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Phytoremediation of Selected Heavy Metals in Simulated Water Using Algae (*spirogyra spp*)

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Abstract— *The present study demonstrated the phytoremediation potential of algae for the removal of Cd, Ni, Fe, Pb, Cr and Mn. Young plants of equal size were grown in H_2O and supplemented with 1.0 mg/dm^3, 3.0 mg/dm^3 and 5.0 mg/dm^3 of multi component metal solution for a period of 15 days. The experiment showed that all samples was able to accumulate at all concentrations. The accumulation of metals increased with increase in the initial concentration of the solution. In conclusion algae can be used in remediating waste water.*

I. Introduction And Literature Review

Environmental pollution is one of the stern problems across the globe, the root can be traced back to anthropogenic activities such as ore mining and crude oil exploration, the processing and use of mining products that generate wastes which directly or in directly finds its way into water bodies, soils or emitted into the atmosphere leading to pollution. These pollutants when accumulated may have adverse effects on living organisms causing gradual degradation of ecosystem (Banach *et al.,* 2012).

Metals are also introduced into the aquatic ecosystems as a result of weathering of soil and rocks from volcanic eruptions and from a variety of human activities such as processing and use of metals and/or substances contaminated with metals (Worku and Sahu, 2014). In addition to this development, technology has widely disrupted the aquatic ecosystems by various pollutants damaging the aquatic ecosystems and water quality (Irfan *et al.,* 2015).

Heavy metals are metals with a density of 5g/cm^3 (Vardanyan and Ingole, 2006) According to Duffus, (2002), heavy metals can be defined in terms of their density (specific gravity), atomic weight (relative atomic mass), atomic number, chemical property, toxicity, and non-chemical aspect (guns/great ability). Some of the effects of these metals are as follows: cadmium can cause high blood pressure, kidney damage and sterility in males. Long term exposure can cause bone to become brittle, chromium is toxic to animals and humans but less so to plants (Miroslav and Vladimir, 1999).

Biotic remediation is a method involving the use of living organisms for the removal of pollutants in the environment which is presently at developmental stage. This technique called biotic remediation which is broadly used for water purification and soil reclamation, is classified based on the organism used; bioremediation (microorganism), mycoremediation (fungi), and phytoremediation (plant). phytotechnologies are an effective and valid alternative for the remediation of contaminated water bodies not only under experimental conditions but also under natural condition (Miretzky, 2004). Phytoremediation involves the use of plants for removal of environmental pollutants or detoxification of pollutants to make them harmless in the environment (Worku and Sahu, 2014).

II. Materials and Methods

Sampling

Three aquatic plants namely alga (*Spirogyra sp.*), water hyacinth (*Eichhornia Crassipes*), and water lettuce (*Pistia stratiotes*) were selected for the study. The alga was obtained from Usmanu Danfodiyo University, Sokoto (UDUS) fountain, water hyacinth from Kandoleshella stream, kwalkwalawa and water lettuce from Sokoto metropolis (behind sutan's palace). The plants were authenticated and identified by a curator in the Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto. The plants were then washed, collected in a clean plastic container and kept in biological sciences botanical garden for a period of one week to acclimatize to the environmental condition of the garden (Swain *et al.,* 2014).

Experimental Set Up

The experimental sets up for alga and water lettuce were conducted in a transparent container, while water hyacinth placed into a container of 15dm^3 capacity. The solutions of Cd, Cr, Ni, Mn, Fe and Pb were prepared at varying concentrations of 1 mg/l, 3mg/l and 5mg/l, nutrient in the form of Hoagland solution was added, while control sample contained only Hoagland solution as reported by the technique in John (2007), pH of the water was adjusted to 6 using acetic acid. The plants were weighed before administering the heavy metal solution and reweighed after. 50cm^3 of water sample was collected from each container after 24hrs, 5days, 10days and 15days of the experiment and 5 drops of HNO_3 acid was added for storage before AAS analyses which is in line with the technique used by John (2007).

The experiment was conducted for a period of 15 days in a shady area according to the technique reported by Banach *et al.,* (2012), at the end of the 15th day the plants were harvested, washed, dried and then analysed for metal accumulation using standard techniques below.

Sample Treatment

The plant was harvested on the 15th day and washed with distilled water to remove any adhered substance, it was then allowed to dry, and the air dried sample was grinded and sieved.

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Digestion

The ashing procedure was done in order to prepare the sample for elemental analysis. This was done using dry ashing method. (Miroslav and Vladimir, 1999).

Wet Digestion

Air dried sample (1.00g) was placed in a beaker, followed by the addition of HNO_3 ($10cm^3$, 6M) and $HClO_4$ ($2\ cm^3$). Each beaker was covered with a watch glass for an hour to allowed the initial reaction to subside. The beakers were heated on a hot plate with the temperature not exceeding 90^oC for 30 minutes. The contents were then filtered and transferred into volumetric flask ($50cm^3$) with subsequent washings and diluted with distilled water to the mark (Miroslav and Vladimir, 1999).

Procedure for AAS Analysis

The alpha 4 model of Atomic Absorption Spectrophotometer was set according to the manufacturer's instructions with the wavelength corresponding to that of the metal under investigation. Standard solutions prepared from the stock solutions and the blank were used to obtain the calibration curves. The absorbance of each sample was measured in triplicate with an automatic calculation of the average concentration in parts per million (ppm).

Statistical Analysis:

The analysis was carried out on three independent replicates for every parameter and the results presented in Table 3.1 are reported as mean ± standard deviation (SD). Data were analyzed by SSPS Anova two ways considering significance at an alpha level of 0.05.

III. RESULTS

The result of heavy metal analysis of the plant under investigation in a multi component metal solution is summarised in Table 3.1 and represented in Figures 3.1 to 3.6. The bioconcentration factor of the heavy metals is shown in Tables 3.2.

Results are expressed as mean±SD. Values with superscript are the first five best bioaccumulating plant/plant part at alpha 0.05.

3.1.1: Cadmium Concentration

Figure 3.1 reveals increase in heavy metal concentration in the sample with increase in concentration of solution, (i.e 1 mg/ dm^3 having least accumulation and 5 mg/ dm^3 having the best). The plant was able to accumulate fair amount of cadmium.

3.1.2: Nickel concentration

The variation in nickel concentration in algae in different solution under investigation is shown in Figure 3.2. Nickel concentration increases with increase in concentration of working solution as observed with respect to Cadmium absorption.

Table 3.1. Bioaccumulation of Heavy Metal by Algae

Plant(mg/kg)	**Concentration (mg/dm^3)**	**Heavy Metal**					
		Cadmium	Nickel	Iron	Lead	Chromium	Manganese
Algae (control)		ND	ND	910.13±0.02	ND	94.64±0.02	305.62±0.01
Algae	1	37.38±0.99	10.96±4.74	3.10.23±0.1	250.12±2.94	66.78±0.02	195.79±0.02
	3	133.34±0.99	79.18±4.74	11.18±0.18	292.37±6.1	178.89±0.03	388.39±0.01
	5	268.74±2.86	112.76±2.28	80.80±0.84	539.46±17.06	671.28±0.02	567.30±0.1

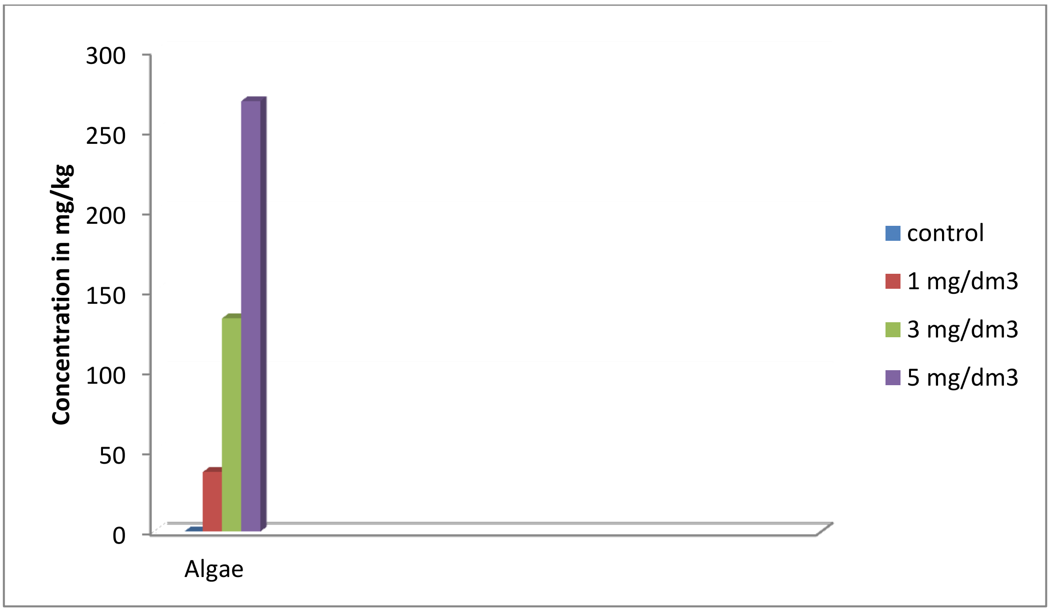

Figure: 3.1: Cadmium concentration in algae.

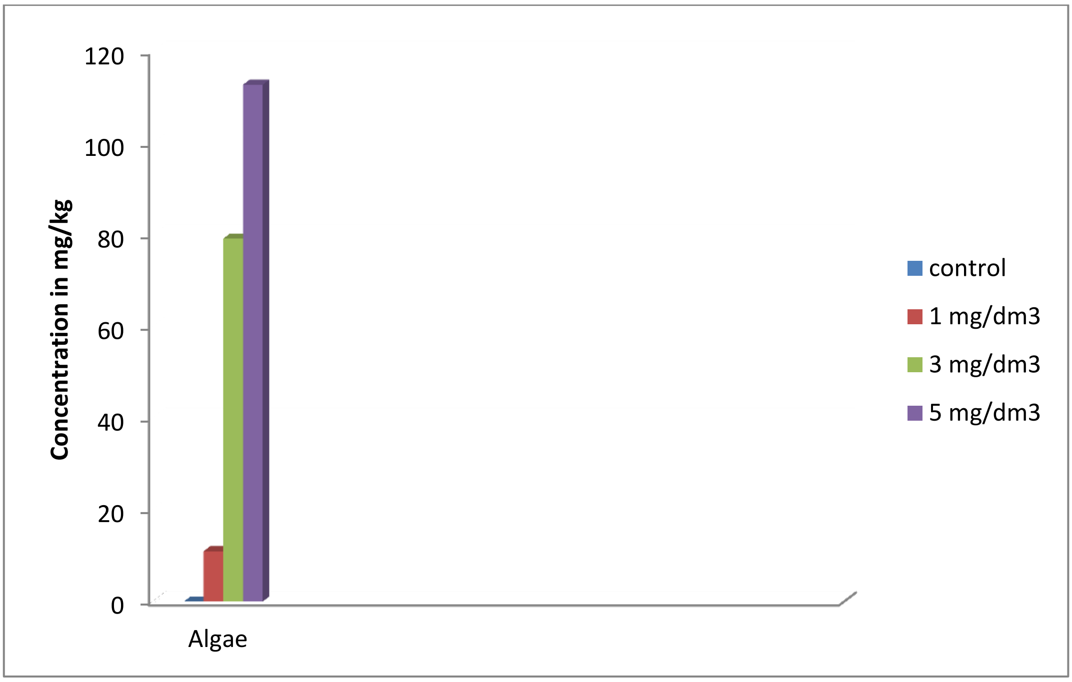

Figure 3.2: Nickel concentration in algae

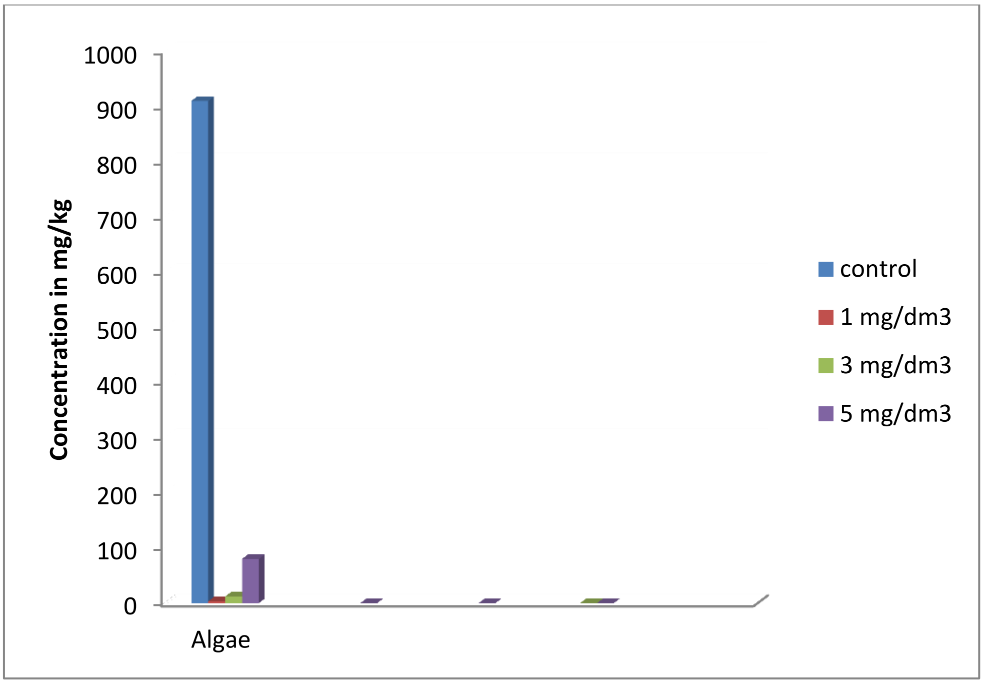

Figure 3.3: Iron concentration in algae

3.1.3: Iron concentration

Figure 3.3 gives the concentration of Iron in various solutions under investigation with similar trend in other metals having higher concentration of metal in plants as a result of increase in the concentration of the working solution.

3.1.4: Lead concentration

Concentration increase in the plant sample increases with increase in the working solution and in the control sample no lead was detected and the plant was able to accumulate fair amount of the metal with highest value in 5ppm solution.

3.1.5 Chromium Concentration

Figure 3.5 potrays chromium accumulation in algae under investigation, of all the metals in view highest accumulation was observed in chromium, with highest value in 5 mg/dm^3 solution.

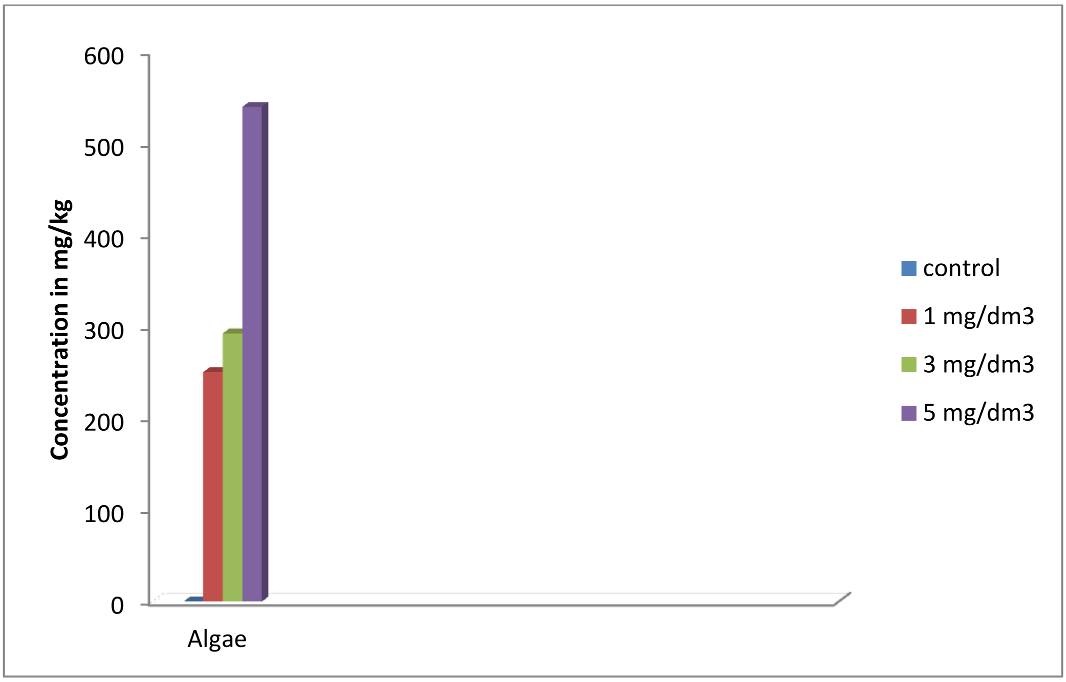

Figure 3.4: Lead concentration in algae.

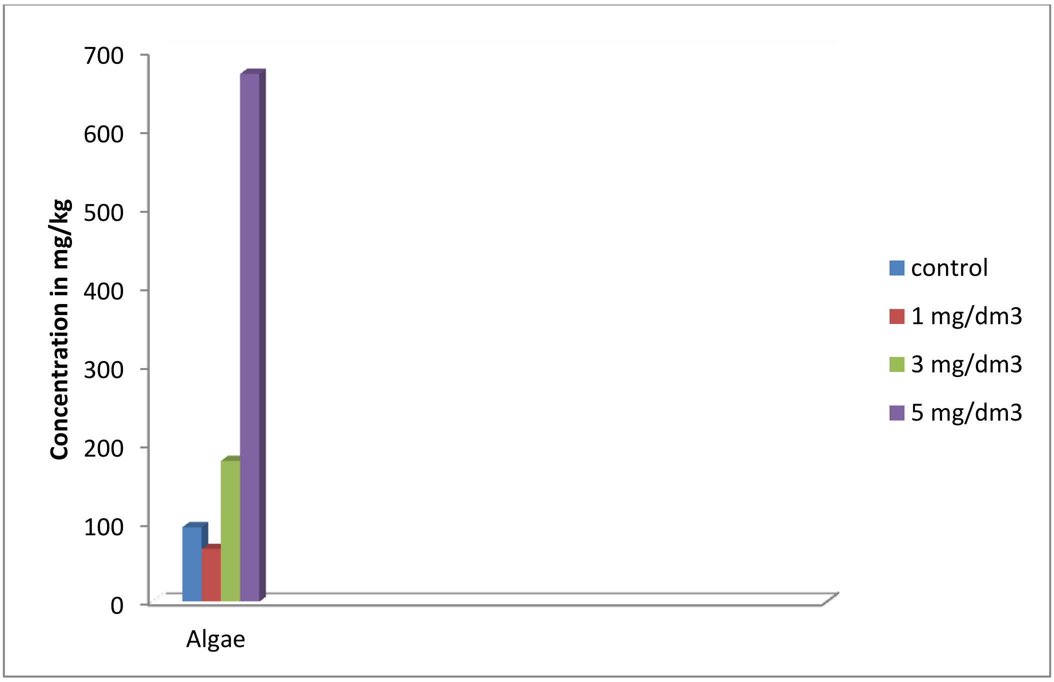

Figure 3.5: Chromium concentration in algae.

3.1.6: Manganese Concentration

There was considerably large amount of manganese detected in control sample, and high amount was accumulated by all various solutions under investigation, from Figure 3.6 below it can be observed that algae has the highest accumulation of manganese concentration following similar trend like the previous metals.

Bioconcentration Factor

Bioconcentration factor is a useful parameter for assessing the potential of trace metal accumulation. When metal concentration in water increases the amount of metal accumulated in plant increases thus the bioconcentration factor decreases. Bioconcentration factor also provides index of the ability of plant to accumulate metal with respect to the concentration of that metal in the substrate (Swain *et al*., 2014). Results in Table 3.1showed bioconcentration factor of algae. Larger value of the bioconcentration factor implies better accumulation capability (John, 2007).

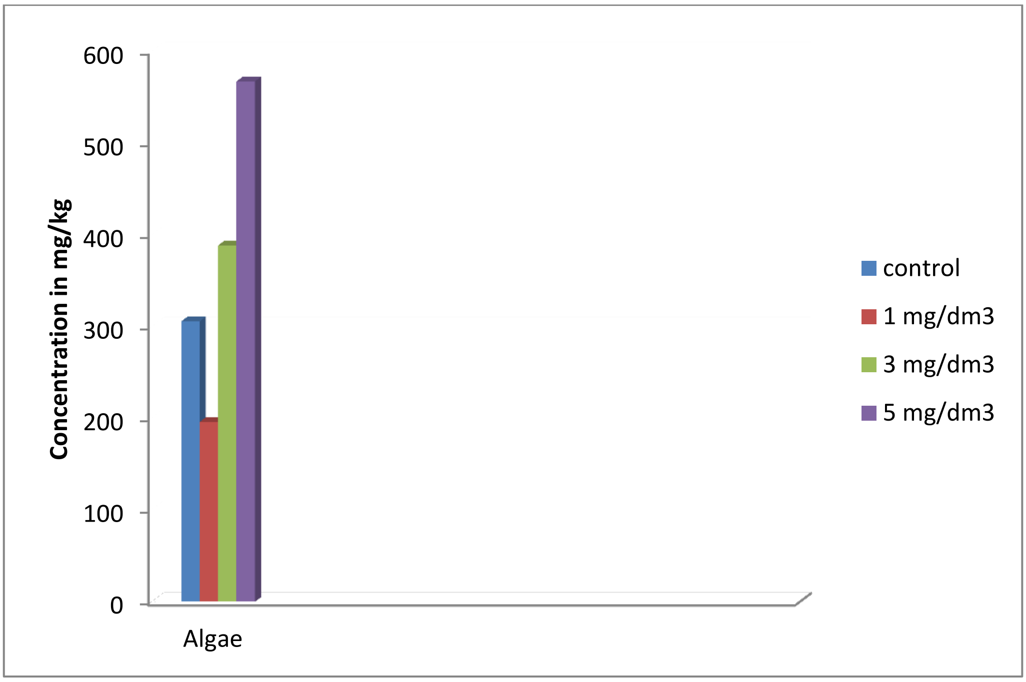

Figure 3.6: Manganese concentration in algae.

Table 3.2 Bioconcentration factor of Algae

Plant	**Concentration(mg/l)**	**Heavy metals(mg/kg)**					
		Cd	Ni	Fe	Pb	Cr	Mn
Algae	1	37.38	10.96	3.10	250.1	66.78	19.79
	3	44.45	26.39	3.73	97.46	59.63	129.46
	5	53.75	22.55	16.16	107.89	134.26	113.46

Bioconcentration factor =a/b, where a = metal concentration in plant and b = metal concentration in water (Ugya *et al.,* 2015;Swain *et al.*,2014).

3.2 Discussion

Heavy Metal Accumulation in Plants

Cadmium concentration was found to be highest in algae with 268.74 mg/dm^3. Their bioconcentration for the above metal followed similar pattern. In comparison with Yasar *et al*., (2013) the values are higher. This is an indication that algae is a good option for the decontamination of cadmium in aqueous medium. But in line with the trend of absorption Cadmium is second to the last, which is due to its size and smaller ionic radius.

As seen in Figure 3.2 and Table 3.1 algae shows greater potential to be used in Nickel decontamination but Nickel is an essential micro nutrient (Narain *et al*., 2010) . The result also revealed that, the plant root has higher affinity for Nickel ion than shoot. This could probably be as a result of some physiological barriers against metal transport.

Iron concentration in algae was (80.80 mg/kg) at 5 mg/l of the multi component metal solution which could be as a result of physiological activities such as photosynthesis (Victor *et al*., 2016). It also serve as an essential micro nutrient (Narain *et al*., 2010).

Lead concentration in the plant under investigation was quite high. Though in rooted plants accumulation in root could be attributed to avoidance of metal toxicity (Briat and Lebrun, 1999). It could also be as a result of greater surface area hence more adsorption capacity of the roots compared to the shoots (Irfan, 2015), on the other hand algae in 5 mg/dm^3 multi metal solution emerged the best accumulator. In comparison with Chimwieleska and Jad (2001) the values obtained here are a bit higher.

Chromium concentration in the plants under investigation recorded highest value in algae with (671.28 mg/kg), comparing the values in this work with those in Yasar *et al*., (2013) the values are very high. The values obtained in this work showed that algae is of paramount importance serving as a good candidate for decontamination of chromium in aqueous medium.

Manganese concentration in the plants was high and a noticeably large amount was recorded in control sample this could be as a result of its need in physiological activities because it is an essential micro nutrient for plants (Narain *et al*., 2010). Algae recorded highest concentration having (567.30 mg/kg), compared to results in Al Homaidan (2011) the values obtained here are within the range. The studied plant showed no heavy metals in control sample except manganese, iron and chromium, according to the findings in Table 3.1, heavy metal concentration increases with increasing concentration of the multi component metal solution.

Of the metals Chromium has the highest accumulation in the following decreasing order by Algae 5ppm, followed by the second most accumulated metal Lead in Algae 5ppm, followed by manganese with highest result in algae 5ppm,followed by iron in ,followed by cadmium in Algae

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5ppm followed by nickel which is the least with highest accumulation in Algae 5ppm.

In summary the total accumulating metals decreased in the following order Cr>Mn>Pb>Cd>Ni>Fe.

Under condition of adequate Mn supply the micro nutrients concentration is greatest in the chloroplasts and as a result, its accumulation is very high in the aerial parts. Studies done by Pedas et al., (2005); Kering et al., (2009) supported this observation. Also in the case of Fe, which is a micro nutrient, a considerably high amount has been recorded in control sample of the plants under investigation. Similar observation was reported by (Vardan and Ingole, 2006).

This could possibly be as a result of control by ferritin, or the plant has accumulated high amount already which is in accordance with findings by Miretzky, (2004). But judging from the small amount of iron left in the solution after the experiment the iron has precipitated out. This is what explains the presence of these metals (i.e. Mn and Fe) in the control sample and in high amount but as for chromium there could possibly be chromium pollution in the water ways since the plant's base is in a river that flows around inhabitation of people.

Since algae is a green plant, presence of Fe and Mn is not out of the ordinary. Fe is essential for the chlorophyll synthesis, ferritin which stores and releases Fe makes up about 75% of the content of chloroplast in leaf cell. Fe also participates in the electron transport in the process of reduction via cytochromes and ferredoxin (Marschner, 1995; Mengel and Kirkby, 2001; Malavolta, 2006), apart from that Cr, Mn and Fe have the smallest rating of size Table 1.3.

Following the trend in Table 1.3, lead accumulation in this order could be as a result of its size and also electronegativity, then Nickel is the least even though it has the highest electronegativity and a size rating as the last.

The result in water hyacinth and water lettuce (Table 3.1) showed higher concentration of heavy metals in the roots than in shoot which is as a result of metal distribution that is supported by certain factors such as genetic factors (Tarradellas *et al.*, 1996).

There was high absorption level of all metals which implies that all the plant under investigation can serve as good remediating tool. The plant showed its ability to accumulate more than one metal at a time which is an added advantage in phytoremediation.

IV. CONCLUSION

In this study, algae was used in order to assess its phytoremediative ability and it has proved to be a good accumulator of heavy metals serving as good biological filters of waste waters, which gives a good alternative for heavy metals removal from waste waters because it is cheaper and is environmentally friendly. Algae has a high result because it was able to accumulate large amount of the metals in use. The plant was able to accumulate more than one metal and has proved its phytoremediative ability towards decontaminating waste waters.

Recommendations

More work should be done in order to improve phytoremeditive capability of algae under investigation and its like, one of the ways is through exploring its genetic characteristics. Similarly, further work should be carried out using similar plants but different pH to ascertain the best pH for optimum absorption of metals. The analysis should also be extended to real waste water especially mining tails and tannery effluents.

Algae is easily obtainable some consider this plant as a serious invasive weed but it has found use in the field of phytoremediation.

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