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Screening of Aquatic Plants for Potential Phytoremediation of Heavy Metal Contaminated Water

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ABSTRACT

Bioremediation is a new green economic approach in providing solutions for cleaning up contaminated sites. Phytoremediation uses plants as a tool for remediation purposes. The usage of plant species offers higher potential solution to remediate heavy metal contaminated sites. This study aimed on screening potential plant species for phytoremediation of heavy metal contaminated water. The potential of three aquatic macrophytes species (*Eichorrnia crassipes*, *Pistia stratiotes* and *Ipomoea aquatica*) for chromium and nickel phytoremediations was tested. The plants were exposed for 10 days under hydroponic conditions in heavy metal contaminated water. *E. crassipes* showed the highest chromium and nickel concentrations in its biomass, 1.60 and 2.40 µg/L respectively. Meanwhile, *P. stratiotes* had chromium and nickel concentrations detected at 0.89 and 0.081 µg/L, respectively; chromium and nickel concentrations of *I. aquatica* detected were, 0.49 and 0.08 µg/L, respectively. The ability of these plants to accumulate heavy metals and survived throughout the experiment demonstrates the potential of these plants to remediate metal-enriched water. Among the three tested aquatic plants, *E. crassipes* was proven to be the most suitable plant species that can phytoremediate heavy metal contaminated water followed by *P. stratiotes* and *I. aquatica.*

INTRODUCTION

Nowadays a lot of heavy metal trace elements are reported to contaminate the environment and each of these elements affects the public health. Some heavy metal elements are naturally available in nature. However, the addition of heavy metals released from point and nonpoint sources promotes accumulation and difficult to control. The main point sources are recognized as household sludge, sewage treatment and industrial activities while other point source includes residential area, solid waste disposal sites and commercial lots [1].

Heavy metal polluted water can affect human health and there have been cases of heavy metal contamination in China due to large amount of chromium slag discharged from chemical industries [2]. Chromium (Cr) is the seventh most abundant element on earth's crust [3]. Exposure to high levels of Cr in a short time can produce irritation at the site of contact, including ulcers on the skin and irritation of the nasal mucosa [4]. Nickel (Ni) is the 24th most abundant element in the earth crust, 3% of the earth's composition. It is an essential nutrition trace metal for several species of animals, plants and microorganisms [5]. Nickel is widely used in industries for the production of stainless steel and other high corrosion and temperature resistance alloys. Nickel metal and its alloys are used widely in the metallurgical, chemical and food processing industries, especially as catalysts and pigments [5].

In Malaysia, the water quality in rivers is deteriorating as many rivers and coastal areas are reported to be contaminated with heavy metal. Rivers and water bodies especially in industrial areas have been experiencing increasing levels of heavy metal contamination. A study showed, there was heavy metal contamination of various heavy metals including Cr and Ni in soil beneath the waste disposal site at Dengkil, Selangor [6]. Port klang coastal areas have been reported to have significant presence of these heavy metals in water and sediments [7] as well as Sungai Buloh River with a severe concentration of Zn, Cu, Ni and Pb [8]. This condition increases the concern of clean water availability in the future and raises the concern of the treatment cost.

Therefore, researchers are interested in finding cost-effective and greener options as an attempt to solve these problems. Bioremediation offers great potential and efficiency and many studies have been conducted to explore this process especially by using microorganisms. Phytoremediation started receiving increase attention as some constituent in contaminants such as metal elements cannot easily be treated by microorganisms. There were also studies on the combination of microorganisms and plants [9].

Phytoremediation is a set of technology that uses plant to extract and degrade chemical compounds including metals, pesticides, hydrocarbons, and chlorinated solvents. It is also widely known as a green remediation and receives interest for its low cost, effectiveness and eco-friendliness. Phytoremediation has several different types, which include phytofiltration or rhizofiltration, phytostabilization, phytovolatilization, phytodegradation and phytoextraction [10]. Phytoextraction is where the plant can absorb metal from soil and translocate the metals to its shoot [10]. Maize is an example of a phytoextractor since it can decrease the percentage of cadmium in the planted soil [11]. Phytostabilization is generally used to describe plants that could remediate soil, sediment and sludge. This type of phytoremediation allows limitation of contaminant mobility in the soil by the plant roots. The reduction of water percolation would prevent a direct contact with the polluted soil and consequently prevent the spreading of toxic to other sites [10]. Rhizofiltration targets a low concentration of contaminants from ground water, surface water and wastewater. This type of phytoremediation utilizes plants that can retain the contaminant within its roots [12].

As this paper focuses on heavy metals in contaminated water, aquatic plants offer suitable and accessible properties as candidates. Three species of macrophytes that were chosen for this study were *Eichhornia crassipes*, *Pistia stratiotes* and *Ipomoea aquatica*. Macrophytes are aquatic plants that can grow in or close to water and can either be emergent, submergent or floating. This includes emergent macrophytes, floating leaved macrophyte, submerged macrophyte and free-floating macrophyte. They can absorb nutrients through their root systems and are very productive [13]. *I. aquatica* is categorized under emergent macrophytes; and *E. crassipes* and *P. stratiotes* are categorized under free-floating macrophytes [14]. The emergent macrophyte, *I. aquatica* has elliptic leaves and adventitious roots [15]. *E. crassipes* has thick and rounded leaves and has feathery root, meanwhile, *P. stratiotes* has thick hairy leaves and adventitious roots [16].

MATERIALS AND METHODS

Sample collection and preparation

All the plant samples, *E. crassipes, P. stratiotes* and *I. aquatic* were harvested from lakes in Universiti Putra Malaysia, Serdang Selangor, Malaysia. The plants were washed thoroughly to remove mud, dirt and particulate matters and acclimatized for three days. The fertilizers used in this study were Hoagland nutrient solution and hydroponics fertilizer [17].

Treatment of plant samples with heavy metals

Heavy metal treatment started after the acclimatization period. Chromium and nickel were added into separate containers with different concentrations 1, 2, 3, 4, 5 µg/L, respectively. Each batch of heavy metal had triplicates for each biological sample. The concentration of heavy metal was monitored for 7 days before the plants were taken out and air-dried at room temperature. The air-dried sample was then harvested for extraction. The treatment was done in a hydroponic system, which used liquid as the medium instead of soil [18]. Each species was put into a separate container (**Fig.** 1).

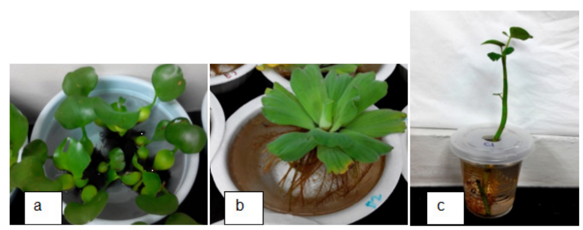

Fig. 1. The macrophyte plants used in this study (a) *E. crassipes* (Keladi bunting) (b) *P. stratiotes* (Kiambang) and (c) *I. aquatica* (Kangkung).

Heavy metal extraction and standard curve preparation

The air dried sample was harvested for extraction. An acid digestion technique [19] was used to extract heavy metal from the samples. About 0.3 g of roots was weighed and then added with 2 mL of 30% hydrogen peroxide (H_2O_2) and 8 mL of 60% nitric acid (HNO_3) followed by heating at 60 °C for 3 h. The sample was cooled to 40 °C and added with distilled water up to 50 mL. The sample was filtered using whatman 150 mm filter paper and the absorbance reading was recorded. In order to determine the wavelength of maximum absorbance of heavy metal, 1 mL of chromium (Cr) and nickel (Ni) solutions were prepared. The solution with known concentration absorbance reading of Cr was taken from 300 nm to 600 nm with 20 nm intervals while the absorbance reading of Ni was taken from 200 nm to 600 nm with 20 nm intervals. The recorded data was then analyzed using standard curves (data not shown). Two heavy metal solutions (Cr and Ni) of known concentration were prepared with HNO_3 and H_2O_2 as the dilution agent with the same portion in extraction to allow a comparable result with the filtrate samples.

RESULTS AND DISCUSSION

The uptake of Cr and Ni in *Eichorrnia crassipes*

E. crassipes showed a gradual increase in Cr accumulation with different Cr concentrations (**Fig.** 2). The average chromium detected was 0.7, 0.92, 1.06, 1.34 and 1.60 µg/L, respectively. The result for each sample was compared with control plant, k value (1.06 µg/L). From five treatment concentrations, the first two treatment concentrations (1 and 2 µg/L) displayed lower values (0.70 and 0.92 µg/L) than the k value while the last two treatment concentrations (4 and 5 µg/L) showed higher concentrations (1.34 and 1.60 µg/L) compared to the k value.

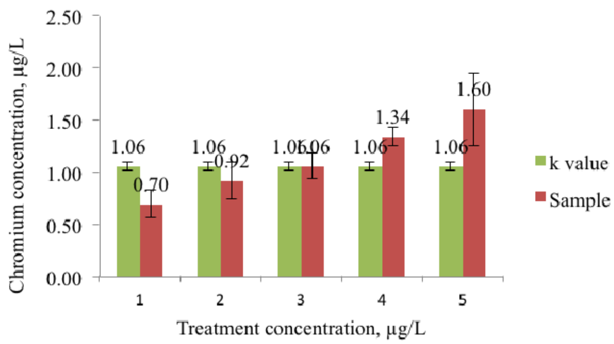

Fig. 2. *E. crassipes* treated with different concentrations of Cr. The graph shows concentration of Cr detected in five different concentrations of Cr treated *E. crassipes* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

The average concentration of ni detected under different treatment concentrations was 0.97, 1.84, 2.24, 2.40 and 2.30 µg/L, respectively (**Fig.** 3). The result of Ni treated sample was compared with control plants, k value (1.53 µg/L). From five treatment concentrations, four treatment concentrations (2, 3, 4 and 5 µg/L) showed higher concentration of Ni (1.84, 2.24, 2.40 and 2.30 µg/L) than the k value except in 1 µg/L treatment where the concentration was recorded at 0.97 µg/L.

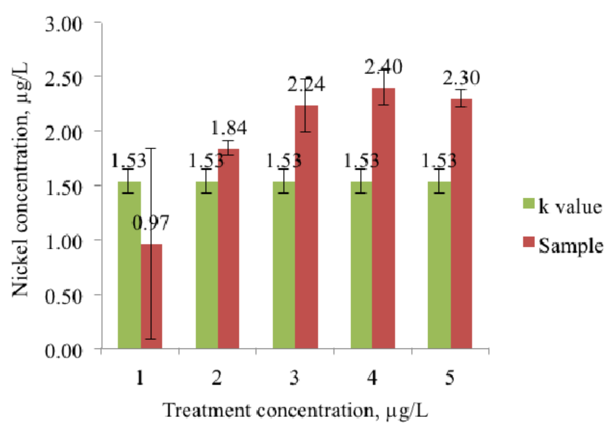

Fig. 3. *E. crassipes* treated with different concentrations of Ni. Concentration of Ni detected in five different concentrations (1, 2, 3, 4, 5 µg/L) of Ni treated *E. crassipes* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

The ability of this species to absorb both heavy metals was comparable with previous study of Sood et al. [14]. They reported that *E. crassipes* could absorb up to 1.25 and 1.68 mg/L of Cr and Ni, respectively; it was almost ten times higher in concentration from our study.the ability of the *E. crassipes* to absorb Cr has also been reported by Mishra and Tripathi [20] and Akinbile et al. [21].

Plants have three different patterns of metal uptake which are; true exclusion that totally restrict metals from being absorbed into the plant, shoot exclusion that accumulate metals only in root by restricting metal translocation to shoot and accumulation that allow metals to be in plant parts. The hyper accumulators can withstand uptake of high levels of certain heavy metals that can be toxic to organisms [22]. The pattern from the analyzed data from this study postulates that *E. crassipes* is an accumulator.

The uptake of crand Ni in *P. stratiotes*

A fluctuating trend across the increased concentration treatment of 1, 2, 3, 4 and 5 µg/L Cr was detected in *P. stratiotes* (**Fig.** 4). The average concentration of cr detected under different treatment concentrations was 0.74, 0.58, 0.89, 0.86 and 0.65 µg/L, respectively. From five treatment concentrations, three treatment concentrations (1, 3 and 4 µg/L) displayed higher cr concentration (0.74, 0.89 and 0.86 µg/L respectively) than the k value while the other 2 treatment concentrations (2 and 5 µg/L) showed lower concentration (0.58 and 0.65 µg/L), respectively compared to the k value.

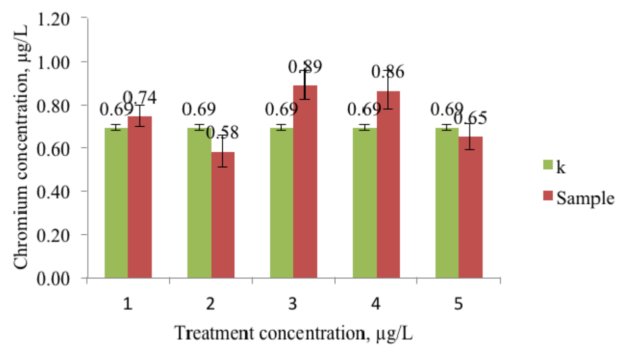

Fig. 4. *P. stratiotes* treated with different concentrations of Cr. Concentration of Cr detected in five different concentrations (1, 2, 3, 4, 5 µg/L) of chromium treated *P. stratiotes* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

Fig. 5 shows nickel concentration detected in *P. stratiotes* increased to 3 µg/L and decreased afterwards with average concentrations of 0.065, 0.07, 0.081, 0.063 and 0.054 µg/L, respectively. The nickel treated sample was compared with k value (0.06 µg/L) and five out of four treatment concentrations (1, 2, 3 and 4 µg/L) showed greater value (0.065, 0.070, 0.081 and 0.063 µg/L) than the k value.

The ability of the *P. stratiotes* to absorb Cr and Ni had also been reported by Lu et al. [23]. The study involved 14 elements (Al, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, Pb, and Zn). Cr and Ni were reported to absorb and accumulate within the root as compared to the other metals that were deposited on external root. Thus, this study postulates that *P. stratiotes* is an accumulator that did not retain the metals in its root as there are samples that has lower concentration of heavy metals compared to the k value [24].

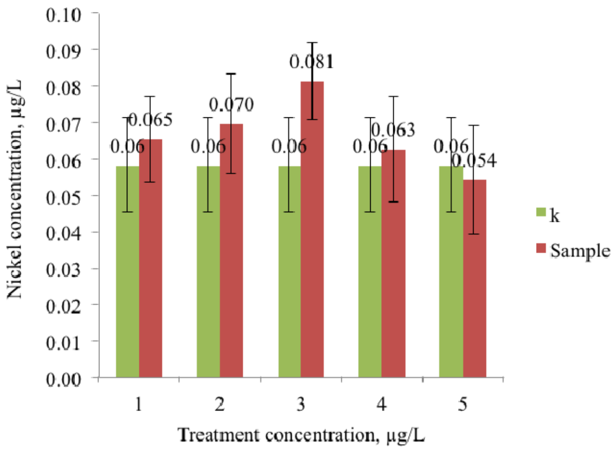

Fig. 5. *P. stratiotes* treated with different concentrations of Ni. Concentration of Ni detected in five different concentrations (1, 2, 3, 4, 5 µg/L) of Ni treated *P. stratiotes* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

The uptake of crand Ni in *I. aquatic*

Ipomoea aquatic shows significant difference between heavy metal treatment sample and k value, as control (**Fig.** 6). *I. aquatica* in Cr displayed increasing trend as average concentration of Cr detected were 0.29, 0.32, 0.49, 0.48 and 0.48 µg/L, respectively.

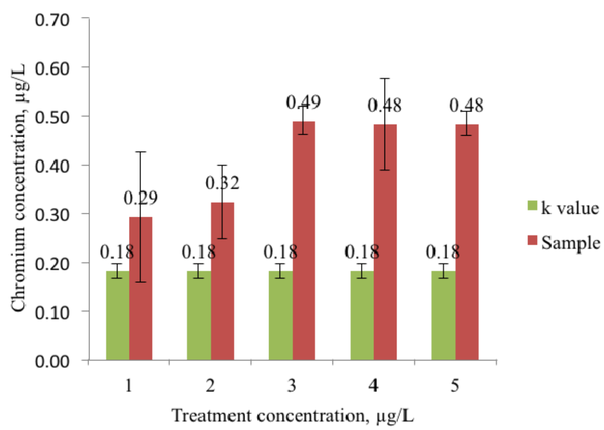

Fig. 6. *I. aquatic* treated with different concentrations of Cr. Concentration of Cr detected in five different concentrations (1, 2, 3, 4, 5 µg/L) of Cr treated *I. aquatic* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

I. aquatica in Ni-treated water showed increasing trend of nickel detected up to 3 µg/L and the concentration detected decreases with average reading of 0.012, 0.018, 0.03, 0.03 and 0.015 µg/L, respectively (**Fig.** 7). The Ni concentration increased in the first three treatment concentrations (1, 2 and 3 µg/L) and decreased in the following treatment concentrations (4 and 5 µg/L).

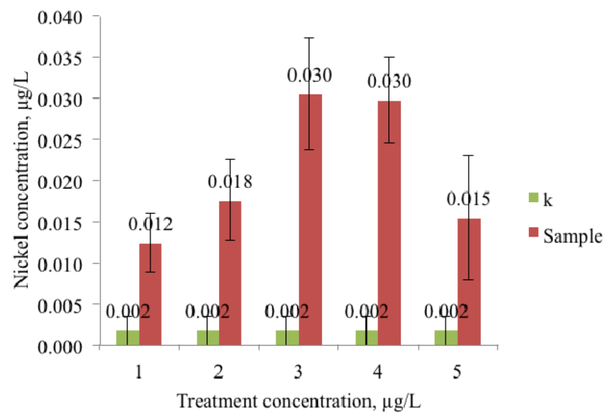

Fig. 7. *I. aquatic* treated with different concentrations of Ni. Concentration of Ni detected in five different concentrations (1, 2, 3, 4, 5 µg/L) of Ni treated *I. aquatic* with k value (green bar) indicates untreated sample and sample (red bar) indicates heavy metal treated samples. Vertical bar indicates standard error.

It has been reported that *I. aquatica* can absorb up to 13,217 mg $kg^{-1}$of Cr [25], almost a hundred times treatment concentration compared to the concentration of Ni used in this study. Therefore, this study postulated that *I. aquatica* is a shoot exclusion considering its significant difference of heavy metals concentration accumulated in root compared to the k value.

Among the three species, the highest Cr concentration detected was in *E. crassipes* with the highest average concentration of 3 µg/L. However, *I. aquatica* showed great difference of concentrations between the control and the heavy metal treated samples. Ni accumulation was also the highest in *E. crassipes*. Therefore *E. crassipes* was more permeable to ni and cr compared to *I. aquatic* and *P. stratiotes*. *I. aquatica* showed great difference between the control and the heavy metal treated sample, hence the species is postulated to be a shoot exclusion that retains heavy metals in its root. Meanwhile, *E. crassipes* and *P. stratiotes* were most likely the accumulators that can transport heavy metals to the other parts of plants. The accumulator has mechanisms to transport, distribute, and detoxify heavy metals to be used in plants, thus not retaining heavy metal in its form. The accumulator can detoxify heavy metals by heavy metal distribution in the apoplast cell wall [26]. Other than that, a study by Dahmani-muller et al. [27] showed that accumulators are able to detoxify heavy metals in leaf which required metal immobilisation from roots.

CONCLUSIONS

E. crassipes, P. stratiotes and *I. aquatica* showed good potential to phytoremediate water contaminated with heavy metals. Our data has shown that *E. crassipes* has the highest potential compared to both *P. stratiotes* and *I. aquatica*. The efficiency of the metal absorption is dependent on the morphology of the plant and characteristics of the plant and heavy metals.

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