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Phytoremediation of some Trace Metals in Polluted Water using Water Lilies

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ABSTRACT: This research work was aimed at using locally available plants (water lilies) to remove heavy metals in water samples from Woji Creek in Port Harcourt, Rivers State. The pH, temperature, dissolved oxygen (DO), total dissolved solid, oxidation reduction potential, and electrical conductivity of the water were assessed. The result of the physicochemical parameters showed that the mean value of the electrical conductivity (EC) was highest in June (46. 34±91. 5 µS/cm), temperature was high in June (27.9±1.0 °C), DO was highest in August (9.8±2.0 mg/l), pH had high value for July (7.4±0.7) and TDS for June (3548.0±1638.3 mg/l), which were lower than national and international permissible limit standard except for EC, DO and TDS. The uptake of cadmium by the roots of the water lily was very high for station 4 (0.7 mg/kg) and shoots for station 4 (2.1 mg/kg). The most uptakes for the Copper by the roots was for station 4 (0.9 mg/kg) and shoot for station 5 (1.3 mg/kg).The percentage of uptake of Cd for the root of the water lily was 19.3% while the shoots was 32.9% respectively. Cd had the highest accumulation of metals The water lily plants were significantly good for phytoremediation agents and as such could be employed by industries for such purposes.

Keywords: Water lilies, dissolved oxygen, electrical conductivity, heavy metals

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INTRODUCTION

Over the years there has been an increasing trend of contamination in land, groundwater and surface waters, arising from industrial and other anthropogenic activities. The build-up of toxic metals not only affects the natural resources but also poses a major problem to the ecosystem. The contamination of terrestrial and aquatic ecosystem by heavy metals has been a major global environmental problem. Land and water pollution by heavy metals is a worldwide issue. All countries have been affected, though the area and severity of pollution vary enormously. The mining and unsafe discharge of solid and liquid wastes by industries is considered to be the major source of heavy metals in the environment. In the urban areas the load of heavy metals in freshwater resources is at alarming level probably due to disposal of untreated or partially treated sewage and industrial wastewaters. Due to acute toxicity associated with heavy metals, these are considered as environmental priority pollutants and are targeted for cleanup processes.

The problem of soil and water contamination of toxic metals is becoming more and more serious with increasing industrialization and disturbance of natural biogeochemical cycles by human activities and climate change (Ali *et al.,* 2013). For example, in Europe, an estimated 52 million hectares of land more than 16% of the total land area of the continent are affected by some level of soil degradation (Peuke and Rennenberg, 2005).The field of phytoremediation is harnessing greater acceptance because phytoremediation technique can offer the only effective means of restoring hundreds and thousands of square kilometers of land area and water that have been polluted by irresponsible activities of humans (Meagher, 2000).Water lily otherwise known as *Nymphaeaceae* is a family of flowering plants which live as rhizomatous aquatic herbs in temperate and tropical climates around the world. The family contains five genera with about 70 known species (Christenhusz, 2016). Water lilies are rooted in soil in bodies of water, with leaves and flowers floating on or emergent from the surface (Wikipedia). Water lily has been reported by many researchers as a good remedy for cleaning up a metal polluted site. Presently, water and land pollution remain the major global problem, because it is the leading cause of deaths and diseases, as reported during the United Nations World Water Day released on March 22, 2010. Around 2.2 million people a year die from diarrheal diseases caused by drinking contaminated water and poor hygiene (Hunter *et al.,* 2010). About 97% of the world's water are saline (seawater), whereas freshwater represents only 3% of the total global water resources. However, only one-third of the freshwater is accessible for human activities due to the fact that the2% occurs as snow and ice in the polar and the alpine region of the world. Moreover, the most part of the freshwater (98%) is locked in the ground as „groundwater", with only about 2% of it easily available as surface water (rivers and lakes), for human consumption, agriculture and industrial activities. As a result, freshwater is seen as a finite and limited resource, especially in the arid regions (Christensen, 2013; Awang *et al.,* 2015).Currently, over 80% of the world population faces intricate water security problems. Nearly all countries in the world are affected by the water security threat of consuming water resources that are not safe through either endemic water diseases due to lack of proper water treatment capabilities and/or decreased in annual precipitation due to severe climatic change (Hanjra and Qureshi, 2010). Generally, the global water resources are polluted mainly through human activities (anthropogenic), because the industrial revolution contributed immensely to the global environmental degradation (Sayyed and Sayadi, 2011). Correspondingly, the natural water is also under severe stress as a result for the rising demand of freshwater caused by the increase in world population, urbanization and industrialization (Gleick and Palaniappan, 2010). It was estimated that the world population would increase to 92 billion at the end of this century and more than 80% of this population would live in the cities (DESA, 2009; Godfray *et al.,* 2012). These could lead to a remarkable growth of both urban and industrialized areas and the possibility of providing enough water for the growing population will be very challenging. The rapid growth in population coupled with the massive industrialization and agricultural activities have raised the water demand to a greater extent, even countries with sufficient quantities began considering sustainable water resource management to avoid water insecurity in the near future (Peasey *et al.,* 2000). At the moment, the demands for freshwater and world population growth are at the rate of 64 billion cubic meters and 80 million people per annum, respectively (Godfray *et al.,* 2012). However, the water demand and population growth increase annually at the rate of 12% and 1.8, correspondingly (Reed, 2015). Consequently, all these variables have direct or indirect impacts on the water problems as experienced by several developing countries. Therefore, improved awareness of harnessing water resources is a crucial component in addressing current world water security which is the only sustainable goal of living in the 21st century (Nature *et al.,* 2011).The discharge of domestic and industrial effluents into water bodies without adequate removal of the unwanted constituents' results in water pollution. The three major sources of river pollution are domestic sewage, agricultural and industrial effluents (Rafia Afroz *et al.,* 2014). Based on the Department of Environment (DOE) registration conducted in 2006, a total number of 18,956 water pollution point sources were identified. The data revealed that sewage treatment plants (47.79%) and manufacturing industries (45.07%) together accounted for more than 90% of the total number of water pollution sources. Meanwhile, animal farms and agro-based industries accounted for only 4.50% and 2.55%, respectively (Malaysian 1st Mathematics in Industry Study Group, 2011).Heavy metals are persistent and non-perishable in environment, which are from increased discharged of untreated or partially treated wastes of industries such as metal plating, mining activities, smelting, battery manufacture, tanneries, petroleum refining, paint manufacture, pesticides, pigment manufacture, printing and photographic industries (Wan Ngah and Hanafiah, 2008). According to Barakat (2011), heavy metals such as Cu and Cd are mostly released from chemical industries. The agricultural runoff, which is from pesticides and fertilizers, can also cause heavy metals contamination (Hou *et al.,* 2007; Megateli *et al.,* 2009). These heavy metals are transferred to aquatic environment through the food chain (Parlak *et al.,* 2013), and can be easily transported and accumulated in tissues, especially the living organisms (Barakat, 2011; Wan Ngah and Hanafiah, 2008).In the present, heavy metal pollution is a major environmental problem in the world. Whether these heavy metals occur in river, stream, pond or ditch, they affect human health. The local people who stays nearby the contaminated zone are affected directly (Miretzky *et al.,* 2004). It is reported that heavy metals can cause anemia, diseases of the liver and kidneys, brain damage and ultimately death. Their effects are not only on human but also on fauna, flora and ecological systems (Algarra *et al.,* 2005; Gavrilescu, 2004; Nagajyoti *et al.,* 2010). Even though at low concentration, it is still toxic on living things (Nagajyoti *et al.,* 2010). Therefore, heavy metals must be treated before discharge to the environment. Kamal *et al.,* (2004) and Ning *et al.,* (2011), stated that heavy metals cannot vanish easily and appropriate cleanup is usually required for their removal. Several technologies, chemical, physical and biological methods, have been widely used to remove heavy metals from environment, but these technologies are costly (Hou *et al.,* 2007). It‘s more expensive if these technologies are used for large volumes of contaminated water or soil with low metal concentration, and when high standards of cleaning are required (Sasmaz and Obek, 2012). In contrast, phytoremediation has been considered the cost effective and eco-friendly technology for heavy metals removal from environment such as soil, surfaced water including groundwater (Ha *et al.,* 2009; Hou *et al.,* 2007). Phytoremediation is a biological process that offers an attractive and economical alternative to detoxify the contaminated site. It is defined as the use of plants to remove pollutants from the environment or to render them harmless. Phyto-extraction is the name given to the process where plant roots take metal contaminants from the soil and transport them to their above ground tissues. A plant used for phytoremediation needs to be heavy-metal tolerant, grow rapidly with a high biomass yield per hectare, have high metal-accumulating ability in the aboveground parts, have a profuse root system and high bioaccumulation factor. Many researchers have found that Lemma minor and water lily plants can be used to remediate heavy metals polluted site. Water hyacinth (Echhornia crassipes) has shown the ability to accumulate many heavy metals according to (Agunbiade *et al.,* 2009), and CN-Ebel *et al.,* 2007).

MATERIALS AND METHOD

Study site

The study site is one of the several adjoining creeks of the Bonny River Estuary. It lies between longitude $7^0$3'N and $7^0$1'3''N and latitude4'48'E and 4'52'E of Port Harcourt. The creek has its head located at Rumuodara and flows unidirectionally downstream through Rumuodara swamp and traverses Port Harcourt-Aba express road. The water remains fresh and flows downstream in one direction until it reaches Mini Okoro Bridge at Rumuogba. It has a confluence with the refinery creek at Okujagu which forms the main tributary that drains into the Bonny River. Along the shores of the creek are located the Port Harcourt industries, markets, the main abattoir house and Port Harcourt zoological garden. It was observed that there was periodic accidental oil spill in the river brought about by illegal and ill-equipped vessels transporting crude oil from location of production to location of illegal refining. These have led to visible oil-sheen in the water and a visible high level of hydrocarbon deposit in the sediment. The study area is also the site for an abattoir where burning of wood, rubbers and cattle skin occurs (Figure 1). These activities in the abattoir produce a visibly high level of pollutant around the area.

Pre-sampling activity

Before embarking on the sampling journey, the different sample containers were thoroughly and properly washed, rinsed with distilled water and air dried. The containers were accurately labeled. Winkler's solution I & II, Mercury-in-glass thermometer, concentrated HNO_3 acid were all made available. A tour was made to the study area. Five sampling stations were identified based on the nature of activities going on along the river stretch and accessibility.

Materials collection

The main materials used for the study were water lily plants which were collected from a rain-fed pond in the Regional Centre for Bioresearch (University of Port Harcourt Botanical Garden in Rivers State, Nigeria). They were cultivated with Steinberg medium for 10 days (ISO 20079, 2005) under natural conditions. Water samples from the woji creek were then poured into a 100 ml plastic container, and 20 individuals of uniform size of water lilies were introduced per container. Pond water was pond into the same container for control. Three replicates (A, B, C) of the setup were prepared for each sampling point and control.

Sampling procedures

Polluted water samples from Woji creek were collected at five points. The samples were subjected to physicochemical analysis. Surface samples were collected approximately 20cm below the surface. The samples were then placed in sterilized plastic containers which were preserved in ice chest box before taking to the laboratory for further analysis. Samples for biochemical oxygen demand (BOD) and dissolved oxygen (DO) were collected in BOD glass bottles; care was taken to avoid bubbles being trapped before fixing the stopper and later taken to the laboratory for further analysis. Then, 150 g of water lily was then introduced to each container that is, into the DO and BOD bottles and appropriately labeled with its respective set-up and contact time (3, 6, and 9 days).

Metals analysis

The plants tissues (leaves and roots) were digested using the standard methods by APHA, (1998). Plant samples were decomposed to dry matter by heating at 120°C for 24 hours in a hot air oven and the ash were digested with nitric acid and hydrochloric acid then filtered into a volumetric flask. The final volume was made up with deionized water and heavy metals analysis was carried out using a Solar 969Unicam series atomic absorption spectrophotometer (AAS). The concentration of metals that remained in the residual solution was also measured using AAS. The differences between the initial metal concentration and remaining metal concentration in the

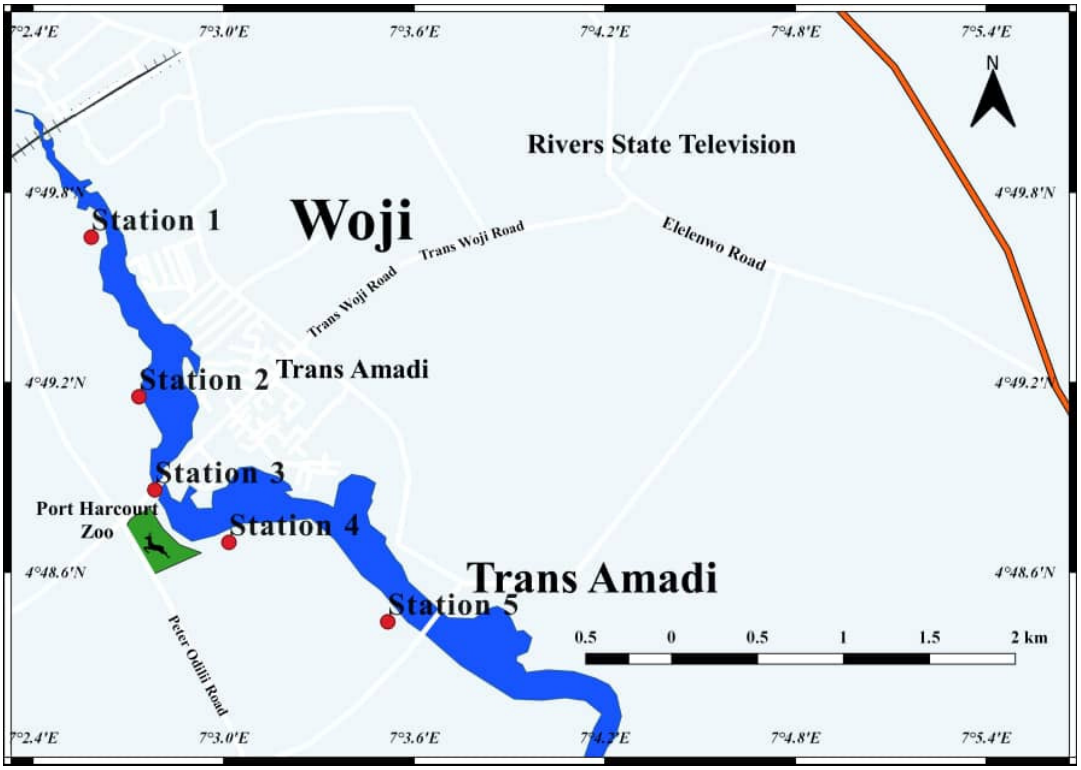

Figure 1: Map of the study area

solution were taken to be metals bound to the plant. The ability of the plant to accumulate metals with respect to the metal concentration in the substrate is known as the bio-concentration factor (BCF). Zayed *et al.* (1998) reported that BCF can be calculated as follows: BCF = Concentration of metal in plant tissue/Initial Concentration of metal in external solution.

RESULTS AND DISCUSSION

Results of the physiochemical parameters study on water samples from Woji creek is shown in (Table 1). The mean value of the electrical conductivity (EC) across the months showed higher value than the World Health Organization permissible limit of 400 µS/cm. However, the values obtained were higher than values reported in the streams, wells and bore-hole water in Nasarawa Eggon local government area of Nasarawa State, Nigeria by Aremu *et al.,*(2011). This may be due to differences in geochemical conditions and soluble ions in the locations analysed. This is an indication of the presence of dissolved chemicals in the water which hamper aquatic lives. It can be deduced that human activities within the area had great impact on the water quality. This shows that the water contains dissolved salts and other inorganic contaminants which could help to elevate its EC value. The temperature were all lower which means slight human effect, though the month of June was highest which might be from a lot of factors; like human activities, level of rain fall, global warming effect and others. High temperature affects the amount of oxygen in water as such affecting aquatic life. High temperature may also aid the easier dissolution of contaminants in water as such increases the level of contamination of such water. The values reported in this work are within the range recommended by WHO (30ºC) and National Guideline and standards for water quality (20 ºC-33 ºC) in Nigeria for aquatic life, industrial and agricultural uses (FME, 1992). Ehimeh *et al*., (2011) reported lower result (21.0 ± 0.1ºC) for rivers Inachalo and Niger in Idah, Kogi State, while Manilla and Frank (2009) and Clarke *et al.,* (2004) reported 25.3 ± 0.03 ºC. Although, there is seasonal fluctuation in creek water temperature values, this may be due to function of the climatic conditions at a particular geographical location and period. The DO showed high values for July (8.3±1.0 mg/l), August (9.8±2.0 mg/l) and September (11.2±2.2 mg/l) were all higher than the WHO standard of 7.5 mg/l. The findings from this result were also higher than reports by Oluyemi *et al* (2010) (4.48 to 9.48 mg/l) on levels of DO on water sources around Ife North Local government area of Osun state. DO is an important water quality parameter and is significance for the survival of aquatic lives (Willock *et al.,*1981). The high value of DO within July, August and September may due

Table 1: Mean values of the physicochemical parameters in water from the study area.

Period	EC	Temp.	DO	pH	ORP	TDS
June	46.34±91.5	27.9±1.0	6.1±1.2	6.8±1.5	0.1±0.0	3548.6±1638.3
July	3645.6±959.4	25.4±0.8	8.3±1.0	7.4±0.7	0.1±0.0	1760.0±440.4
August	1707.6±889.0	24.5±0.4	9.8±2.0	6.7±0.2	0.1±0.0	787..6±356.6
September	1211.8±676.1	23.2±0.4	11.2±2.2	6.9±0.2	0.1±0.0	505.2±131.4
WHO	1000	30	7.5	6.5-8.5	-	1000

Table 2: Uptake of metals (mg/kg) on phytoremediated parts of water lily plant and its concentrations in the surrounding waters

Metal	Root					Shoot					Water				
	Stations					Stations					Stations				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Cd	0.4	0.5	0.6	0.7	0.6	0.6	0.9	0.8	2.1	0.7	2.4	3.1	1.4	7.0	1.6
Cu	0.7	0.6	0.8	0.9	0.8	0.9	0.8	0.6	1.1	1.3	1.7	3.1	2.9	2.3	4.5

Table 3: Percentage uptake of metallic ions by water lily across the stations.

Metal	Root (%)					Shoot (%)				
	1	2	3	4	5	1	2	3	4	5
Cd	16.0	16.1	42.9	10.0	37.5	25.0	29.0	57.0	30.0	43.8
Cu	41.2	19.3	27.6	39.1	17.8	52.9	25.8	20.7	47.8	28.9

Table 4: Percentage Bio-concentration factor of the metal from the water lily.

Metal	Root (mean)	% BCF	Shoot (mean)	% BCF	% Total BCF
Cadmium	0.6±0.3	19.3	1.02±0.7	32.9	52.2
Copper	0.8±0.4	27.6	0.9±0.5	31.0	58.2

to intense disposal of toxic effluent in to the creek within this period of time. The pH values were more acidic except for the month of July with a basic value of 7.4±0.7 which were all within the WHO value of 6.5 to 8.5. These values were lower than reports by Oluyemi *et al.,* (2010), (8.16±0.38) on water sources within Ife north local government area of Osun State, Nigeria. It is clear though human activity may have affected the quality of water but the pH will likely favour aquatic life. The Oxidation reduction potentials were all relative low for the entire period of investigation. These care very low indicating that the creek lakes its ability to recreate itself, therefore indicating effect of anthropogenic activities. The TDS for August and September were lower that the WHO standard but June (3548.0±1638.3 mg/l) and July (1760.0±440.4 mg/l) were higher. These low and high values could probably be attributed to low and high human activities within these months. Reports by Alabaster and Lloyd, (198), suggest that excessive concentration of suspended and dissolved solid might be harmful to aquatic organism, because they decrease water quality, inhibit photosynthetic processes and eventually lead to increase in bottom sediment and decrease of water depth. The value of TDS may also have depended on differences in organic composition of the water bodies and as well the source of effluent discharge that gets into the water (Ogbeibu and Anagboso, 2004).

The result from the (Table 2) above showed that the contaminants absorbed both in the root and in the shoot of the water lily. In the different stations the rate of absorption differed slightly which could be as result of time, temperature, pH and changes in other variables. The uptake of cadmium by the root ranged from 0.4 to 0.7 mg/kg, with the highest uptake recorded for station 4 (0.7 mg/kg). The other stations had cadmium uptake at lower amount with the results in the order of station 4>5, 3>2>1. The shoot had more uptake of cadmium than the root with the range of values from 0.6 mg/kg to 2.1 mg/kg. The highest uptake for the shoot was in station 4 with a value of 2.1 mg/kg. this also shows that station 4 was more polluted than the rest of the stations. The order of uptake for the shoot was station 4>2>3>5>1 respectively. The result from the table above also showed the uptake of Cu by the water lily. The root had least uptake as compared with the shoot. The most uptakes for the Cu atom by the root was for station 4 (0.9 mg/kg). The other stations had Cu uptake lower than the said value. The shoot uptake was higher than that of the root. The highest was for station 5 (1.3 mg/kg) which was higher than the rest of the stations followed by stations 4, 1, 2 and 3 respectively.

Table 3 above indicate the percentage uptake by the water lily root and shoot across the stations. The percentage uptake of Cd by the root indicates that station 3 had the highest uptake with values at 42.9% while for Cu is station 1 with values at 41.2%. The shoot had the most uptake percentage for Cd in station 5 with its percentage at 43.8%, while the percentage uptake for Cu was highest for station 1 also with value of 52.8%.

The percentage of the bio-concentration factor shows that shoot was more effective in uptake than the root for both metals (Table 4). The percentage of uptake of Cd for the root of the water lily was 19.3% while the shoot was 32.9% respectively. These values for Cd were lower than that reported by Verma and Suthar (2014) on pH influence on removal of Cd in water using aquatic plants with values over 50 to 80 %. The percentage uptake of Cu by the shoot was also higher than the root. The overall accumulation of Cu by the water lily was 58.2 % which is lower than that of cadmium but is high enough for the water lily to serve as good phytoremediator. The result from this report indicates that the ability of water lily to accumulate Cd and Cu over time was influenced by various factors like time, concentration, pH, DO and TDS. It is pertinent to note that though the percentage removal of Cd by the water lily is high enough to serve a better alternative to the use of chemicals in remediation of metal polluted water. The research work was aimed at using locally available plants (water lilies) to remove heavy metals in water samples from Woji Creek in Port Harcourt, Rivers State and also investigate the water quality. This was achieved by checking the pH, temperature, Dissolved Oxygen (DO), Total Dissolved Solid, Oxidation reduction potential, and Electrical conductivity of the water and assessing the efficiency of *Lemna minor* and water lily as phytoremediation agent for cadmium and copper. The result of the physicochemical parameters showed mean value of the electrical conductivity (EC) was highest in June (4634.4±915.1 µS/cm), temperature was high in June (27.9±1.0 ^{o}C).The DO was highest in August (9.8±2.0 mg/l), pH had high value for July with a basic value of 7.4±0.7, TDS for June (3548.0±1638.3 mg/l) and July (1760.0±440.4 mg/l) were higher. The uptake of cadmium by the root of the water lily ranged from 0.4 to 0.7 mg/kg, with the highest uptake recorded for station 4 (0.7 mg/kg) and shoot had range of values from 0.6 mg/kg to 2.1 mg/kg, with station 4 (2.1 mg/kg) having highest uptake. The most uptakes for the Cu atom by the root was for station 4 (0.9 mg/kg) and shoot was highest for station 5 (1.3 mg/kg).The percentage of uptake of Cd for the root of the water lily was 19.3% while the shoot was 32.9% respectively. Overall accumulation of Cu by the water lily was 58.2 % which is lower than that of cadmium. The result indicated that Cd had the highest accumulation of metals in the duckweed on station 4 (3.0 mg/kg), and Cu was in station 5 (2.2 mg/kg).The amount of Cd was highest in the month of June (4.9±2.5 mg/kg), followed by September (4.2±0.4 mg/kg), July (3.1±0.5 mg/kg), August (2.4±0.3 mg/kg) respectively.

Conclusion

The result obtained from this very research work indicated that the physicochemical parameters of the Woji creek were within the World Health Organization standard limits for water bodies except for the Electrical conductivity and the Total dissolve solids which were high and could pose threat to the immediate users of such water. seed samples are good reservoirs of nutritional components and bioactive phytochemicals. The water lily a plants were significantly good phytoremediation agents and as such could be employed by industries for such purposes. The percentage of cadmium and copper removed were high as such it could be deduced that the plants have the capacity to act as phytoramediators

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