***Chrysopogon nitrigana* (VETIVER GRASS) and *Chymbopogon citratus* (LEMON GRASS) AS BIOSORBENTS FOR HEAVY METAL CONTAINING EFFLUENT: A COMPARATIVE STUDY.**

 **ABSTRACT**

The introduction of foreign materials such as the heavy metals into the environment is attributed to the major reason of environmental adulteration and this has been linked to increased industrialization. Plants have shown great potency in the biosorption of these toxic elements from the environment*. Cymbopogon citratus* (lemon grass) and *Chrysopogon nitrigana* (vetiver grass) both terrestrial plants have been used in the present study for the uptake of heavy metals from a heavy metal containing effluent. A 50% (TRT 1) and 25% (TRT 2) effluent concentrations was used to hydroponically culture the plants for the heavy metal bioaccumulation. *Cymbopogon citratus* significantly (P<0.05) reduced the concentrations of cadmium by 97% and 99% in both TRT 1 and 2. Lead by 66% and 57%, chromium 48% and 38% and copper 99% and 73% in both treatments. *Chrysopogon nitrigana* significantly (P<0.05) too up the heavy metals from both treatments at 94% and 98% for cadmium, 29% and 20% for lead, 20% and 12% for chromium and 73% and 79% for copper. The data obtained from this study revealed that both plants have the capacity to take up heavy metals however, *Cymbopogn citratus* showed a higher removal efficiency of all the four studied heavy metals than *Chrysopogon nitrigana*.

**Keywords: Vetiver grass, Lemon grass, Heavy metals, Bioremediation, Hydroponics, Greenhouse.**

**INTRODUCTION**

Heavy metals discharged form industrial effluents, domestic wastes, physical and chemical weathering of rocks, soil erosion, agricultural run offs and sewage release could substantially damage the natural aquatic bodies (Alloway and Ayres, 1993). Heavy metals have been studied epidemiologically showing that they make up for a significant proportion of pollutants via industrial waste (Khalifa and Alkhakf, 2018). Chemical pollutants (such as the heavy metals) are involved greatly in the pollution of the aquatic ecosystem and they originate from dangerous chemical using industries left as a derivative of production process (Bill, 2010; Maczulak, 2010; Laboratory Chemical Waste Management Guidelines., 2016). Arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), mercury, lead (Pb), zinc (Zn) and nickel (Ni) are the eight top usual heavy metals linked with pollution according to Environmental Protection Agency (Athar and Vohora, 2001). Furthermore, subjection to heavy metals such as mercury, lead, cadmium and arsenic is the major cause of hazard to human health (Zeitoun and Mehana, 2014). The containment of heavy metals in the environment does more harm to life than good and should be as a matter of urgency not let into the biota by any means. Currently, heavy metals are a set of pollutant causing the most highly rated chaos in the environment including both land and water altering the natural ecosystem and giving rise to geo-ecological variances universally (Liu *et al,* 2003; Chabukdara and Nema, 2012). Threat is brought to humans, animals, aquatic resources and plants by the presence of heavy metals.

The process of utilizing plants for the eradication of harmful pollutants such as the heavy metals from the environment (soil, water and sediments) either via in situ or ex situ approach is referred to as phytoremediation (Aisien *et al,* 2010). The elimination of heavy metals from different parts of the ecosystem using plants has developed into an exploratory and practicable perspective (Mojiri., 2012). Phytoremediation was first discovered in the 16th century by Andrea Cesalpino and reports have shown that plants were proposed to be used in the remediation of polluted waters more than three hundred years back (Hartman, 1975; Brooks, 1998). According to Gupta *et al* (2013), a number of plant species adapt to growing in habitats contaminated with heavy metals. Plants employ two approaches to shield the organelles from the perturbing effects of heavy metals and they include; keeping under control the heavy metal take-up and applying tolerance mechanisms (Nedjimi, 2021). Phytoremediation involves biological, chemical and physical actions of plants in the biosorption, bioaccumulation and bio-detoxification of toxic pollutants in the ecosystem (Cunningham and Berti, 1993). Because phytoremediation is an inherent process of decontamination of toxic heavy metals, it does not require the use of heavy machineries (Babu *et al,* 2021). Plants present discriminating prospects in the bioaccumulation of heavy metals via phytoremediation (Bhargava *et al,* 2012). In the last two decennia, quite a number of plants with phytoremediation potentials for both polluted water and soil have been recognized (Liao and Chang, 2004). Hyper accumulator plants have gained much interest in recent times as a result of its usefulness in remediating heavy metal pollution (Nedjimi, 2021). So far, the use of plant based remediation particularly for the hyper accumulation of heavy metal waste has shown significant results and quite a number of examination have been carried out (Zhu *et al,* 1999). According to Baker *et al* (2000), records have it that over four hundred plant species have exhibited hyper accumulation potentials. Some plants have been successfully used in phytoremediation (Zhu *et al,* 1999; Gardea-Torresdey *et al,* 2005; Aisien *et al,* 2010).

Vetiver grass is a tropical plant known to withstand exceedingly frigid climatic conditions (Dorafshan *et al*, 2023). Vetiver grass has been extensively proliferated all over in the tropical locality and also all around the world for a long period of time (Gnansounou *et al*, 2017). Recently, Vetiver grass (*Vetiveria zizanioides*) was reclassified as *C. zizanioides* and it belongs to the family gramineae and was initially applied in the preservation and maintenance of water and soil (Roongtanakiat *et al*, 2007) and can also, an important tool in balancing of slope and controlling of erosion (Paz-Alberto and Sigua, 2013). According to Dorafshan et al (2023) and Paz-Alberto and Sigua (2013), Vetiver grass has the ability to breed and withstand very severe ecological situations. Due to the morphological and physiological attributes of Vetiver grass and high biomass production, it is therefore, regarded as a more potent and suitable agent in bioremediation and phytoremediation processes (Boonsong and Chansiri, 2008; Leguizamo *et al*, 2017; Davamani *et al*, 2021). Vetiver grass has exhibited very significant resilience to high concentrations of toxic metals like arsenic, chromium, lead, copper, nickel, zinc, mercury and selenium (Truong and Baker, 1996; Danh *et al*, 2009). Recently, macrophytes that possess rhizosphere microorganism (such as the Vetiver grass) have gotten recognition owing to the notable bioremediation properties they possess (Worku *et al*, 2018) and now has a global acceptance in the remediation of both inorganic and organic contaminants (Panja *et al*, 2020). Vetiver grass is a terrestrial macrophytes that grows favorably around wet ecosystem and does very well when subjected to soil-less culture media (Xia et al, 2000; Boonsong and Chansiri, 2008) and has been successfully cultivated hydroponically for the treatment of different types wastewater (Roongtanakiat *et al*, 2007; Ho *et al*, 2013; Darajeh *et al*, 2019; Davamani *et al*, 2021).

Lemon grass which is scientifically known as *Cymbopogon citratus* stapf (*C. citratus*) is a perennial crop or herb belonging to the section of Andropogoneae called *Cymbopogon* of the family of poaceae/gramineae possessing a somewhat branched partly aerial rhizome (Vaqar *et al,* 2007). They are very fast-growing plants also known as plants of warm climatic conditions (Vaqar *et al,* 2007; Shamsheer *et al,* 2020). Lemon grass contains citral; this is a cyclic monterpene the gives rise to the lemon-like smell hence, the prefix “lem” (Manvitha and Bidya, 2014). Lemon grass is made up of about 500 genus approximately and 8,000 herbs with a life span of about 5 years (Manvitha and Bidya, 2014; Sobh *et* *al,* 2014). This tropical aromatic herb or grass have the ability to grow up to a height of six feet also, it possesses bulging stem and glabrous leaves (Shah *et al,* 2011). Subject to the habitat, lemon grass contains a collection of compounds such as flavonoids, alkaloids and terpenes (Babarinde *et al,* 2016). The biosorption of heavy metals (copper, nickel, zinc, cadmium and lead) have been studied using lemon grass biomass (Zou *et al,* 2012; Lee *et al,* 2014; Sobh *et al,* 2014; Babarinde *et al,* 2016). The metabolism of glutathione and organic acids play a very vital role in the tolerance of plants to metals in which organic acids detoxifies the metals by creating complexes with the existing metals (Prasad and Freitas, 2003). *Cymbopogon citratus* (lemon grass) have been successfully grown under hydroponic conditions (Mairapetyan and Tadevosyan, 1999).

**MATERIALS AND METHODS.**

The present research was conducted in the University of Abuja, FCT, Nigeria. Abuja is located at the Nigerian central with latitude 8o86’N to 8o95’N and longitude 7o18’E to 7o29’E with a population of about 1,406,239 (Nnodu *et al,* 2017).



 **Map 1: Map of Abuja, FCT with the six Area Councils (Obi-Anike *et al*, 2017).**

**Effluent** **Collection**

The collection of the effluent was done according to Nanda *et al* (2010) and Woldeamanuale and Hassen (2017). Untreated paint effluent was obtained from a paint industry located within Abuja, Nigeria. The preservation of the collected samples was done by storing the samples at the temperature of 4oC until use.

**Plant Collection and Preparation**

As described by Savitha and Rajan (2018), viable plants of the same size *Cymbopogon citratus* were collected from a neighborhood in Gwagwaglada, Abuja while *Chysopogon nigritana* was collected from the faculty of Agriculture, University of Abuja all within the FCT. The collected plants were allowed to acclimatize and established to their new environment for a period of time (Gupta *et al*, 2022).

**Acid Digestion of the Effluent and Heavy Metal Determination**

The method described by by Juliani *et al* (2021), was adopted for the digestion of the effluent sample and the heavy metal contents of the paint based industrial effluent in this study was analysed using the Atomic Absorption Spectroscopy (AAS) Hanna HI Variian AAS SpectraAA 240FS, Italy. A standard solution was run with certified reference material in line with (Zulkafflee *et al,* 2020).

**Experimental Setup**

The method illustrated by Kumar *et al* (2018), Wang *et al* (2018) and Savitha and Rajan (2018), for greenhouse effect with a slight modification was adopted in the present study. Adapted plants of *Cymbopogn citratus* and *Chrysopogon nitrigana* were relocated into a 5 litres size tub with two effluent concentrations (50% and 25%). The tubs were kept were the plants could get enough sunlight. Sample was withdrawn after a six week period (Echiegu *et al*, 2021) for the final heavy metal determination. Both treatment period and acclimatization time lasted for about 63 days.

**Determination of Removal Efficiency (%) of the Test Plants**

The evaluation of the removal efficiency was carried out using the following formula (APHA, 2012).

Removal Efficiency (%) = Ci - Ce x 100 …………………….. Equation I

 Ci

Where;

Ci = initial concentration of the pollutant.

Ce = final concentration of the pollutant.

**Determination of Relative Growth Rate (RGR) of the Test Plants**

The relative growth rate (RGR) was evaluated at the beginning (before exposure to the effluent) and the end of the plant exposure to the effluent. The weight of the plant was determined with the use of analytical weighing scale while the length of the leaves and root was taken with the use of measuring tape in inches before conversion to centimeters (cm) as described by (Kumar *et al,* 2018) and (Gupta *et al*, 2022).

The formula below was used to calculate the relative growth rate (Aron, 1949).

Relative Growth Rate (RGR) = Ln W2 – Ln W1  ………… Equation II

 T2 – T1

Where;

W1 = initial weight of fresh biomass

W2 = final weight of the harvested biomass

T1 and T2 = the duration of the experiment in days.

**Statistical Analysis**

All values described were the means of the replicates. Microsoft excel were used in the calculation of mean, standard error, standard deviation and in the plotting of statistical graphs. Statistical Package for the Social Sciences (SPSS) version 25 for Windows was used for the Descriptive statistics, Kolmogorov-Smirnov-Test, One-way Analysis of Variance (ANOVA) and Turkey Post Hoc Test.

 **RESULTS**

**Heavy Metal Uptake**

Two varying dilution factor of the effluent was used labelled TRT 1 (50% effluent concentration) and TRT 2 (25% effluent concentration). The uptake of four heavy metals (lead, cadmium, chromium and copper) was studied using *Cymbopogon citratus* and *Chrysopogon nitrigana*. In TRT 1, *Cymbopogon citratus* recorded the highest (P<0.05) uptake for all the four elements. *Cymbopogon citratus* significantly reduced the concentration of lead from 5.34 mg/L to 1.79 mg/L. Cadmium was reduced from 7.77 mg/L to 0.1 mg/L. chromium had initial and final concentrations of 0.20 mg/L and 0.13 mg/L and copper was reduced from 0.53 mg/L to 0.002 mg/L (figure 1). *Chrysopogon nitrigana* also significantly (P<0.05) reduced the concentrations of the heavy metals from the effluent. Lead was reduced to a final concentration of 3.79 mg/L, cadmium 0.13 mg/L, chromium 0.195 mg/L and copper 0.14 mg/L all by *Chrysopogon nitrigana* (figure 1).

**Figure 1. Heavy Metal Uptake by *Cymbopogon citratus* and *Chrysopogon nitrigana* in TRT 1.**

The second treatment (TRT 2) was also analysed for the heavy metal uptake by both plants. A similar pattern as obtained in TRT 1 was also observed in TRT 2 where the highest (P<0.05) heavy metal uptake was observed in the tub with *Cymbopogon citratus*. It reduced the concentration of lead from the initial concentration of 2.9 mg/L to 1.22 mg/L. Cadmium went from 3.96 mg/L to 0.12 mg/L, chromium (0.14 mg/L to 0.12 mg/L) and lastly, copper went down 0.03 mg/L from 0.27 mg/L. For *Chrysopogon nitrigana*, lead was reduced to 2.9 mg/L from 2.9 mg/L, cadmium, chromium and copper had the final concentrations of 0.24 mg/L, 0.12 mg/L and 0.03 mg/L (figure 2).

**Figure 2. Heavy Metal Uptake by *Cymbopogon citratus* and *Chrysopogon nitrigana* in TRT 2.**

**Removal Efficiency (%) of the Heavy Metals by the Plants.**

The removal efficiency for each plant on the four heavy metals was evaluated and *Cymbopogon citratus* had 66% and 57% lead removal from both TRT 1 and 2. 97% and 99% for cadmium, 48% and 38% for chromium while copper recorded 99% and 73% removal. The removal efficiency recorded for *Chrysopogon nitrigana* for lead in both treatments were 29% and 20%. Cadmium 94% and 98%. Chromium was at 20% and 12% while copper had a total removal upto 73% and 79% (figure 3).

**Figure 3. Removal Efficiency by *Cymbopogon citratus* and *Chrysopogon nitrigana* in TRT 1 and 2.**

**Relative Growth Rate (RGR) of the *Cymbopogon citratus* and *Chrysopogon nitrigana***

Both plants recorded reduced growth at the end of the study however, higher plant biomass reduction was observed in the TRT 2 pots. The TRT 1 pot had a more slight reduction. *Chrysopogon nitrigana* recorded a higher reduced growth rate compared to *Cymbopogon citratus* in this study (figure 3).

**Figure 4. RGR of *Cymbopogon citratus* and *Chrysopogon nitrigana* in the Effluent.**

**DISCUSSION**

The use of terrestrial plants in environmental cleanup has become a great and promising step in combating the menace of environmental pollution particularly that with heavy metals. Considering the hazardous effects of these elements, their eradication from the environment has become a matter of necessity and urgency. In this study, two terrestrial plants, *Cymbopogon citratus* and *Chrysopogon nitrigana* were studied to check for the plant with better heavy metal uptake from an effluent sample. In all the treatments (TRT 1 and 2) and for all the heavy metals (cadmium, lead, chromium and copper) analyzed, significant uptake has been observed by the two plants. However, *Cymbopogon citratus* gave the highest uptake for the four heavy metals when compared to its counterpart, *Chrysopogon nitrigana*. A phytoremedial approach was employed in an integrated industrial wastewater for the bioremoval of chromium, iron, copper, lead and zinc using *Cannabis indica* and the removal efficiency recorded ranged between 40%-100% (Gupta *et al*, 2022). Jha et al (2023) also studied the bioaccumulation of heavy metals via plant roots and recorded 99.86%, 87.09% and 70.25%. *Chrysopogon zizanoides* was used in a hydroponic system for the bioremoval of lead and cadmium from the medium and the removal efficiency reported for both metals were 65.63% and 64.29% respectively (Davamani *et al*, 2021). Chromium has also been reduced by 92% while lead, copper and cadmium were reduced by more than 60% in a hydroponic system (Rababah and Al-Shuka 2009). *Cymbopogon citratus* has been reported in previous report to have significantly brought low the levels of lead and cadmium in a system by 95% and 83% (Oguche et al, 2022).

In the present study, the maximum weight was recorded in *Cymbopogon citratus* at 1.35kg final weight and the maximum length of plant was observed in *Chyrsopogon nitrigana* at 120.65 cm even with the reduced weight of the biomass that was observed. According to Hassan et al (2020), there was a reduction in the length of vetiver grass with elevated levels of leachate; however, there was no significant change in the biomass. Studies have shown that the biosynthesis of chlorophyll has been altered due to the accumulation of heavy metals in vital parts of the plants and as a result, has hindered the process of photosynthesis and also, alteration of cellular division in the leaves (Igwe and Nwachukwu, 2016). On another view, there were a few green leaves left on some of the plants (*Cymbopogon citratus* and *Chrysopogon nitrigana*). There could be lack of obvious growth inhibition by different plants from cadmium bioaccumulation even at saturation level, a great menace on the security and safety of food and the environment (Akinola and Ekiyoyo, 2006; Dong *et al*, 2007; Igwe and Nwachukwu, 2016).

**CONCLUSION**

The present study aimed to investigate the potential of two terrestrial plants (*Cymbopogon citratus* and *Chrysopogon nitrigana*) that could be efficient in the uptake of heavy metals (lead, cadmium, chromium and copper) from a metal containing effluent. Cadmium in particular was reduced from an initial concentration of 7.77 mg/L to 0.1 mg/L (which is the permissible concentration limit given for effluent by the WHO) by both plants. Significant reductions were also observed for the other heavy metals. The results obtained proved that both plants can be used effectively for the purpose of phytoremediation with special reference *to Cymbopogon citratus* which showed a higher and more efficient uptake of all heavy metals that were studied.

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