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CASE STUDY OF PHYTOREMEDIATION OF CU, FE AND ZN BY USING VETIVER GRASS (*VETIVERIA ZIZANOIDES*)

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Abstract:

The intensification of geological and anthropogenic activity has rendered the presence of heavy metals a significant environmental problem globally. The significant discharge of heavy metals into the soil induces enduring consequences on the environment, as they cannot be decomposed into non-toxic forms. Phytoremediation is an economical green method suitable for environmental preservation. The success of phytoremediation depends on numerous parameters, including plant behaviour and the uptake levels of both organic and inorganic contaminants. Vetiver Grass (*Vetiveria zizanioides*) possesses significant potential for phytoremediation due to its remarkable characteristics. The objective of the study is to assess the presence of heavy metals in Vetiver Grass and soil, as well as the correlation between the plant and the soil. The research was performed in a greenhouse to ensure adequate sunlight for the plants while protecting them from precipitation. This study used a one-factor completely randomised block design in which the concentrations of a synthetic Zn mixture (treatment) are established at five distinct levels (control, 250 ppm, 450 ppm, 750 ppm, 950 ppm), determined by the soil toxicity level aligned with the tolerance range of Vetiver Grass to that toxicity. The concentrations of copper and iron in the soil were established as criteria. During the experiment, all three elements, Cu, Fe, and Zn, accumulated in the plants during the course of one month of growth. The results indicated a statistically significant change in available Zn before and after the planting of Vetiver Grass and treatments (p value: 0.006). This investigation may enhance understanding of the mechanisms of pollutant uptake and the build-up of heavy metals under varying analytical settings.

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Introduction

Heavy metal pollution is a global health concern due to its long-lasting effects on ecosystems. It originates from industrial sewage, fuel production, mining, smelting, chemicals, agriculture, brick kilns, and coal combustion. Conventional methods involve transportation, chemical treatment, and disposal, but these are costly and can damage soil structure and ecology. Phytoremediation, a promising green technology, uses green plants to purify contaminated areas (Salt et al., 1998; Truong et al., 2010; Valderrama et al. 2013; Banerjee et al. 2016; Rashid & Mohd, 2024). Vetiver Grass, a more productive plant, is suitable for phytoremediation due to its adaptability to high concentrations of pollutants and its ability to produce high biomass (Suelee et al., 2017). The Vetiver System, developed in farmlands for soil and water conservation, has been successfully applied in Australia to conserve mine residue and contaminated soils with heavy metals. It is widely used for environmental protection due to its effectiveness, affordability, and practicality. Malaysia's rapid industrialization and urbanization have led to the incorporation of pollutants, including heavy metal pollution from geologic and anthropogenic activities. This pollution disrupts the environment and ecology, causing reduced growth of plants and animals. The accumulation of heavy metals in plants and uptake in human tissues contributes to high risks of human health and environmental concerns. Heavy metals in agricultural runoff can also harm aquatic plants and animals. The study aims to determine the availability of heavy metals in Vetiver Grass and soil, and the relationship between plant and soil. The significance of this study is that it will provide information on the uptake mechanism of contamination, physicochemical parameters, and heavy metal absorption by Vetiver Grass from contaminated soil. This could enhance knowledge about phytoremediation and contribute to the development of a Vetiver System in Malaysia. Additionally, it could establish guidelines for treating contaminated soil and improve soil quality.

The entire objective of phytoremediation technique is to provide a solution that can conserve environment and human health. This study was conducted to determine the availability of heavy metal in the Vetiver Grass and soil, and relationship between plant and soil.

Vetiver Grass is mostly used for control soil erosion and slope stability in Malaysia, thus phytoremediation is yet to be explored. Based on the study, it would give the information on the measure of contaminations uptake mechanism, as far as physicochemical parameters and heavy metals by Vetiver Grass. Besides that, this study highlighted on potential absorption of heavy metal by Vetiver Grass from the contaminated soil. It is hypothesized that Vetiver Grass would remove higher amount of heavy metal from the contaminated soil. In this this study, it will enhance the knowledge about phytoremediation by using Vetiver Grass in the soil under different level of soil toxicity and tolerance level of Vetiver Grass to the toxicity. Furthermore, this could contribute as the standard data for the development of Vetiver System in Malaysia. In additional to that, this study could contribute in establish guidance associated contaminated soil quality improvement by providing guidelines in treating contaminated soil.

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Literature Review

Phytoremediation

According to Pivetz (2001) Phytoremediation technique is the utilization of vegetation to reduce certain contaminants such as polluted soil, mud, silt, underground water, lakes, and sewage. Darajeh *et al*. (2014) were suggested phytoremediation is the usage of plants to remediate contaminated soils, water or sediments. Roongtanakiat and Chairoj (2001) were claimed phytoremediation is a second method that utilize green plants to conserve a polluted area. Plants exhibit reactions to mitigate the effects of various heavy metals. The prolonged accumulation of heavy metals in soils adjacent to industrial areas include iron (Fe), copper (Cu), nickel (Ni), cobalt (Co), cadmium (Cd), zinc (Zn), arsenic (As), and mercury (Hg). Many of these metals are essential micronutrients that govern various normal plant functions (Soumya et al., 2022). Phytoremediation involves employing plants to eliminate, absorb, or stabilise potential pollutants, including heavy metals, metalloids, trace elements, organic compounds, and radioactive substances from soil or water. Metallophytes possess distinct genetic and morphological characteristics that enable them to develop biological mechanisms for survival and reproduction in metalliferous soils, while being toxic (Yuan et al., 2019). A variety of engineering methods and biological procedures have been developed to improve the phytoremediation process. Phytoremediation has been employed to validate the efficacy of particular plants for environmental remediation (Iqbal et al., 2019 as cited in Rashid & Mohd, 2024). The accomplishment of phytoremediation a green technology is depend on several factors. At this point, the vegetation must produce adequate biomass to accumulate high concentrations of heavy metal. Next, agricultural practices must be responsive to the metal-accumulating plants such as practice repeated cultivation and removing of the toxic tissues (Paz-Alberto & Sigua, 2013). Phytoremediation is a different technique to purify polluted soil that are easy to implement, reduce remedial cost as well as environmental friendly in comparison with other methods such as chemical treatment and soil excavation method (Roongtanakiat & Chairoj, 2001). There are several types of phytoremediation process: phytoextraction, phytostabilization, phytodegredation, phytovolatilization, and rhizofiltration (Lee, 2013 as cited in Pulford *et al*., 2003)

Special features of Vetiver Grass suitable for Phytoremediation

Morphological Attributes

Dudaia, Putievskya, Chaimovitcha, and Ben-Hurb (2006) were confirmed Vetiver grass (*Vetiveria zizanoides*) belongs to the Poaceae family and it's one of the perennials grasses. According to Paz-Alberto and Sigua (2013) the Vetiver Grass has several unique characteristic (as cited in the National Research Council). Vetiver grass is a perennial grass that can penetrate their roots three meters into the ground and lengthwise their shoots two meter high. Vetiver Grass can grows and strive both condition of xerophytic as well as hydrophilic. It has a vigorous dense of roots that grow vertically. It has narrow blade, long and coarse that sprout from the base of the clumps. The leaf able to extend 45 to 100 centimeter long and 6 to 12 centimeter wide (Paz-Alberto & Sigua, 2012).The plant has a strongly massive and vertical root system which is descending two to three meters deep in soil for the first year, eventually can extend five meters deep under tropical climate. The ability of root hairs to penetrate deep in soil enhance tolerance of plants toward shortage of water, able to great infiltrate soil moisture and extend through hard soil film. The plant has ability to form solid vegetative barrier that decrease water flow and filters as well as trap sediments in runoff water. It is also strongly tolerance to

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infestation pests and diseases, fire, and high grazing stress (Truong & Loch, 2004). According Chomchalow (2003) the plant has ability to filter both fine and coarse sediment effectively due to special feature of thick growth forming living porous barrier. The finely structured and massive root system of plant enhances the environment to stimulate microbiological processes in the rhizosphere (as cited in Truong & Baker, 1998).

Physiological Characteristics

Phytoremediation by using Vetiver Grass is highly recommended due to its extraordinary features. Special feature of Vetiver Grass such as large and deep penetration of root system, drought and flood tolerance, able to thrive in submergence, fire and extreme cold condition as well as heat of waves make it appropriate to be used for phytoremediation (Paz-Alberto & Sigua, 2012). Truong and Loch (2004) was reported Vetiver Grass is resistance to temperature levels reaching from 20 degrees Celsius to 55 degree Celsius and it has been found to stand under high rainfall ranging from 300 millimeters to 6000 millimeter per year. It also has ability to recover quickly when contrived by adverse conditions such as drought, frost, fire and saline after weather improves. According to Truong (2000) Vetiver Grass has capability to adapt a wide range of soil pH ranging from 3.0 to 10.5. Truong (2000) assert that it highly adaptable to high acidity of growing medium, soil alkalinity and salinity. In addition, it also extremely greatly tolerant to heavy metals such as Cd, As, Pb, Ni, Cr, Hg, Zn and Se in the soils (Truong , 2000). According to Dudaia *et al.* (2005) Vetiver Grass able to thrive in various soil condition including sandy, clay, loamy sand, sandy clay loam, crushed limestone, as well as peat mixture.

Heavy Metals in Soils

Heavy metal pollution by metals has become a severe problem all over the world particularly in developing countries (Krami *et al.,* 2013). Heavy metal contamination could emerge naturally or by activities of human. Sources of natural occurrence of heavy metal contamination are explosion of volcanic, forest combustion as well as seepage of rock. The major contribution of heavy metal contamination is human activities including activity of mining, operation of smelting and a lot of other industries such as battery production industries and thermal power plants (Sharma *et al*., 2014). Sow, Ismail and Zulkifli (2013) has confirmed that heavy metal such as Cd, Pb, Zn, Ni, and Cu develop from chemical fertilizers application in agriculture, pesticides usage, and plowing activities, therefore prolong the accumulation of heavy metals in soil. Metals such as iron (Fe), copper (Cu), nickel (Ni), cobalt (Co), cadmium (Cd), zinc (Zn), arsenic (Ar), and mercury (Hg) are among the heavy metals that have been found to accumulate in the soils in close proximity to industrial locations over an extended period of time. The majority of these metals are essential micronutrients that govern a variety of processes that are routinely performed by plants (Soumya et al., 2022 as cited in Rashid & Mohd, 2024).

Methodology

Study Location

Experiments were performed in a greenhouse at Universiti Teknologi Mara Kampus Jasin Melaka to ensure adequate sunlight for the plants while protecting them from precipitation.

Experimental Design

This study consists of a one-factor completely randomised block design. The treatments in this experiment were duplicated five times. The experiment involved five distinct zinc treatments:

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(1) zero concentration (control), (2) 200 ppm, (3) 450 ppm, (4) 750 ppm, and (5) 950 ppm. The choice to employ five replications is intended to achieve a precise examination of heavy metal buildup in plants subsequently.

Experimental Methodology Flora Acquisition
This study utilised Vetiver grass, scientifically designated as *Vetiveria zizanioides*. The Vetiver grass (VG) was obtained from the Humid Tropic Centre (HTC) under the supervision of the Department of Irrigation and Drainage (JPS Malaysia). The vetiver grass utilised in this experiment was five months old. Vetiver grass was pruned to a height of 30 cm and a root length of 10 cm to 15 cm for each treatment prior to transplantation into polybags.

Synthetic Composite of Designated Heavy Metals
The treatment concentrations for the experiment were determined according to the environmental zinc threshold and the tolerance range of Vetiver Grass to Zn, as detailed in Appendix 2. The treatment concentrations above the environmental threshold for Zinc, resulting in soil toxicity, although remained within the tolerance range of Vetiver Grass (Truong, 2000). A synthetic solution of 1.0 M Zinc sulphate ($ZnSO_4$) was created by dissolving 287 g of $ZnSO_4$ in 250 ml of distilled water, after which this solution was utilised as a therapy.

Preparation and Cultivation of Plants
Prior to initiating the experiment, the plants were meticulously rinsed without direct exposure to flowing tap water to avert injury, particularly to the roots. Subsequently, they were cleaned with distilled water to eliminate any pollutants present on the plant that could influence the experiment. Vetiver grass was pruned to a height of 30 cm, with root lengths ranging from 10 cm to 15 cm for each treatment. The treatments were conducted in 25 experimental polybags, measuring 10’’ X 13’’, filled with 4 kilogrammes of a synthetic heavy metal soil mixture, within the rain shelter. Each polybag contained one set of vetiver slips, each comprising three to four tillers, depending on the size of the tillers. The medications were administered one week after the plants had grown. The soil in each polybag was analysed before and after the application of treatment. The plants were harvested after one month of growth, and subsequently, the leaves and roots of the harvested plants were analysed.

Soil Sampling
The soil sampling was conducted prior to the execution of the experiment. This investigation comprised three parameters: the availability of copper and iron in the soil. The cleaned auger was utilised to take soil samples from three plots at the UiTM Jasin sharing farm. Soil samples were taken at a depth of 15 centimetres from each plot to acquire the topsoil. The samples were packed in new polyethylene bags and promptly transported to the laboratory for analysis. The soil samples were collected on a typical day, ensuring the absence of severe rainfall to prevent excessive moisture. Three readings were recorded for each chemical measurement to get the average value. The pH level of the soil and the organic matter content were analysed for each sampled plot. Approximately 20 ml of each sample solution was transferred into vials for analysis using the Inductively Coupled Plasma (ICP) equipment to identify the concentration of specific heavy metals.

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Soil Analysis

The study aimed to determine the available concentrations of zinc (Zn), iron (Fe), and copper (Cu) in soil using the Mehlich No. 1 extraction method. The Mehlich No. 1 extracting solution was prepared by combining 4 mL of concentrated hydrochloric acid (HCl) and 0.7 mL of concentrated sulfuric acid (H_2SO_4) in a 1000 mL volumetric flask, and subsequently diluting the solution to volume with distilled water to obtain a mixture of 0.05 N HCl in 0.025 N H_2SO_4. For extraction, 5.0 g of oven-dried soil, sieved to 2 mm, was weighed into a 50 mL plastic vial. Twenty millilitres (20 mL) of the Mehlich No. 1 extracting reagent was added to the soil, and the mixture was agitated for 5 minutes on a reciprocating shaker. The suspension was then filtered through Whatman No. 2 filter paper, and the filtrate was collected and stored for subsequent analysis of Zn, Fe, and Cu concentrations as shown in Figure 1.

Plant Analysis

To determine the total concentrations of Zn, Fe, and Cu in plant tissues, a dry ashing technique was employed on both leaf and root samples. The collected plant tissues were placed in paper envelopes and oven-dried at 60°C for 24 to 48 hours, or until a constant weight was achieved. The dried samples were ground into a fine powder using a mechanical grinder. An aliquot of 0.5 g of the ground tissue was weighed into a clean porcelain crucible. The samples were burned in a muffle furnace, first at 300°C for 1 hour, and then the temperature was increased to 550°C and maintained for 6–7 hours. After cooling, a small amount of distilled water was added to moisten the ash. Then, 2 mL of concentrated HCl was added and the mixture was evaporated on a hot plate. Subsequently, 10 mL of 20% nitric acid (HNO_3) prepared by diluting 200 mL of concentrated HNO_3 to 1000 mL with distilled water was added to the residue. The crucibles were heated in a water bath at 100°C for 30 minutes. The contents were then transferred and diluted to 100 mL with distilled water in a volumetric flask, followed by filtration into a 100 mL vial. The elemental concentrations of Zn, Fe, and Cu were determined using an Inductively Coupled Plasma (ICP) spectrometer as shown in Figure 1.

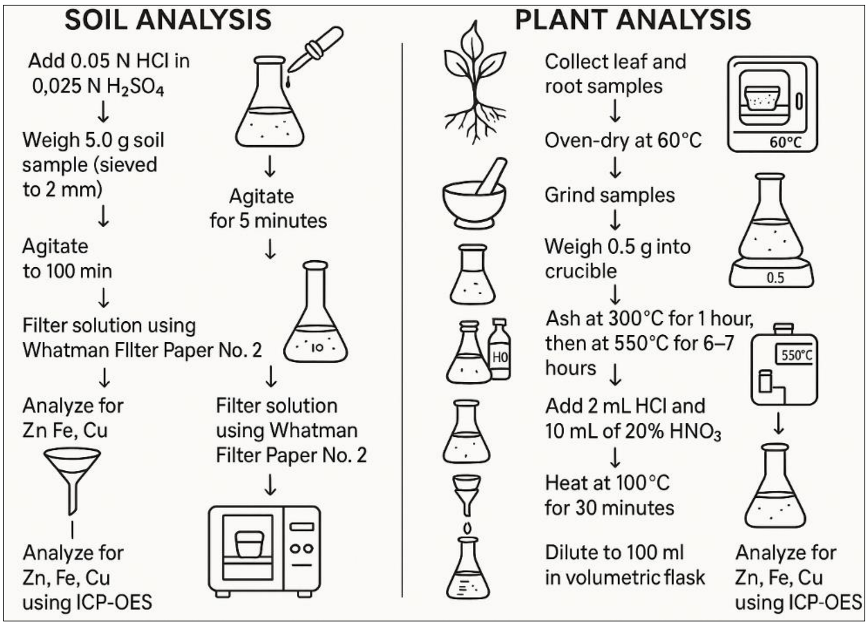

Figure 1: Soil Analysis and Plant Analysis procedures

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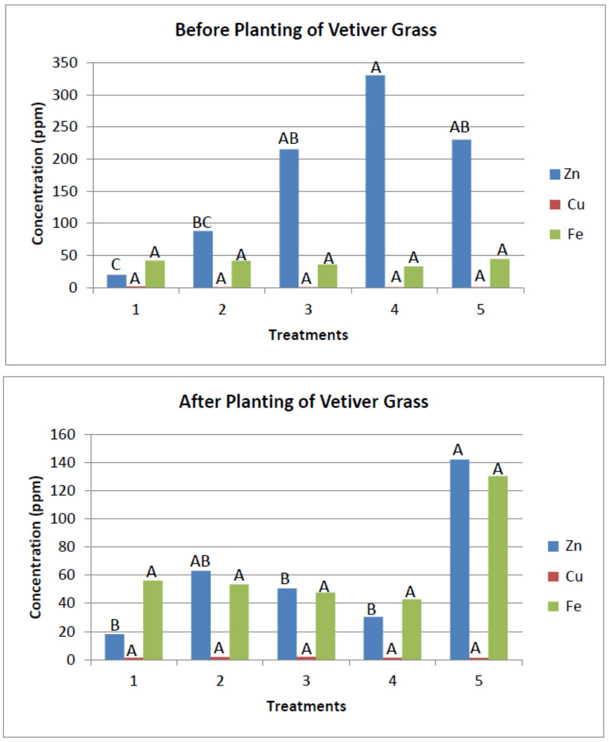

Figure 2: Concentration Of Zn, Cu And Fe In Soils Before And After Planting Vetiver Grass

From the bar chart above, the availability of Zn, Cu, and Fe in soils between before and after planting of Vetiver Grass (VG) shows statistically significant different. Data analysis shows Zn (p value: 0.000), Cu (p value: 0.044), Fe (p value: 0.000). However, there is no significant different between Cu and treatments, Fe and treatments except Zn (p value: 0.006) after 30 days planting of VG. Results of the study proved that Zinc, Fe and Al were not available in VG except Cu after 30 days planting.

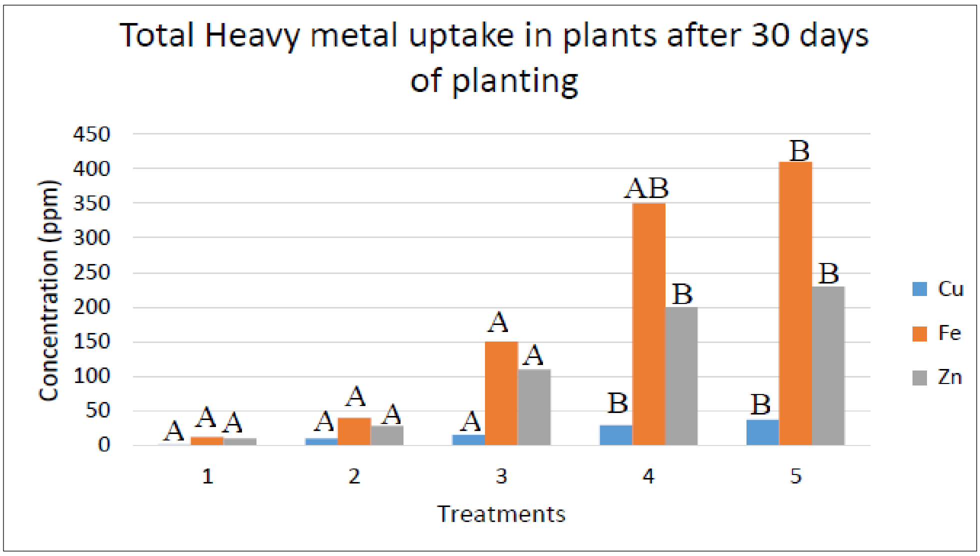

Figure 3: Accumulation of Cu, Fe and Zn in Plant Shoots And Roots

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The bar chart indicates that Cu, Fe, and Zn exhibit increased accumulation in VG following treatments administered 30 days post-planting. The data demonstrates a considerable difference between each treatment, particularly between treatments 4 and 5. This demonstrates that VG is highly recommended for the absorption of heavy metals in soils, particularly for Cu, Fe, and Zn elements.

Discussion

Soil properties that have been used in the study as shown in Figure 2 may affect availability of Zn in VG. According to Alloway (2008) the soil factors such as pH, clay soil, presence of organic matter, calcium carbonate, and concentrations of other trace elements control the amount of zinc in the soil solution and its sorption-desorption from or into the soil solution. Those factors affect availability of Zn in soil and plant. The clay soil that has been used in experiment may affect the availability of Zn in VG. Adsorbed and exchangeable zinc held on surfaces of the colloidal fraction in the clay soils control the concentrations of Zn in the soil solution as well as Zn availability to plant roots. In addition, Kabata-pendias (2000) confirmed clays soils are capable to hold Zn very strongly. Therefore, Zn solubility in soils is less than Zn hydroxide, Zn carbonate, and Zn phosphate in pure experimental system (as cited in Lindsay, 1972). Furthermore, Thompson and Troeh (2005) argues that Zn can occur inside clay structures by isomorphic substitution in octahedral sites as the Zn^{2+} is strongly adsorbed on the cation-exchange sites of clays (as cited in Obrador *et al*., 2003). MacBride and Blasiak (1979) claims the nucleation of $Zn(OH)_2$ on surface of clay soil of can result the highly pH dependent detainment of Zn in soil .The competition between Zn and other cations resulted, Zn easily mobile and leach from light acid soil due to adsorption of Zn^{2+} can be decreased at lower pH (<7) . Kabata-pendias (2001) declares availability of Zn in soil and plant is connected mostly with hydrous Al and Fe oxides ranging from 14 percent to 38 percent of total Zn and with clay minerals ranging from 24 to 63%, while 1 to 20 and 1.5 to 2.3% freely mobile fractions and complexed by organic matter (as cited in Zyrin *et al*., 1976). Recent discovery have admitted these calculation, reveal that the fraction of clay govern up to 60% of Zn available in soils (Kabata-pendias, 2001 as cited in Kabata-Pendias *et al*., 1995). In her 2000 book, Kabata-Pendias argues that soluble silicic acid enhance Zn adsorption by clays (as cited in McBride, 1978). Moreover, Garcia-Miragaya, Cardenas and Page (1986) reported that low concentrations of Zn were sorbed more strongly by kaolinite due to a high fraction of fragile acidic edge sites exist on surface of kaolinite. Rieuwert *et al*. (1998) believes existing of Hydrous Fe and Mn oxides arise in clays as a thin layer on phyllosilicates and as mobile crystal and gels (as cited in Kinniburgh & Jackson, 1981). The existence of Hydrous Fe and Mn oxides in clay soil may decrease concentrations of Zn in soil solution due to precipitation process and adsorption reactions (Elliott, Liberati, & Huang, 1986; Chuan, Shu, & Liu, 1996).

Besides that, reaction of Zn with soil pH affects the availability of Zn in soils and plants. According to Prasad, Shivay, and Kumar (2016) solubility of Zn reduce 100 fold as each unit of pH increase (as cited in Lindsay, 1991). Rupa and Tomar (1999) were supported the view a sorption of Zn increase with each unit increase in pH ranging from 4.25 to 6.75. The Zn solubility in pH-dependent decrease as the pH increase resulting Zn+2 interact with organic matter. As a result, thirtyfold reduction in solution Zn+2 typically have been observed for every unit PH increase between 5 and 7 (Alloway, 2004). Availability of Zn decrease with increase of soil pH (Kaleeswari, *et al*., 2013). Kaleeswari *et al*. (2013) acknowledges Al and Fe oxide, organic matter and $CaCO_3$ increase as the soil pH increase. Prasad and Power (1997) supported as the pH-dependent charge increase, the sorption of Zn on hydrous Fe and Al oxides, soil OM, and clay soil minerals will increase. Rieuwerts et al. (1998) states critical pH values adsorption

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of Zn on Fe oxides are approximately and 5 to 6.5 for Zn (as cited in Kinniburgh & Jackson, 1981). Farrah and Pickering (1977) were confirmed at pH level of more than five increase proportions of metal ions bound as hydroxy complexes. Xian and Shokohifard (1989) supported the view that a decline in soil pH ranging from 7 to 4.55 increased exchangeable levels Zn.

The presence of organic matter in the soil that has been used in this study may affect availability of Zn in soil and plant. The main sorbents in soils are organic matter and clay minerals. Those soils components play an important role in micronutrient retention that release in soil (Hooda, 2010).Brady and Weil (2008) were explained more than half of soil solution Zn^{2+} complexed by organic matter cause it is low in soil solution (2-70 ppb). Brady and Weil (2008) generally mentioned the negative charges that presence from various soil particles have tendency to engage and bind cations and inhibit them from being soluble and disperse to surface of root hence, reduce bioavailability of metal in soil. This is a major problem that can affect successful of phytoremediation technique (Roongtanakiat, 2009). According to Roongtanakiat (2009), application of organic fertilizer reduced metal toxicity in VG through the binding of the heavy metals into the OM. Alloway (1995) was supported organic matter has a high adsorptive capacity at pH levels of 5 and above.

Next, the figure 2 shows statistically significant different between availability of Cu in soil before and after planting of VG (p value: 0.044). The result shows the availability Cu increase significantly after planting of VG, whereby only Cu is available in VG after 30 days planting. Yet, the experiment shows the availability of Zn in soil before and after planting of VG significantly decreased (p value: 0.000). However, there is no Zn accumulated in VG after 30 days planting. Based on the study, there are connection between availability of Cu and availability of Zn in soils and plants. According to Kabata-Pendias (2001), Cu is very versatile and stable cation in soils and capable to chemically interact with mineral greatly. Kabata-Pendias (2001) confirmed the interaction of Cu and Zn was normally observed (as cited in Graham, 1981; Rinkis, 1972). The author in her book says these metals supposedly absorbed through same carrier sites in absorption mechanism; therefore aggressively prevent root absorption of each other. Kabata-Pendias (2001) was reported the antagonistic interaction between Zn and Cu have been identified in which the uptake on one elements was competitively inhibits by other. According to Prasad *et al*. (2016), Cu and Zn being divalent cations and compete with each other for adsorption sites on soil clay minerals, even though affinity for Cu is more than that for Zn (as cited in Eliott *et al*., 1986). Metals are adsorbed specifically based on the preferential order Cd < Zn <Cu < Pb (Alloway, 1995). According to preferential order, root surface prefer to adsorb Cu than Zn, thus Cu is available in plant. Adsorption of metals may occur due to affinity of metal cations toward particular adsorption site that specifically adsorb metal cations (Basta &Tabatabai, 1992).

Available Fe in soil may affect availability of Zn in plant and soil. Based on study, the availability of Fe significantly increases after planting of VG (p value: 0.000). Kabata-Pendias (2001) confirmed that the interaction of Fe and Zn is clearly connected with the franklinite precipitation that can depress the availability of both metals (as cited in Pluford, 1982). The relative mobility of Fe was found to be inversely related to the mobility of Zn and interfered more with absorption of Fe and translocation (Kaleeswari *et al*., 2003). Species and genotype of VG may also influence availability of Zn in VG due to its differential responses towards metals and soil components. According to Roongtanakiat (2009), VG require sufficient

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biomass to absorb the heavy metal as it is non-hyperaccumulator plants. The Zn was not available in VG also due to insufficient time to produce adequate biomass for metal accumulation as the VG was planted only 30 days as shown in Figure 3. The author express enormous amount of biomass is needed to increase efficiency of phytoremediation technique. Paz-Alberto and Sigua (2012) were supported that less amount of biomass production can limit efficiency in phytoremediation process. Plant must produce sufficient biomass in order to accumulate high concentrations of Zn. Roongtanakiat and Chairoj (2001) were mentioned that VG prefer to absorb Cu than other metals at low soil pH (5.4) resulted increase in Cu absorption, thus available in VG (as cited in Baker & Senft, 1997). Roongtanakiat and Chairoj (2001) claims the highest mean metal accumulations in the shoot of the three VG ecotypes for Zn (253.8 mg kg-1) lower than the toxic tolerance levels in VG shoot in comparison for Cu (46.6 mg kg-1) that was more than the toxic threshold level of VG (13-15 mg kg-1) (as cited in Truong, 1999). Based on that proves, it supported that VG prefer to accumulate Cu in their shoot in comparison of Zn.

Conclusion

This project is implemented in order to explore the potential of phytoremediation technique in monitoring heavy metal in soil by using Vetiver Grass (VG). The objectives are to determine the availability of heavy metal in plant and soils, and to determine the relationship between plant and soil. This project is considered to have successfully satisfied the objectives stated in this project.

Throughout the experiment, the VG is believed adaptable toward high toxicity level of Zn. Based on the observation; VG still looked vigorous and healthy after a week applying synthetic mixture of Zn in soil. Yet, symptoms of the VG towards the treatment have been observed after 20 days planting. Some of VG started show up chlorosis symptom followed by browning of leaves. To summarize, there is no plants died during the experiment although symptom of chlorosis has been show up on a few plants.

Recommendation

The study has limitations, including the short project duration, unhealthy plants, and inaccuracies in synthetic mixture preparation. The one-month cultivation period is insufficient for plants to grow and absorb metals, and the short harvesting interval may limit success. The plants used were unhealthy due to late transplanting and the method used for preparing the mixture may not accurately reach the required concentration. Recommendations include implementing the project over a longer period, increasing the time interval for harvesting, and ensuring the plants are in a vigorous and healthy condition. The use of standard solutions for preparing the synthetic mixture is also recommended.

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