

Effectiveness of Bio-Phytoremediation on Heavy Metal Contaminated Wastewater using Vetiver Grass

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The high concentration of heavy metals in wastewater highlights the urgent need to explore alternative treatment methods. Partially treated wastewater with elevated heavy metal levels can have severe environmental consequences, ultimately affecting the food chain. This study evaluates the effectiveness of bio-phytoremediation in treating heavy metal-contaminated wastewater using perennial grasses. The research analyzed one-year average effluent results for Pb and Cd, comparing their removal efficiencies at an initial concentration of 10ppm after introducing Vetiver grass (*Chrysopogon zizanioides*) and Elephant grass (*Pennisetum purpurem*). The compliance levels of different remediation approaches were assessed against South African wastewater discharge limits and World Health Organization (WHO) guidelines. Various remediation methods were considered, with a particular focus on bio-phytoremediation using selected grass species to remove heavy metals from contaminated wastewater. The findings indicated that Vetiver grass demonstrated a higher removal efficiency for Pb compared to Cd.

1. Introduction

Rapid population growth necessitates a substantial supply of products and services to sustain livelihoods worldwide (Shang et al., 2019). The increasing demand for clean water has become a critical global issue, exacerbated by rapid industrial expansion (Al Sharabati et al., 2021). Routine human activities and industrial operations generate a range of organic and inorganic water pollutants that negatively impact aquatic ecosystems. The environmental crisis is already apparent, driven by unsustainable development, unchecked economic expansion, and a disregard for environmental sustainability (Vita et al., 2019).

Over the past ten years, significant environmental concerns have emerged regarding heavy metal contamination. Although the words "heavy metals" lacks a precise explanation, it therefore encompasses areas such as toxicological, biological, physical, and chemical aspects (Katheresan et al., 2018). It remains widely used in scientific discourse due to the absence of a suitable alternative. The presence of heavy metals in wastewater poses serious risks to ecosystems and human health, as they can accumulate in the food chain (Naseem et al., 2023).

Heavy metals are a major worry due to their ability to cause adverse impacts in humans even with extremely low concentrations (Levchuk et al., 2018). These adverse impacts include Cancer-causing potential, developmental toxicity, and genetic mutations, even with low-level or sub-chronic exposure. Proper disposal of heavy metal-containing waste is essential to prevent ecosystem contamination (Masinire et al., 2020). While some heavy metals (such as Cu, Fe, Mn, Zn, and Mo) play vital roles in biological growth at trace levels, excessive amounts can negatively impact development and progression (Raji et al., 2023). Naturally occurring as part of the Earth's composition and found in various ores, heavy metals are indestructible. Their xenobiotic concentrations pose significant environmental risks due to their toxicity, nonbiodegradable (El-Naggar et al., 2018).

Plant roots take in heavy metals within the soil, particularly in soil in which there's infection (Jordao et al., 2006). When those heavy metals are taken up through plant roots, it results in chlorosis, susceptible plant growth, yield reduction, decreased nutrient uptake, problems in plant metabolism and decreased cappotential to repair molecular nitrogen in leguminous flora (Guala et al., 2010). Uptake of those heavy metals through flora and

accumulation in meals chain will become a risk to animal and human health (Sprynskyy et al., 2007). High stage ingestion of poisonous metals has an unwanted impact on people which will become apparent simplest after numerous years of publicity to it (Khan et al., 2008). A lower in bacterial species richness and a relative growth in soil actinomycetes or maybe decreases in both the biomass and variety of the bacterial groups in infected soils are due to presence of heavy metals (Jordao et al., 2006).

The want to research sports of soil microorganisms in eco-structures which have been uncovered to long-time period infection through heavy metals has been suggested (Wang et al., 2007). The popular growth of heavy metallic content material within the soil has been in large part due to crude oil spillage (Anoliefo and Vwioko., 1995). Heavy metallic pollutants of the soil is due to numerous metals particularly copper, Nickel, Cadmium, Zinc, Chromium, and lead (Hinojosa et al., 2004). It has been found that the pollutants due to heavy metallic does now no longer have only consequences in destructive outcomes on numerous parameters regarding plant exceptional and yield but also reasons adjustments in the size, composition and microbial activities (Yao et al., 2003)

Toxicity of heavy metals at the increase and overall performance of plant life varies consistent with the heavy metallic concerned withinside the process. Heavy metals including Pb, Hg, As and Cd do now no longer have a useful position in plant increase, therefore, little or no awareness of those metals in increase medium may want to reason detrimental results to the plant. Some plant life now no longer most effective gather metals withinside the roots however additionally translocate metals from root to the leaves and shoots. For the ones metals which might be useful to plant life, small concentrations of the metals in soil can enhance plant increase and development; however, better concentrations can also additionally cause a discount of plant increase (Baker et al., 2000).

Manivasagaperumal et al., (2011) stated in his look at that zinc awareness of 25mg/l of soil answer stepped forward increase and body structure of cluster beans, because the awareness improved to 50mg/l of zinc increase discount and detrimental impact on plant body structure changed into observed. In a scenario wherein the surroundings is polluted with multiple heavy metallic including sewage sludge disposal, metallic mining waste, and crude oil drilling, there may be each synergistic and hostile relationships among the heavy metals which can also additionally have an effect on plant metallic toxicity.

Research has proven that a aggregate of Pb and Cu at each excessive awareness (1000mg/kg each) and coffee awareness (500mg/kg) resulted to a speedy and whole demise of the leaves and stem of *Lythrum salicaria* (Nicholls and Mal, 2003). Their end result found out no synergistic interplay among those heavy metals, in all likelihood due to the fact the concentrations used withinside the look at have been too excessive for an interactive dating to be observed among the metals. Ghani, (2010) in his paintings tested the impact of six heavy metals (Cd, Co, Mn, Cr, Pb and Hg) at the increase of maize on soil. The effects confirmed a discount withinside the increase and protein content material of maize. The toxicity of the heavy metals came about withinside the order of Cd>Co>Hg>Mn>Pb>Cr. This look at additionally confirmed that the blended impact of or extra heavy metals was as harmful as the effect of the most toxic heavy metal. This he attributed to be due to the antagonistic relationship which exists between heavy metals.

The method's attractiveness stems from its numerous benefits, including easy operation, simple design, and producing high-quality water (Katheresan et al., 2018) Due to the frequent uses and release of these HMs in most domestic and industrial activities and notorious environmental impacts this research discussion is centred more towards these the two (Iqbal et al., 2019; Khan et al., 2019). Methods through which the tolerance of perennial grass can be enhanced, with the pros and cons are also discussed. This research also iterates the knowledge related to the use of perennial grasses in research as the model vegetation in the field of phytoremediation.

In this context, selecting plants capable of absorbing heavy metals with minimal risk of biomagnification is highly beneficial. Therefore, Vetiver grass (*Chrysopogon zizanioides*) was chosen for this study. This research examines the effectiveness of perennial grass in the removal of heavy metals, specifically evaluating its response to Pb and Cd. Due to its high toxicity, accumulation potential, and persistence, Pb is considered one of the most hazardous environmental pollutants (Nazik et al., 2023; Jia et al., 2020). Major sources of Pb contamination in water include traffic emissions, mining activities, batteries, and a range of industrial operations (Ahmad et al., 2022; Jin et al., 2019). This research supports Sustainable Development Goals (SDG) 6, which targets to ensure the supply and sustainable control of water and sanitation for all.

2. Material and methods

Vetiver grass slips were supplied by Free Choice Progressive Learning (Pty) Ltd (White River, South Africa). while, Elephant grass was obtained from SMR Africa (Pty) Ltd in Bronkhorstspruit, South Africa. Lead nitrate $Pb(NO_3)_2$ was used to prepare the synthetical Pb solutions, while the Cadmium compound in the form of Cadmium nitrate ($Cd(NO_3)_2$) was used to prepare the Cd solution. This was because they are both soluble in

water. To determine the desired concentration of 10ppm, the following formulas were used to calculate the mass for both lead compound and Cadmium Salt respectively.

$$\text{Mass} = M \times V \times \text{Molar Mass.} \quad (1)$$

Where M is the molarity, V is the volume in litres, and the molar mass of $\text{Pb}(\text{NO}_3)_2$ is 331.2g/mol.

$$\text{Mass} = C \times M \times V \quad (2)$$

Where C is the concentration(mol/L) and M is the molar mass of salt $\text{Cd}(\text{NO}_3)_2$ which is 236.42g/mol and V is volume of solution (L).

2.1 Vetiver grass

Vetiver grass which was originally planted in soil was moved and introduced to water. To ensure that the roots remained intact with less if no damage to the roots, the roots were handled with care. Vetiver grass was washed and left in water for one weeks for acclimatization. To ensure the growth of roots and shoots to the desired length nutrients such as P, K, N and Ca were added into the water. The shoot length was adjusted to a height of 20 cm that is when acclimatization was observed. To start with the experiment Vetiver grass which was in the water was then transferred to 2 transparent buckets with 4Liters with 10ppm concentration of both Pb and Cd solutions. The decrease in water level in each bucket over time was accounted to evaporation and uptake by the vetiver grass. It was presumed that the evaporating water did not contain Pb and Cd, and water was topped to each bucket before sampling. This was to ensure that the change in concentration was not due to evaporation. Water samples were collected once every day for analysis for period of 10 days.



Figure 1: Vetiver grass in 10ppm Pb Solution



Figure 2: Vetiver grass in 10ppm Cd Solution

3. Laboratory Experiment

3.1 Pb Analysis

Water sample was collected using a 50mL syringe and put stored in a 100mL glass beaker. To prevent Pb from adhering to the wall of the glass beaker, 0.5mL of nitric acid HNO_3 was added to the sample to lower the pH to <2 . The sample was filtered using a $0.45\mu\text{m}$ filter just to separate dissolved Pb from suspended solids. The water sample was mixed with 0.1mL of hydrochloric acid (HCL) to break down the organic matter and release Pb. The mixture was gently heated in a micro wave until the solution became clear and then cooled and diluted with deionized water. Analysis was conducted using Atomic Absorption Spectroscopy (AAS).

Table 1: Pb results after when Vetiver grass water introduced for period of 10 days

Time/d	Pb Effluent at point Zero (mg/L)	Vetiver in Pb Solution(mg/L)	SA Wastewater limits for Pb (mg/L)	WHO Guidelines(mg/L)
Day 1	0.15	0.003	0.01	0.01
Day 2	0.15	0.003	0.01	0.01
Day 3	0.15	0.004	0.01	0.01
Day 4	0.15	0.006	0.01	0.01
Day 5	0.15	0.007	0.01	0.01
Day 6	0.15	0.007	0.01	0.01
Day 7	0.15	0.007	0.01	0.01
Day 8	0.15	0.009	0.01	0.01
Day 9	0.15	0.009	0.01	0.01
Day 10	0.15	0.009	0.01	0.01

3.2 Cd Analysis

Water sample was collected using a 50mL syringe and put stored in a 100mL glass beaker. To prevent Cd from adhering to the wall of the glass beaker, 0.5mL of HNO₃ was added to the sample to lower the pH to <2. The sample was filtered using a 0.45µm filter just to separate dissolved Pb from suspended solids. The water sample was mixed with 0.1mL of hydrochloric acid (HCL) and 0.1mL of hydrogen peroxide (H₂O₂) to break down the organic matter and release Cd. The mixture was gently heated in a microwave until the solution became clear and then cooled and diluted with deionized water. Analysis was conducted using Atomic Absorption Spectroscopy (AAS).

Table 2: Cd results after when Vetiver grass water introduced for period of 10 days

Time/d	Cd Effluent at point Zero (mg/L)	Vetiver in Cd Solution(mg/L)	SA Wastewater limits for Cd(mg/L)	WHO Guidelines(mg/L)
Day 1	0.2	0.002	0.01	0.03
Day 2	0.2	0.002	0.01	0.03
Day 3	0.2	0.005	0.01	0.03
Day 4	0.2	0.005	0.01	0.03
Day 5	0.2	0.007	0.01	0.03
Day 6	0.2	0.008	0.01	0.03
Day 7	0.2	0.008	0.01	0.03
Day 8	0.2	0.008	0.01	0.03
Day 9	0.2	0.008	0.01	0.03
Day 10	0.2	0.008	0.01	0.03

Pd and Cd were measured to assess the efficiency of Vetiver grass wastewater treatment. Calculating the removal efficiency of these parameters was made using the removal efficiency equation (Eq. 1):

$$\chi = \frac{C_0 - C_t}{C_0} \times 100. \quad (3)$$

where C₀ is the initial concentration of the parameter, C_t is the final concentration of the parameter at time t (de la Luz-Pedro et al., 2019). The results obtained are presented in table 1 and table 2 above.

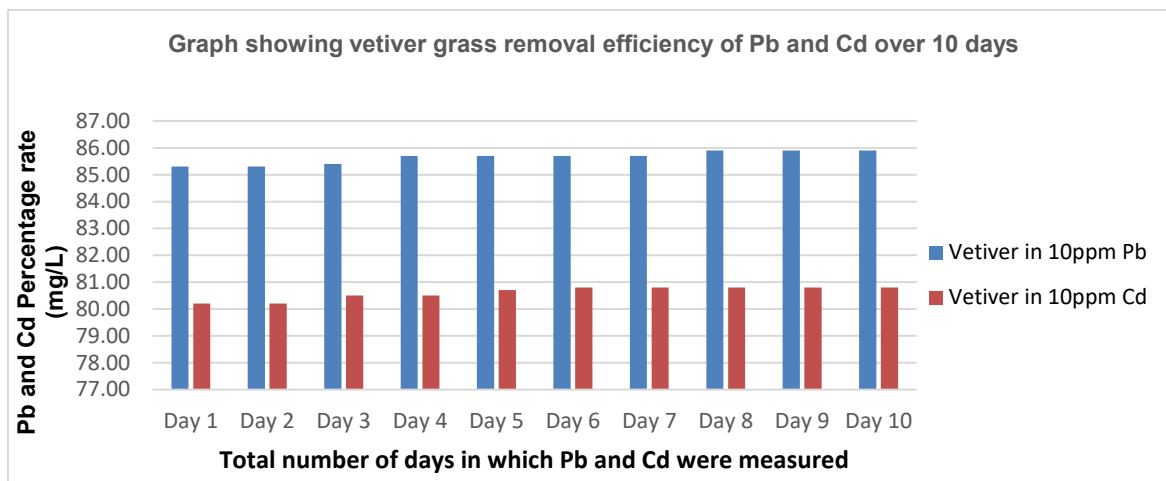


Figure 3. Shows the Vetiver grass removal efficiency of Pd and Cd over the period of 10 days.

4. Conclusion

The results indicated that Vetiver grass proved to have the most removal efficiency for both Pd and Cd and met both the South African wastewater discharge limits into the resource and the WHO guideline for the entire duration of the study. Removal efficiency for Pb was between 85.3% and 85.9% while that for Cd was between 80.2% and 80.8%. Overall, Vetiver grass had the highest removal efficiency on Pb as compared to Cd, even though the removal efficiency for both heavy metals was great. Further research to assess which parts (roots, shoots, and stems) of the vetiver grass were storing these heavy metals is recommended.

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