Asian Journal of Biochemistry, Genetics and Molecular Biology

Volume 16, Issue 10, Page 1-9, 2024; Article no.AJBGMB.122581
ISSN: 2582-3698

Effects of Citric Acid on the Phytoextraction Potential of *Psium sativium* L Grown on Heavy Metals Contaminated Soil

Nkwocha John Nnaemeka [a]*, Nwauche K. T. [b] and Amadi B. A. [b]

[a] *Department of Biochemistry, University of Port Harcourt, Rivers State, Nigeria.*
[b] *Department of Biochemistry, Rivers State University, Port Harcourt, Rivers State, Nigeria.*

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/ajbgmb/2024/v16i10407

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/122581

Original Research Article

Received: 05/07/2024
Accepted: 07/09/2024
Published: 27/09/2024

ABSTRACT

This study assessed the effect on citric acid on the phytoextraction of *Psium sativium* on heavy metals. The experiment was an outdoor experiment which was done using soil samples obtained from auto-mechanic workshop. The soil sample was analysed for the presence of heavy metals and the presence of the following heavy metals were detected in this order of concentration: Zn>Cu>Fe>Pb>Cd>Cr>Hg. The four plastic cups labeled A, B, C and D were treated with the following concentrations of citric acids: 0, 0.5, 1.0 and 1.5g/kg of citric acid. The plants were allowed to grow for twenty (20) days before they were harvested. The harvested plants were air- dried. The

**Corresponding author: E-mail: johnnkwocha400@gmail.com;*

Cite as: *Nnaemeka, Nkwocha John, Nwauche K. T., and Amadi B. A. 2024. "Effects of Citric Acid on the Phytoextraction Potential of Psium Sativium L Grown on Heavy Metals Contaminated Soil". Asian Journal of Biochemistry, Genetics and Molecular Biology 16 (10):1-9. https://doi.org/10.9734/ajbgmb/2024/v16i10407.*

biomass and the length of the root and shoot of the dried plant sample were obtained by weighing with a chemical balance and measuring tape respectively. The heavy metal bioaccumulation of the root and shoot of *Psium sativium* obtained from plastic cup without citric acid was estimated and the following heavy metals were detected in the root and shoot: mercury, chromium, cadmium, zinc, copper, iron, and lead. There was an increase in the biomass and length of the dried root and shoot of *Psium sativium* that were planted on soil sample treated with 0.5 g/kg and 1.0 g/kg citric acid but an increase in the concentration of citric acid to 1.5 g/kg decreased the biomass and length of root and shoot when compared with *Psium sativium* that was planted on soil sample that was not treated with citric acid. The application of citric acid increased uptake of chromium, cadmium, zinc, copper, iron, and lead by the root and shoot of *Psium sativium* in this order of increasing citric acid concentration: 1.5 g/kg> 1.0 g/kg> 0.5 g/kg while 1.5 g/kg concentration of citric acid reduced the heavy metal uptake by the root and the shoot. The bioconcentration factor for all the heavy metals that were analyzed increased with an increase in citric acid concentration except for mercury that had a reduced bioconcentration factor at citric acid concentration of 1.5g/kg. The traslocation factor of *Psium sativium* for copper, zinc and iron increased with an increase in citric acid concentration. *Psium sativium* had the least translocation factor for cadmium and highest translocation factor for chromium at varying concentration of citric acids when compared with other heavy metals. The result of this study shows that *Psium sativium* is an effective phytoremediation agent of some heavy metals. The study also shows that the application of citric acid could be a useful agent in increasing the phytoextraction of heavy metals from contaminated soil by *Psium sativium* plants.

Keywords: Phytoextraction; Psium sativium bioaccumulation; heavy metal; citric acid; phyroremediation.

1. INTRODUCTION

Bioremediation is a process whereby organic wastes are biologically degraded under regulated conditions to levels below concentration limits established by regulatory authorities. The use of plants to carry out bioremediation by is known as phytoremediation [1]. Environmental contamination can occur by several inorganic and organic pollutants such as heavy metals, petroleum products, explosives and hazardous waste materials [2].

Heavy metals are defined as metals with specific density which is more than 5g/cm^3. Heavy metals are non-biodegradable and have long biological half-life and accumulate in different organs of the body which results to health challenges [3-6].

Heavy metals are considered as serious contaminants in the ecosystem due to their non-biodegradable nature and toxicity in little concentration [7].

The accumulation of heavy metals in humans results to adverse health conditions. Some of the health threatening toxic heavy metals which are usually encountered are aluminium (Al), copper (Cu), mercury(Hg), nickel (Ni), iron(Fe), chromium(Cr), zinc(Zn), arsenic (As), Lead (Pb), and Cadmium (Cd). The processes through which the heavy metals find their way into the body include ingestion, absorption through the skin and inhalation [8]. The problem of heavy metals exposure has become a global challenge. Exposure to non- toxic concentration of heavy metals can result to health challenge because the heavy metals have the ability to interact with biological processes in the cell [9]. The use of plants and micro-organisms have been shown to be effective in reducing the exposure level as well as the environmental contamination of heavy metals [10]. There were three main classes of plants based on their heavy metal absorption capacities [11]. These are low heavy metal accumulators, medium heavy metals accumulators and high heavy metals accumulators. The plants that belong to the family *Brassicaceae* are the most heavy metals bioaccumulators [12]. Many plants mainly the plants that belong to the *Cucurbitaceae* family can bioaccumulate significant quantity of heavy metals from contaminated soil. Study carried out on summer squash showed that it is a potential agent of phytoremediation for the removal of cobalt, chromium, lead and nickel from polluted soil [13] and the following heavy metals: cobalt, chromium, lead, zinc and nickel could be accumulated into the leaves and root [14]. The poor bioavailability and mobility of heavy metals within the rhizosphere hinders phytoextraction that occurs naturally [15]. The ability of heavy metals to dissolve in the soil is greatly influenced by PH [16]. This has led to the use of chemicals to facilitate heavy metal solubility and bioavailability [15]. Citric acid is one of the chemicals used as chelating agents to enhance phytoextraction of heavy metals [17].

Psium sativium L. (pea) is one of the world`s oldest cultivated legume. It has been used in several genetic research and it has undergone numerous selections and breeding for characteristics desirable for cultivation, consumption and other uses. *Psium sativium* has a tap root but most of the secondary roots are within the top soil layers [18]. It is among the most efficient nitrogen fixation crops. *Psium sativium* survives in heavy metals contaminated soil which shows its bioremediation potential, hence its selection for this study.

The objective of this study is to ascertain the phytoextraction potential of *Psium sativium* in a soil sample that is contaminated with multi-heavy metals along with the help of citric acid as a phytoextraction enhancer.

2. MATERIALS AND METHODS

Study Area: This research was an out-door experiment which was done between July and august 2023 at Elelenwo, Port Harcourt, Rivers state, Nigeria.

2.1 Sample Collection and Preparation

The soil that was used for this study was collected from the top layer (0-15 cm) at auto-mechanic workshop in Elelenwo, Port Harcourt, Rivers State, Nigeria. The soil was exposed to air for air-drying. The heavy metal composition of the soil was determined using the atomic spectrometry method.

Psium sativium seeds were obtained from University of Agriculture Umudike Research Institute, Umudike, Abia State, Nigeria. *Psium sativium*. The seeds were sown in small plastic cups filled with soil sample obtained from a mechanic workshop. The plastic cups were labeled A, B, C, and D and the following concentrations of citric acid: 0, 0.5, 1.0 and 1.5 g/kg were introduced into A, B, C, and D respectively. The plants were allowed to grow for 20 days, after which, they were harvested. The roots were treated with deionized water. The roots were excised, blotted with filter paper and then weighed with a chemical balance. The samples were subjected to drying with an oven at a temperature of 85◦C. The digestion of the samples was done with the use of sulphuric acid and hydrogen peroxide. The concentrations of the heavy metals in the soil and plant samples were analyzed at the University of Agriculture, Umudike Research Institute using atomic absorption spectrophotometer by adopting the method described by [12].

Bioconcentration Factor (BCF) = metal content in mg/g dry plant tissue / metal content in mg/g soil sample

Translocation Factor (TF): This was determined using the formula according to Anamika, et al., 2009; Liao and Chang, 2004 maize citric acid).

Translocation Factor (TC) = Shoot metal content in mg/g / Root metal content in mg/g

2.2 Statistical Analysis

The data were analyzed for statistical differences between treatment groups by means of One Way Analysis Of Variance (ANOVA). In all, $p<0.05$ was considered significant. Data are presented as mean ± standard deviation.

3. RESULTS

Treatment of the soil sample with different concentrations of citric acid increased the biomass of the root and shoot of *Psium sativium* at citric acid concentration of 0.5 and 1.0 mg/g compared to the non-citric acid treated soil but an increase in the concentration of the citric acid to 1.5 g/kg reduced the biomass of the root and shoot.

Treatment of the soil sample with different concentrations of citric acid increased the lengths of the root and shoot of *Psium sativium* at citric acid concentration of 0.5 g/kg and 1.0 g/kg compared to the non-citric acid treated soil but an increase in the concentration of the citric acid to 1.5 g/kg reduced the lengths of the root and shoot.

Bioaccumulation of heavy metals at the root and shoot increased with an increasing citric acid concentration. The concentration of the heavy metal in the root and shoot increased in plants cultivated in soil sample treated with 0.5 and 1.0 g/kg concentration of citric acid but an increase in the citric acid concentration of the soil to 1.5 g/kg reduced the bioaccumulation of mercury in the root and shoot of *Psium sativium.*

Table 1. Concentration of heavy metals in soil sample before planting and after harvesting of *Psium sativium* plant. Result showed that before planting, zinc had the highest concentration followed by copper while lead had the least concentration. After harvesting, the heavy metal with the highest concentration was zinc while mercury had the least concentration

Soil sample	Hg (mg/g)	Cr(mg/g)	Cd (mg/g)	Fe(mg/g/)	Pb(mg/g)	Zn (mg/g)	Cu (mg/g)
Before planting	2.98± 0.02[a]	3.14 ± 0.21[a]	3.45 ±0.01[a]	4.21± 0.11[a]	3.98 ±0.12[a]	5.05 ± 0.03[a]	4.38 ± 0.01[a]
After harvesting	0.69± 0.13[b]	1.03 ± 0.04[b]	0.32 ± 0.11[b]	0.89± 0.04[b]	3.50 ± 0.05 [a]	4.85± 0.01 [a]	4.12 ± 0.08[b]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same column*

LEGEND: Hg = Mercury, Cr = Chromium, Cd = Cadmuim, Pb = Lead, Zn = Zinc, Cu = Copper

Table 2. Effect of treatment of different concentrations of citric acid on the biomass of root and shoot of *Psium sativium*

Mass of Dry Plant plant(g)	0.0 g/kg citric acid	0.5 g/kg citric acid	1.0 g/kg citric acid	1.5 g/kg citric acid
Root (g)	3.54 ± 0.11 [a]	3.67 ± 0.07 [a]	4.89 ± 0.02 [b]	3.02± 0.01 [a]
Shoot(g)	16.12 ± 0.08 [a]	26.88 ± 0.03[b]	30.12 ± 0.04[b]	20.68 ± 0.11[a]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same row*

LEGEND: g/kg =gram per kilogram, g = gram

Table 3. Effect of treatment with different concentrations of citric acid on the lengths of the root and shoot of *Psium sativium*.

Length of dry plant part(cm)	0.0 g/kg citric acid	0.5 g/kg citric acid	1.0 g/kg citric acid	1.5 g/kg citric acid
Root (cm)	4.50 ±0.21[a]	5.67 ±0.31[b]	7.89 ±0.11[c]	4.32 ±0.22[a]
Shoot(cm)	21.97 ±0.04[a]	28.88 ±0.22[b]	30.12 ±0.11[b]	24.68 ±0.22[a]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same row*

LEGEND: g/kg = gram per kilogram, cm= centimeter

Table 4. Effect of different concentrations of citric acid on the bioaccumulation of heavy metals by the root and shoot of *Psium sativium*

Plant parts	Citric acid concentration g/kg	Hg mg/g	Cr mg/g	Cd mg/g	Fe mg/g	Pb mg/g	Zn mg/g	Cu mg/g
Root	0.0	0.98± 0.02[a]	0.89 ±0.21[a]	2.08 ±0.08[a]	1.08±0.04[a]	1.04±0.04[a]	1.09±0.06[a]	1.07±0.06[a]
Shoot	0.0	1.64±0.22[ab]	1.66±0.01[ab]	1.03±0.06[ab]	1.45±0.01[ab]	1.23±0.43[ab]	1.35±0.01[ab]	1.33±0.01[ab]
Root	0.5	1.06± 0.01[a]	0.97 ±0.08[b]	2.28± 0.07[b]	1.17± 0.61[b]	1.27 ±0.04[c]	1.18± 0.61[b]	1.15± 0.61[b]
Shoot	0.5	1.72±0.05[ac]	1.71±0.10[ab]	1.21±0.03[ac]	1.62±0.01[ac]	1.42±0.03[ac]	1.64±0.02[ac]	1.60±0.01[ac]
Root	1.0	1.23 ±0.06[b]	1.03 ±0.11[b]	2.43 ±0.01[c]	1.21 ±0.04[b]	1.38 ±0.02[c]	1.23 ±0.04[b]	1.21 ±0.04[b]
Shoot	1.0	1.98±0.01[ad]	1.85±0.01[ac]	1.30±0.11[ad]	1.89±0.06[ad]	1.66±0.02[ad]	1.90±0.07[ad]	1.86±0.06[ad]
Root	1.5	0.86 ±0.04[a]	1.22 ±0.12[c]	2.49 ±0.12[d]	1.28 ±0.08[c]	1.40 ±0.07[c]	1.26 ±0.07[c]	1.22 ±0.08[c]
Shoot	1.5	1.68±0.03[ab]	1.88±0.01[ac]	1.3;5±0.05[ad]	2.24±0.05[ae]	2.28±0.04[ab]	2.22±0.05[ae]	2.20±0.05[ae]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same column*

LEGEND: Hg= Mercury, Cr= Chromium, Cd= Cadmuim, Pb= Lead, Zn= Zinc, Cu= Copper

Table 5. Effect of different concentrations of citric acid on the bioconcentration factor (BCF) of *Psium sativium*

CITRIC ACID g/kg	Hg	Cr	Cd	Fe	Pb	Zn	Cu
0.0	0.89± 0.01[a]	0.81 ±0.11[a]	0.9 ±0.03[a]	0.60±0.01[a]	0.57±0.02[a]	0.48 ±0.04[a]	0.55±0.03[a]
0.5	0.93± 0.03[a]	0.85 ±0.02[b]	1.01± 0.02[b]	0.66± 0.11[a]	0.68 ±0.05[b]	0.55± 0.11[b]	0.62± 0.31[b]
1.0	1.08 ±0.11[b]	0.92 ±0.21[b]	1.08 ±0.01[c]	0.74 ±0.02[b]	0.76 ±0.02[c]	0.62±0.01[c]	0.70 ±0.04[c]
1.5	0.85 ±0.04[a]	0.98 ±0.11[c]	1.11 ±0.11[d]	0.84±0.05[c]	0.92 ±0.06[d]	0.68 ±0.05[c]	0.78 ±0.02[c]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same column*

LEGEND: g/kg = gram per kilogram

Table 6. Effect of different concentrations of citric acid on the translocation factor (TF) of *Psium sativium*

CITRIC ACID g/kg	Hg	Cr	Cd	Fe	Pb	Zn	Cu
0.0	1.67± 0.02[a]	1.86 ±0.13[a]	0.49 ±0.13[a]	1.34±0.02[a]	1.18±0.03[a]	1.24±0.03[a]	1.24±0.02[a]
0.5	1.62± 0.05[a]	1.76 ±0.02[b]	0.53± 0.02[b]	1.38± 0.21[a]	1.12 ±0.05[b]	1.38± 0.01[b]	1.39± 0.11[b]
1.0	1.61 ±0.21[a]	1.80 ±0.11[b]	0.53±0.02[b]	1.56 ±0.12[b]	1.20 ±0.05[a]	1.55±0.11[c]	1.54±0.02[c]
1.5	1.95 ±0.08[a]	2. 04 ±0.21[c]	0.54 ±0.01[b]	1.75±0.04[c]	0.90 ±0.06[c]	1.76 ±0.15[d]	1.80 ±0.02[d]

**Values are shown in means of triplicate analysis ± Standard Deviation. Values bearing different alphabetical superscript are significantly different (p<0.05) from each other down the same row*

LEGEND: g/kg =gram per kilogram

The bioconcentration factor for all the heavy metals that were analyzed increased significantly with an increase in citric acid concentration except for mercury that had a reduced bioconcentration factor at citric acid concentration of 1.5 g/kg. *Psium sativium* had the highest Bioconcentration factor (BCF) for cadmium and least translocation factor for zinc at varying concentration of citric acids when compared with other heavy metals.

The traslocation factor of *Psium sativium* for copper, zinc and iron increased with an increase in citric acid concentration. *Psium sativium* had the least translocation factor for cadmium and highest translocation factor for chromium at varying concentration of citric acids when compared with other heavy metals.

4. DISCUSSION

The result obtained from atomic absorption spectrometry experiments showed the presence of heavy metals in the contaminated soil sample (obtained from auto-mechanic workshop) in variable concentration in the order as shown : Zn>Cu>Fe>Pb>Cd>Cr>Hg. Auto-mechanic workshop is usually polluted with engine oil as well as metal scraps which is rich in heavy metals and hence make the mechanic workshop site to be rich with these heavy metals [19].

Treatment of the soil sample with different concentrations of citric acid increased the biomass of the root and shoot of the plant samples at citric acid concentration of 0.5 g/kg and 1.0 g/kg compared to the non-citric acid treated soil but an increase in the concentration of the citric acid to 1.5 g/kg reduced the biomass of the root and shoot. This is in agreement with the report of Mohammed et al., [20] who also reported an increase in root dry weight at citric acid concentration of 2.5 and 5mM. Low concentration of citric acid increased plant biomass by mobilizing the slightly soluble plant vital nutrients [21]. While 0.5 g/kg and 1.0 g/kg concentration of citric acid increased the root and shoot biomass production whereas 1.5 g/ kg of citric acid concentration decreased root and shoot biomass, there may be critical concentration between 1.0 g/kg and 1.5g/kg which initiates suppression of root and shoot biomass production. This critical concentration can be obtained through graphical or statistical method.

Treatment of the soil sample with different concentrations of citric acid increased the length of the root and shoot of the plant samples at citric acid concentration of 0.5 g/kg, and 1.0 g/kg when compared with the plants from non- citric acid treated soil and increase in the concentration of the citric acid to 1.5 g/kg reduced the length of the root and shoot.

Bioaccumulation of heavy metals at the root and shoot increased with an increasing citric acid concentration. The bio-concentration of lead, iron cadmium, zinc, copper and chromium in the root and shoot increased in plants cultivated in soil sample with citric acid in this order of citric acid concentration: 1.5 g/kg > 1.0 g/kg > 0.5 g/kg. This demonstrates the effectiveness of citric acid in increasing the bioremediation of metals under conditions of heavy metals contaminated soil [22]. Moreover, citric acid can decrease the phytotoxicity of heavy metals by increasing the activities of antioxidant enzymes [22]. This could be due to the ability of citric acid to convert toxic metals into forms that are less toxic. Studies have also shown that citric acid enhances the soil nutrient availability and uptake by plants by the formation of complexes with nutrients especially in calcareous soils [23]. The growth - promoting role of citric acid under conditions of heavy metals contaminated soil is due to the ability of citric acid to improve chlorophyll content and photosynthesis efficiency in plant leaves [24,25]. Moreover, the chelating ability of citric acid helps in the transfer of these metals from the root to the aerial plant parts [6]. Ma, [26] also reported that citric acid detoxifies salinity stress by chelation by plant root. Citric acid reduces the toxicity level of manganese, cadmium and lead [27]. The heavy metals were more concentrated in the shoot than in the root. This trend is common in most plants due to the ability of the plants to take metal - contaminated fluids and nutrients [28]. An increase in the citric acid concentration of the soil to 1.5 g/kg reduced the bioaccumulation of mercury in the root and shoo*t of Psium sativium*.

The Bioconcentration factor (BCF) and translocation factor (TF) were estimated to check whether the plants were suitable for phytoremediation. Bioconcentration factor (BCF) expresses the capacity of plant to take up metals to the root. Translocation factor expresses the capacity of the plant to transfer those metals from the root to the shoot of the plant [29]. The highest bioaccumulation factor among heavy metals that were analyzed was cadmium, while zinc had the lowest bioconcentration factor (BCF)value. A low BCF shows that the metals cannot be readily absorbed by plants while a high BCF value shows that the metals are both relatively poorly retained by the soil and more mobile in the soil and therefore effectively absorbed by plant root. The relatively decreased transfer of zinc from soil to plant root shown by lower BCF value is in agreement with the result of Hasan et al., [30] and Acuna et al., [31] The high BCF value for cadmium is in agreement with the findings of Hedavatzadeh et al., [32] and Bortoloti and Baron [12]. This shows that the root of *Psium sativium* is more effective in the extraction of cadmium. Chromium had the highest Translocation factor while cadmium had the least translocation factor. The low concentration of cadmium in the shoot shows that *Psium sativium* has cadmium tolerance level. This is in agreement with the findings of Jadia and Fuleka [33], Xiao, et al. [34].

From the findings of this research, all the heavy metals except cadmium had high transcloaction factor values for all treatments. This indicates that large amount of the heavy metal was transported from the root to the shoot tissues. This result is in agreement with the previous findings [32]. The low traslocation factor value of cadmium may be due to the ability of the metal bioaccumulator plants for the extraction of metals from the soil occurring up to a certain level of concentration, hence an increase in the amount of the metal leads to an increase in the phytoextraction of the metal or an increase in the bioaccumulation coefficient of the plant.

The findings of this research showed that *Psium sativium* is an effective phytoremediator of heavy metals from the soils. The phytoremediating capacity of *Psium savitium* is enhanced by the application of citric acid. The contamination of plants occur through phytoremediation hence making the plants to be toxic. Therefore phytoremediating plants need to be treated to prevent further damage to the environment and living things in the environment.

5. CONCLUSION

The current result shows that *Psium sativium* is very effective in the extraction of Fe, Pb, Cd, Cr, and Hg from heavy metal contaminated soil. The result also shows that the application of citric acid to heavy metal contaminated soil enhances the heavy metal bioextraction by *Psium sativium* and also reduces heavy metal toxicity and an increase in biomass production. The phytoextraction potential of *Psium sativium* is significant in the bioremediation process more importantly as a low cost technology in cleaning contaminated environment. Further studies need to be carried out over longer period of time and under different environmental conditions and in real field condition. There should be further studies to determine factors that enhance plant phytoremediation process such as changes in soil PH, concentration of heavy metals and addition of other chelating agents.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

The authors hereby declare that no generative AI technologies such as large language model (ChatGPT, META AI, etc) and text -to- image generators have been used during writing and editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Hussein F, Farrag, W, Yasin, M, Al-Sodany, M, Faleh, G. Phytoremediation and accumulation characteristics of heavy metals by some plants in Wadi Alargy-Wetland, Taif-KSA. World Applied Sciences Journal. 2013;28(5):644-653.
2. Wuana RA, Okieimen FE. Heavy metals in contaminated soils: A Review of Sources, Chemistry, Risk. Journal of Environmental Science. 2011;3:12-16.
3. Ikechukwu UR, Okpashi VE, Nduka FO, Ogbonnaya P. Evaluation of heavy metals in selected fruits in Umuahia Market, Nigeria: Associating Toxicity to Effect for Improved Metal Risk Assessment. Journal of Applied Biology and Biotechnology. 2019;7(4):39-45.
4. Rascio N, Navari F. Heavy metals hyperacummulating plants: How and Why do they do it? And what makes them so interesting? Plant Science. 2011;180:56-87.
5. Rostami S, Azhdarpoor A. The application of plant growth regulatorsto improve phytoremediation of contaminated soil: A review. Chemosphere. 2019;220:818-827.
6. Sobukola OP, Adeniran OM, Odedairo A, Kajihausa OE. Heavy metals from fruits and leafy vegetables from selected markets in Lagos, Nigeria. African Journal of Food Science. 2010;4(2):389-393.

7. Jan AT, Azam M, Siddiqui K, Ali A, Choi I, Haq QMR. Heavy metals and human health: Mechanistic insight into toxicity and counter defense system of antioxidants. International Journal of Molecular Sciences. 2015;16:29592–29630. Available:http://dx.doi.org/10.3390/ijms161226183
8. Tchounwou PB, Yedjou CG, Patlolla AK, Sutton DJ. Heavy metals toxicity and the environment. EXS. 2012;101:133–164. Available:http://dx.doi.org/10.1007/978-3-7643- 8340-4_6
9. Valko M, Morris H, MTD C. Metals, Toxicity and Oxidative Stress. Current Medicinal Chemistry. 2005;12:1161-1208. Available:https://doi.org/10.2174/0929867053764635
10. Gupta N, Gaurav SS, Kumar A. Molecular basis of aluminium toxicity in plants: A review. American Journal of Plant Science. 2013;4:21–37.
11. Kobi T, Noguchi A, Yazaki J. Family dependent cadmium accumulation characteristics in Higher Plants. Plant Science. 2011;9:405-415.
12. Bortotoli GA, Baron D. Phytoremediation of Toxic Heavy Metals by Brassica Plants: A Biochemical and Physiological Approach. Environmental Advancement. 2022;8: 100204.
13. Ciura J, Poniedzialek M, Jedrszczyk F. Possibility of using plants as metal bioremediants. Journal of Environmental Studies. 2005;14:17-22.
14. Aggarwal H, Goyal D. Phytoremediation of some heavy metals by agronomics crops. Development and Environmental Science. 2007;5:79-98.
15. Diarra L, Kotra, K. K, Prasad, S. Assessment of biodegradable chelating agents in the phytoextraction of heavy metals in multi-metal contaminated soil. Chemosphere. 2021;273:126-268.
16. Ashraf S, Ali Q, Asghar HN. Phytoremediation: Environmentally Sustainable way of Remediation of Heavy Metal Polluted Soil. Ecotoxicol, Enviro. 2019;174:714-727.
17. Abou MA, Yasen AA, Zaghoul SM. Phytoremediation of heavy metals principles, mechanisms, enhancement with several efficiency enhancer methods and perspectives: A Review. Middle East Journal of Agriculture Res. 2020;9(1):186-214.
18. Endres G. Field Pea Production. Res. 2016;53:111-130.
19. Adebayo AJ, Jayoye JT, Adejoro F, Ilemobayo IO, Labunmi L. Heavy metal Pollution of Auto-mechanic Workshop Soils within Okitipupa, Ondo State, Nigeria. Acad. J. Environ. Sci. 2017;5(12):215-223. Available:https://doi.org/10.15413/ajes.2017.0131
20. Mohammad DD, Chen J, Zhang G. Comparison of EDTA and citric acid enhanced phytoextraction of heavy metals in artificially contaminated soil by Typha angustifolia. Internationa Journal of Phytoremediation. 2009;1196:558-574
21. Strom L, Owen DL, Jones TM. Organic acute behavior in a calcareous soil: Sorption reaction and biodegradation rates: Soil Biology and Biochemistry. 2001;31:255-259
22. Parveeen A, Saleem HM, Azeem M. Effect of Citric Acid on Growth, Ecophysiology, Chloroplast Ultrastructure, Phytoremediation Potential of Jute Seedlings Exposed to Copper Stress. Biomolecule. 2020;10:592.
23. Aderholt M, Vogelien D, Koether M, Greipsson S. Phytoextraction of contaminated urban soils by *Panium virgatum* L. Enhanced with Application of Plant Growth regulator (BAP) and Citric Acid. Chemosphere. 2017;15:20-74.
24. Afshan S. Citric Acid enhances the phytoextraction of chromium, plant growth and photosynthesis by alleviating the oxidative damages in *Brassica napus* L. Environmental Science Pollution. Res. 2015;22:11679-11689.
25. Ehsan S. Citric acid assisted phytoremediation of cadmium by *Brassica napus*. L. Ecotox Environ. 2014;106:164-172.
26. Ma JF. Role of organic acids in detoxification of Aluminum in higher plants. Plant Cell Physiol. 2000;41:383-385.
27. Chen YX, Lin YM, Luo YFH, Zhen YL, Yu GM, Tian L, Wong MH. The role of citric acid on the Phytoremediation of Heavy Metal Contaminated Soil; 2003.
28. Matavos-Aramyan S, Moussavi M. Advances in Fenton and Fenton based oxidation processes for industrial based effluent contaminants control. A Review. International Journal of Environmental Sciences and Natural Resources. 2017; 23:12-34.

29. Zhuang P, Yang Q, Shu WS. Phytoextraction of heavy metals by eight plant species in the field: Water, Air and Soil. Pollution. 2007;184:235-242.
30. Hassan A. Accumulation and distribution of heavy metals in soil and food crops around the Ship Breaking Area in the Southern Bangladesh and Associated Health Risk Assessment. Applied Science. 2020; 2(155).
31. Acuna E, Castillo B, Tapia Y. Assisted phytoremediation of lead contaminated soil using *Atriplex halimus* and its effects on soil physical properties. International Journal of Environmental Science and Technology. 2020;7:1925-1938.
32. Hedavatzadey F, Banaee M, Shayesteh K. Bioaccumulation of lead and cadmium by raddish and cress under hydroponic growing medium. Pollution. 2020;6(3):681-693.
33. Jadia CD, Fulekar MH. Phytotoxicity and remediation of heavy metals by fibrous root grass in Soil: Vermin Compost Media. J. Appl. Biosci. 2008;10: 491-499.
34. Xiao XC, Tongbin A, Mei L. Potential of Pterisvittata for phytoremediation of sites contaminated with cadmium and arsenic. The tolerance and accumulation. Journal of Environmental Science. 2008;20:62-67.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/122581