESULTS AND DISCUSSION

The presence of metals in water at high concentrations can pollute the water so that it has an impact on environmental pollution and living things in the waters. It will also affect the food chain that impacts humans. By carrying out phytoremediation, it is hoped that it can reduce metals (Zn, Cu, Pb) pollution in the waters of lake Tempe.

3.1 Physicochemical properties of lake Tempe

Table 1 shows the average results of the physicochemical test results of lake Tempe contaminated with Zn, Cu, and Pb in phytoremediation experiments using kale plants. The measurements of physicochemical properties were carried out before and after 30 days of the phytoremediation process.

The comparison of pH, TDS, TSS, N, and P contaminated with Zn, Cu, and Pb metals in Lake Tempe before and after phytoremediation is shown in Figure 2. pH or degree of acidity indicates the level of acidity or alkalinity of a solution by measuring the concentration of hydrogen ions (H+) in the solution. According to Tyas, the pH value can be used to predict water quality. At pH=7 the water is classified as neutral. For pH<7, water is classified as acidic while for pH>7, water is classified as alkaline. Figure 2.a shows the pH comparison of Zn, Cu, and Pb metals which decreased before and after phytoremediation. The pH test before phytoremediation was Zn 7.34; Cu 7.55; Pb 7.42 and after process pH value decreased to 7.32; Cu7.48 and Pb 6.91. The results of the pH analysis showed a decrease in pH in the lake Tempe water samples. This is likely due to the presence of fallen plant parts [34], and the oxidation process of sulfate formation [35]. The presence of absorption of heavy metals by plants also affects the decrease in pH.

Total Dissolved Solids (TDS) or dissolved solids are types that are smaller in size or smaller than suspended solids. Dissolved materials in natural waters are not toxic, but when they are excessive, they can increase the turbidity value, which in turn inhibit the penetration of sunlight into the water and ultimately affect the photosynthetic process in the water [36]. The TDS content is also directly proportional to the level of turbidity in the water, such as the higher the TDS concentration, the higher the turbidity level [37]. The results (Figure 2.b) showed that the TDS value before phytoremediation increased by Zn 147 mg/L to 261 mg/L; Cu 159 mg/L to 225 mg/L; and Pb 137 mg/L to 212 mg/L after phytoremediation. This occurred as a result of the deposition of several organic compounds on the growing media in lake water. The higher the concentration of pollutants, the higher the TDS value in lake Tempe.

Total Suspended Solid (TSS) refers to solids suspended in waste, with a size below 0.45 microns [38]. The suspended material causes turbidity, which can block sunlight from penetrating the water plants [39]. Figure 2.c showed the results of the TSS tests at Tempe Lake show that a TSS value of Zn 94 mg/L to 130 mg/L; Cu 60 mg/L to 158 mg/L; and Pb 86 mg/L to 167 mg/L after phytoremediation. The increase in TSS value is linear with the TDS value, which means that there is an increase in dissolved organic or inorganic compounds in the lake Tempe water that comes from the water spinach growing media.

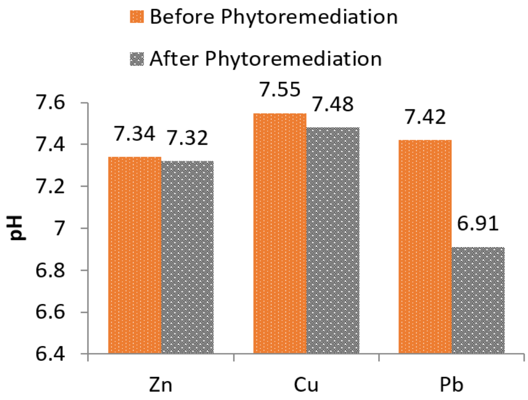
The total nitrogen and phosphate content in water is one of the parameters that determine water pollution. Along with the process of photosynthesis and decomposition in plants, there is also a decrease in the concentration of Total N and P. Figure 2.d showed the measurement of the total nitrogen content was carried out in the lake water obtained from Tempe yield Zn 1.9670 mg/L increasing to 6.9041 mg/L, Cu 1.9670 mg/L increasing to 3.7659 mg/L, and Pb 2.4209 mg/L increasing to 8.7871 mg/L. While the total phosphate as P from Zn 0.0162 mg/L increased to 0.0277 mg/L, Cu 0.0417 mg/L increased to 0.1679 mg/L, and Pb 0.0651 mg/L increased to 0.0688, this is due to the fine roots, plant stems, or fallen leaves, which then rot in the lake Tempe.

The results of the analysis of the Zn concentration in lake Tempe showed a significant decrease. The Zn concentration decreased from 1.6926 mg/L to 0.0743 mg/L. This shows the effect of water spinach as a phytoremediation medium in reducing heavy metal levels. The functional groups contained in water spinach can bind with metal ions such as carbonyl, amino, thiol, hydroxyl, phosphate, and hydroxy-carboxyl which are on the cell walls [40, 41]. The reaction of Zn metal with functional groups contained in plants has been confirmed using the Infra-Red instrument. Statistical analysis in this study was not available because of the limited data available.

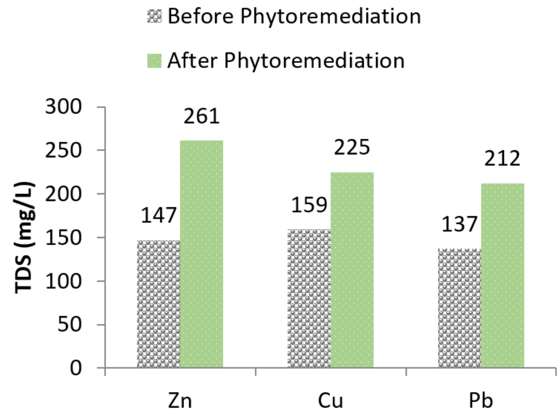
Table 1. Physicochemical properties of Tempe lake + Zn, Cu, and Pb waste in phytoremediation experiments with water spinach plants

Parameters	Before phytoremediation			After phytoremediation			Unit	Test method
	Zn	Cu	Pb	Zn	Cu	Pb		
pH	7.34	7.55	7.42	7.32	7.48	6.91	-	SNI 06-6989.11-2004
TDS	147	159	137	261	225	212	mg/L	SNI 06-6989.27-2005
TSS	94	60	86	130	158	167	mg/L	SNI 06-6989.3-2004
N	1.9670	1.9670	2.4209	6.9041	3.7659	8.7871	mg/L	AOAC Official Method 973.48.18th Ed, 2005
P	0.0162	0.0417	0.0651	0.0277	0.1679	0.0688	mg/L	SNI 06-6989.31-2005

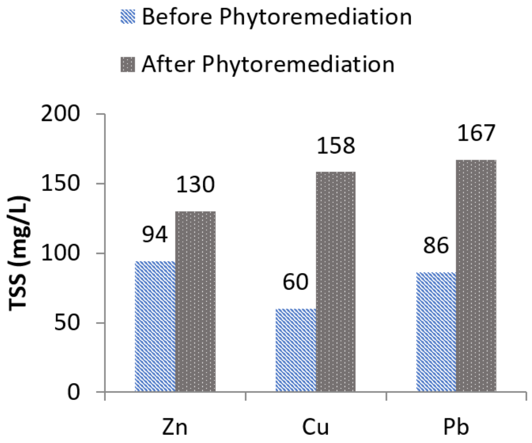
Notes: TDS=Total Dissolved Solids; TSS=Total Suspended Solids; N=Total Nitrogen; P=Total Phosphate.



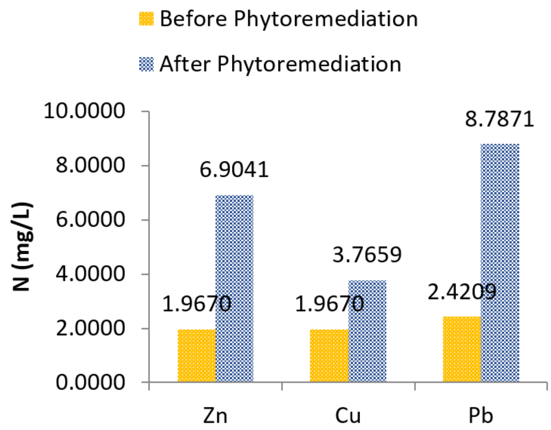
(a) pH comparison of Zn, Cu, and Pb metals in Lake Tempe



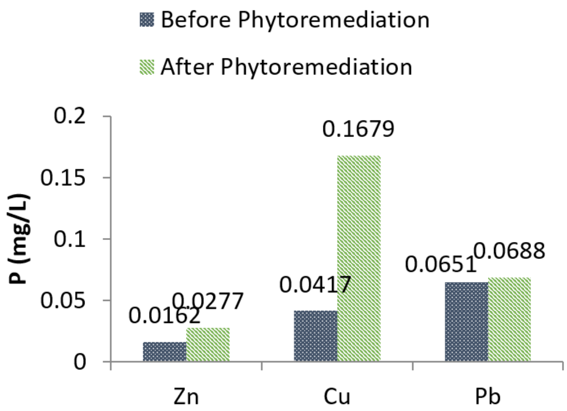
(b) TDS comparison of Zn, Cu, and Pb metals in Lake Tempe



(c) TSS comparison of Zn, Cu, and Pb metals in Lake Tempe



(d) N comparison of Zn, Cu, and Pb metals in Lake Tempe



(e) P comparison of Zn, Cu, and Pb metals in Lake Tempe

Figure 2. a) pH; b) TDS; c)TSS; d) N; and e) P comparison of Zn, Cu, and Pb metals in Lake Tempe before and after phytoremediation

3.2 Concentration of metals in water spinach plants and lake Tempe contaminated with metals

The measurement of the concentration of metals absorbed by water spinach and measurement of metals concentration in polluted lake Tempe for 30 days using the Atomic Absorption Spectrophotometer (AAS) method is shown in Table 2.

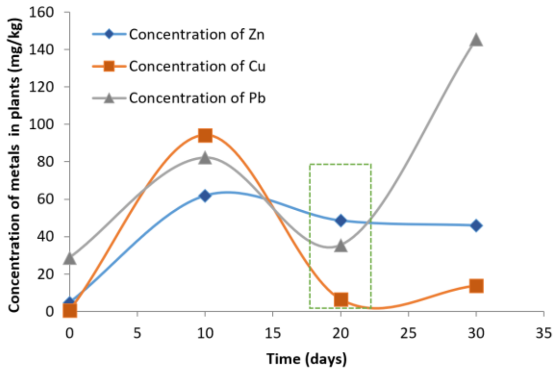
Table 2 shows the results of measurements of metal concentrations of Zn, Cu, and Pb in plants and lakes. Metal content in plants on day 0 measurement was Zn 4.5740 mg/kg; Cu 0.5 mg/kg; and Pb28.6234 mg/kg continued to increase until it reached its maximum absorption point on the 30th day, which was Zn 45.9628 mg/kg; Cu13.6609 mg/kg; and Pb145.3690 mg/kg. Table 2 can be illustrated into the graph in Figure 3 to see the influence of *Ipomoea aquatica* on decreasing metal concentrations.

However, from the 10^{th} to the 20^{th} day (Figure 3.a), the Zn concentration in plants decreased quite drastically. It is believed that this is because water spinach plants have reduced their ability to absorb heavy metals. After all, the plants were already saturated. On the 30th day, the metal concentration in the plant increased again, meaning that the plant could absorb the metal again.

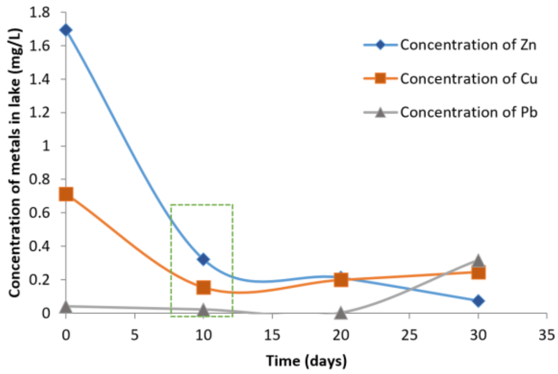
Figure 3.b shows the inverse of metal content in the lake. The 10th day showed a significant decrease in metal concentrations due to the activity of kale as a phytoremediation medium which was quite good. The higher the phytoremediation process, the concentration of kale (as an absorbent medium) increased the plant Zn concentration, while the Zn concentration in Lake Tempe decreased.

Bioconcentration (BCF) is a tendency for a chemical to be absorbed by aquatic organisms. The BCF results from the ratio between the concentration of chemicals in aquatic organisms with the concentration of chemicals in water [42]. The higher the BCF value in an organism, the higher the organism accumulates heavy metals. Table 2 shows the BCF value from the first day (day 0) to 30 has increased in value. This shows that the ability of water spinach as a phytoremediation medium is quite effective in absorbing heavy metals. To prove the accuracy of the BCF results, a statistical test was carried out using the One Way ANOVA method by comparing the three concentration results to the metal absorption activity. Results of testing the effectiveness of water spinach absorption on Zn, Cu, and Pb are shown in Table 3.

The significance value >0.05 indicates that water spinach absorption activity against metals that have polluted the lake has the same effectiveness against metals Zn, Cu, and Pb. So it can be concluded that water spinach is an effective phytoremediation medium in reducing metal pollution.



(a)



(b)

Figure 3. a) Illustrate of comparison concentration metals in Plants and b) Illustrate of comparison concentration metals in Lake

Table 2. The concentration of Zn, Cu, and Pb in water spinach plants and Tempe lake contaminated with metals

Concentration of metals	Time (days)	Plants (mg/kg)	Lake (mg/L)	BCF (L/kg)
	0	4.5740	1.6926	2.70
Zn	10	61.8734	0.3218	192.27
	20	48.5471	0.2121	228.89
	30	45.9628	0.0743	618.61
	0	0.5000	0.7125	0.70
Cu	10	94.2785	0.1548	609.03
	20	6.4804	0.1994	32.50
	30	13.6609	0.2469	55.33
Pb	0	28.6234	0.0408	701.55
	10	82.2266	0.0218	3771.86
	20	35.3788	0.0020	17689.40
	30	145.3690	0.3169	458.72

Table 3. Results of testing the effectiveness of water spinach absorption on Zn, Cu, and Pb

ANOVA

Concentration in plants

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4200.126	2	2100.063	1.155	.358
Within Groups	16364.486	9	1818.276		
Total	20564.612	11			

3.3 Results of identification of functional groups water spinach before and after the phytoremediation process by using infrared spectroscopy

3.3.1 Water spinach IR data before and after the phytoremediation process with lake Tempe water + Zn waste

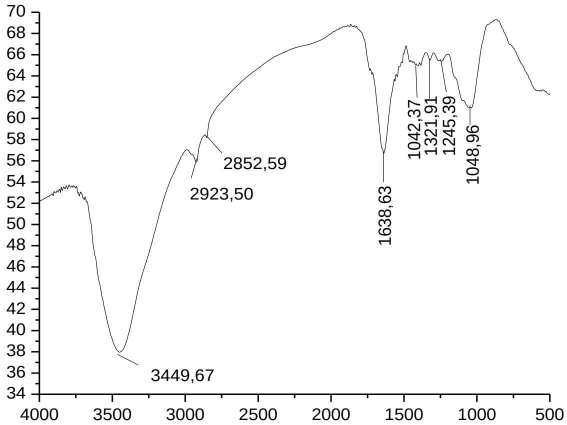


Figure 4. IR water spinach before the phytoremediation process

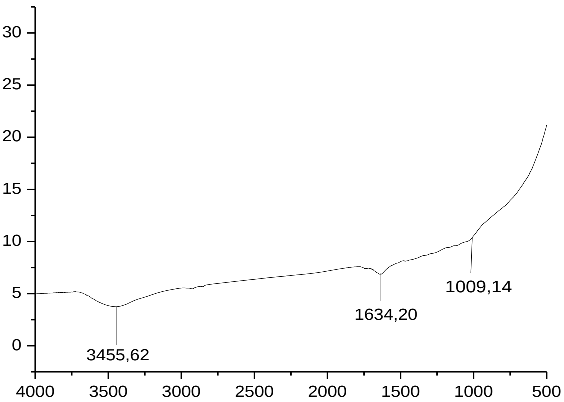


Figure 5. IR Zn content in water spinach after the phytoremediation process

3.3.2 Water spinach IR data before and after the phytoremediation process with lake Tempe water + Cu waste

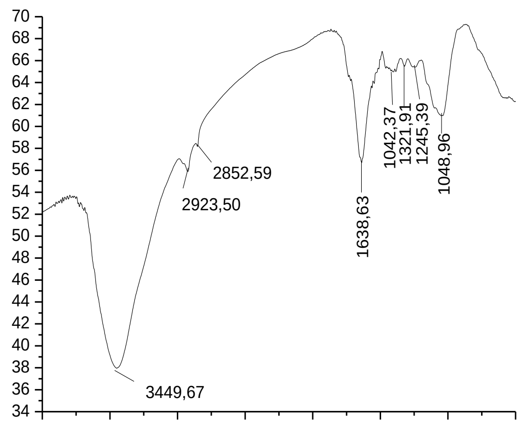


Figure 6. IR water spinach before the phytoremediation process

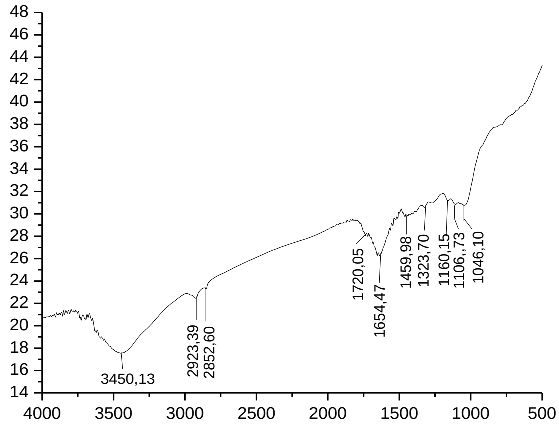


Figure 7. IR Cu content in water spinach after the phytoremediation process

3.3.3 Water spinach IR data before and after the phytoremediation process with lake Tempe water + Pb waste

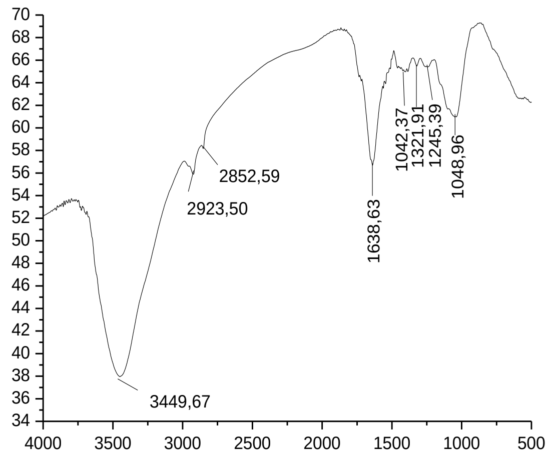


Figure 8. IR water spinach before the phytoremediation process

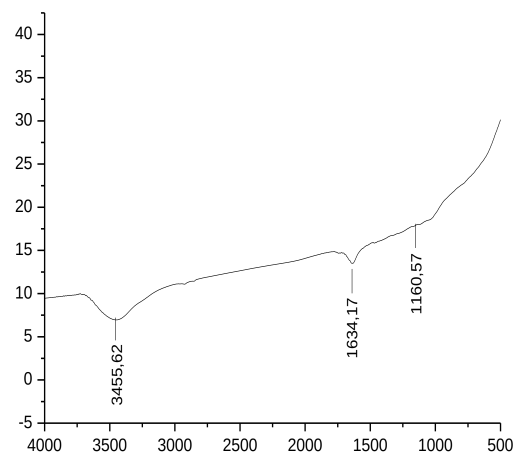


Figure 9. IR Pb content in water spinach after the phytoremediation process

IR data on water spinach powder shown at Figure 4 and 5, that there was a change in the wavelength before and after the phytoremediation process in the C=S group of wavelengths1048.96 cm^{-1} becomes a wavelength of 1009.15

cm^{-1}. This causes a change in the wavelength of about 39.81 cm^{-1}, and the change in the peak indicates a metal ion binding process Zn in the C=S functional group that occurs in water spinach. The C=N group before phytoremediation has a wavelength of 1638.63 cm^{-1}, while after=1634.20 cm^{-1}, there was a change in the wavelength of 4.43 cm^{-1}, which indicates that there is binding to metal ion Zn in the C=N group [43, 44]. The C=N group has the potential to bind metal ions because it has a lone pair. When viewed from the HSAB (*Hard and Soft Acid-Base)* properties of the metal ion Zn(II) is included in the borderline acid category and the C=N functional group found in plants is included in the soft base category so that it allows a strong bond between the Zn (II) metal ion and the C=N functional group. There was also a change in wavelength from 3449.67 cm^{-1} to 3455.62 cm^{-1}, indicating that there was the binding of the metal ion Zn to the OH group.

IR data on water spinach powder shown at Figure 6 and 7, that there is a shift in wavelength before and after the phytoremediation process in the C=S group from a wavelength of 1048.96 cm^{-1} to a wavelength of 1160.15 cm^{-1}. This causes a shift in the wavelength of about 111.19 cm^{-1}. This peak shift indicates that there is a process of binding Cu metal ions to the C=S functional group found in water spinach. The C=N group before phytoremediation had a wavelength of 1638.63 cm^{-1}, while after phytoremediation 1654.47 cm^{-1}, there was a shift in wavelength of 15.84 cm^{-1}, which indicated the binding of Cu metal ions to the C=N group. There was also a shift in wavelength from 3449.67 cm^{-1} to 3450.13 cm^{-1}, indicating the binding of Cu metal ions to the O-H group. The dominant functional group that plays a role in the Cu metal sorption process in water spinach plants is the C=S group. The C=S group has the potential to bind metal ions because it has a lone pair of electrons. When viewed from the HSAB properties, Cu(II) metal ions are included in the borderline acid category and the C=S functional group found in plants is included in the soft base category so that it allows for a strong enough bond between Cu(II) metal ions and the C=functional group S.

IR data on water spinach powder shown at Figure 8 and 9, that there is a shift in wavelength before and after the phytoremediation process in the C=S group from a wavelength of 1048.96 cm^{-1} to a wavelength of 1160.57 cm^{-1}. This causes a wavelength shift of about 51.02 cm^{-1}. This peak shift indicates a process of binding Pb metal ions to the C=S functional group found in water spinach. The C=N group before phytoremediation has a wavelength of 1638.63 cm^{-1}, while after phytoremediation 1634.17 cm^{-1}, there is a shift in wavelength of 11.83 cm^{-1}, which indicates the binding of Pb metal ions to the C=N group. There was also a shift in wavelength from 3449.67 cm^{-1} to 3455.62 cm^{-1}, indicating the binding of Pb metal ions to the O-H group. The dominant functional group that plays a role in the Pb metal sorption process in water spinach is the C=S group. The C=S group has the potential to bind metal ions because it has a lone pair of electrons. If viewed from the nature of the HSAB metal ion Pb(II) is included in the borderline acid category and the C=S functional group found in plants is included in the soft base category so that it allows for a strong enough bond between the Pb(II) metal ion and the C=functional group S.

4. CONCLUSIONS

Phytoremediation significantly facilitates heavy metals absorption, to reduce water pollution, by using plant media. This method is also suitable for contamination by toxic heavy metals, including lead, cadmium, and chromium, in wastewater. In this study, water spinach was used as a phytoremediation agent, to reduce zinc (Zn), copper (Cu), and lead (Pb) pollutants in Lake Tempe water. The largest adsorbed Zn(II) metal ion in water spinach was 61.8734 mg/Kg on the 10th day, Cu(II) metal ion which was the most adsorbed on the 10th day was 94.2785 mg/Kg, and the metal ion Pb(II) which was the largest adsorbed was on the 30th day of 145.369 mg/Kg. There was an increase in the levels of Zn, Cu, and Pb in water spinach plants along with the length of time the phytoremediation process took place, this is in accordance with the theory which states that the longer the phytoremediation time is used, the more metals can be adsorbed. And it was found that there was a decrease in the concentration of Zn and Cu in polluted tempe lake water (plant media) after the phytoremediation process was carried out for 30 days. This indicates that metal adsorption occurs in plants. The IR data supports this in the presence of zinc (Zn), copper (Cu), and lead (Pb) bonds in plants involving the functional groups C=S, C=N, and OH.

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