International Journal of Innovative Environmental Studies Research 10(1):115-123, Jan.-Mar., 2022

 www.seahipaj.org **ISSN: 2354-2918**

Phytoextraction Of Lead Polluted Soil Using *Abelmoschus esculentus* (L.)Moench

Akpan, E. N.*; Denise, E. M. & Ntuen, U. E.

Department of Botany and Ecological Studies,
University of Uyo, Uyo, Nigeria
***Corresponding Author: Elsie Nsuhoridem Akpan,**
Tel: +2348065233717, Email: elsieakpan@uniuyo.edu.ng

ABSTRACT
The potential of *Abelmoschus esculentus* (L.)Moench for phytoextraction of lead was investigated. A pot experiment was conducted under natural conditions for a period of 7 weeks. The pots were filled with 5kg of soil and seeds were planted in soil artificially polluted with 0ppm (control), 10ppm, 20ppm, 30ppm, 40ppm and 50ppm of lead respectively. The physicochemical properties of the soil before and after the experiment were determined using standard methods. Leaves, stems and roots of the plant were subjected to analysis for uptake of lead. The results showed that the plants had accumulated a significant concentration of lead in the leaves (26.61mg/kg), stem (8.27mg/kg) and roots (18.81mg/kg). There was no significant difference in the different concentrations of lead used in the study ($p>0.05$). The phytoextraction ability of the plant was assessed in terms of its Bioconcentration factor (BCF) and Translocation factor (TF). The concentration of lead in the roots and shoots after 7 weeks showed that more concentration of lead was translocated from the root to the leaves. The result obtained suggests that *Abelmoschus esculentus* could be used for phytoextraction of lead from contaminated soil.
Keywords: Bioremediation, Phytoremediation, Phytoextraction, Lead

INTRODUCTION
Heavy metals are a type of pollutant that can be found in the environment. Aside from natural activities, practically every human activity has the potential to produce heavy metals as a byproduct. The spread of heavy metals containing sewage sludge and the migration of these contaminants into non-contaminated areas as dust or leachates through the soil are two examples of occurrences that contribute to ecosystem contamination (Gaur and Adholeya, 2004). Several strategies for cleaning up the environment from these types of toxins are already in use, but the majority of them are expensive and fall short of their potential. Chemical methods produce significant volumes of sludge, which raises costs (Giahi *et al*., 2009). Chemical and thermal methods are all technically challenging and costly, and are capable of degrading the important component of soils (Negri *et al*., 1995). Traditionally, heavy-metal-contaminated soils have been remedied by either onsite management or excavation and disposal to a landfill. This type of disposal only relocates the contamination problem, as well as the risks associated with transporting contaminated soil and contaminant migration from the landfill into the surrounding ecosystem.
Concerns about environmental pollution have prompted the development of devices to determine the presence and mobility of metals in soil (Laiho *et al*., 2004). Phytoremediation is now a viable and cost-effective technological method for extracting inert metals and metal contaminants from polluted soil. The use of plants to clean up contaminants from soils, sediments, and water is known as phytoremediation. This technology is both ecologically friendly and cost-effective in the long run. Hyper accumulator plants are plants that have a high metal-accumulating capability (Kurukote *et al*., 2006). Phytoextraction, which

is part of Phytoremediation, takes advantage of plant root systems' unique and selective absorption capabilities, as well as the entire plant body's translocation, bioaccumulation, and contaminant degrading capacities (Negri *et al*., 1995). Many plant species have been successful in absorbing pollutants from soils, including lead, cadmium, chromium, arsenic, and different radionuclides. Phytoextraction, one of the phytoremediation categories, can be used to remove heavy metals from soil by utilizing its ability to uptake elements that are required for plant growth (Fe, Mn, Zn, Cu, Mg, Mo, and Ni). Cd, Cr, Pb, Co, Ag, Se, and Hg are examples of metals with unknown biological functions that can accumulate (Kurukote *et al*., 2006).

Abelmoschus esculentus (L.) Moench, known as Okra is an annual, erect herb up to 5m (but typically about 2m) tall, with succulent stem and scattered stiff hairs. Okra is a cultigen (a plant that has been altered by humans through a process of selective breeding). The exact origin of okra is unknown, but it is thought to have come from Africa, where it has been grown as a crop for centuries. Evidence suggests it was grown in Egypt as long ago as 2,000 BC. Today it is widely cultivated for its edible green fruits, which are harvested when immature (after 3–5 days of development), and are infamous for their slimy mucilage.

Okra is widespread in cultivation in the tropics, subtropics and warmer temperate zones. It is particularly popular in Africa, India, the Philippines, Thailand, Brazil, Turkey, Spain and the southern USA. It is naturalised in some areas. The related species West African okra *(Abelmoschus manihot*) is restricted to the humid and perhumid (wettest) climates of Africa.

Heavy metals, particularly lead, are major contaminants that endanger the environment as well as human and animal health (Ijah and Ndana, 2008). Heavy metal pollution of soil has existed for centuries, but its scope has grown dramatically in the last fifty years as a result of technical advancements and greater consumer use of metal-containing items (Majid and Argue, 2001). The investigations on the variability of bioaccumulation in non-hyperaccumulator species were based on agricultural or market plants for human use, rather than heavy metal phytoextraction employing plants. Their goal was to find cultivars that would reduce the entrance of heavy metals into food chains and thereby the health effects of soil contamination. However, in the phytoextraction context, studies are carried out with the opposite purpose and plant species are selected for their ability to accumulate and extract metals from polluted soils. One of the key steps in phytoextraction is the selection of plant species and higher heavy metal extracting values have been observed by many scientists in naturally occurring plant population. Hence, the aim of this work was to examine the feasibility of *Abelmoschus esculentus* as a hyperaccumulator plant for lead in soils.

MATERIALS AND METHODS

Collection and Processing of Samples.

The soil sample used for this study was collected from the depth of 0 - 20cm within the University of Uyo main campus, Uyo, Nigeria. The soil was transported into 18 different pots using a soil auger. The taxonomic classification of the experimental soil was sandy with the pH 5.30, mature seeds of *Abelmoschus esculentus* were collected at Akpan andem market, Uyo, Nigeria. The local seeds were used because they have the potential to withstand adverse climatic conditions and they easily adapt to harsh environmental conditions.

Preparation of Heavy Metal Contaminants.

The lead was added to the soil as lead nitrate (Pb $(NO3)_2$, 1.599g of Pb $(NO3)_2$ and 10mL of Nitric acid was dissolved in 1000mL of distilled water to make stock solutions of 10, 20, 30, 40, and 50 milliliters. These different concentrations were then measured from the stock solutions into a 100ml capacity measuring cylinder and made up to the mark to give 10ppm, 20ppm, 30ppm, 40ppm, 50ppm and 0ppm (control) metal concentrations. The soil was spiked with different concentrations of lead and thoroughly mixed

Experimental Design and Treatment.

The study was a pot experiment carried at the green house of Department of Botany and Ecological Studies, University of Uyo, Uyo, Nigeria. The experiment was laid out in a complete randomized design and the treatments were in three replicates. The experimental pots were filled with 5kg lead contaminated soil of different concentrations of lead contaminants. Eight seeds were planted in each pot, which were later thinned down to four after germination of the plant. The plants were watered with 500mls of water for the first two weeks and later reduced to 20mls of tap water daily in each pot for the remaining 5weeks.

Analysis of Lead

After 7 weeks of planting, all the plants were harvested separately according to the soil treatment, separated into three compartments leaves, root and stem. The 18 replicates of each treatment were pooled together to give composite sample of each treatment, the plants were washed in water to eliminate soil, dirt, possible parasite or their eggs, and finally washed using a deionized water. The plants were dried in an oven at 80^oC and grinded to powdered form before determining the presence of heavy metals.

Procedure for Digestion: 1g of sample was weighed into a digestion flask, 20ml of nitric acid was added and 10ml of perchloric acid was also added, a drop of concentrated sulphuric acid was added, the mixture was allowed to stand for 30minutes**.** This was digested using a digestion chamber in the laboratory until the color turned white, this shows that the samples were digested, afterwards it was allowed to cool. 30mls of distilled water was added and was filtered with a filter paper, the filtrate made up to 50mls solution with distilled water, the digested solution was stored in a bottle ready for the determination of heavy metals using atomic absorption spectrophotometer with different lamps in position. Lead concentrations were determined using Atomic Absorption spectrophotometer (AAS).

Determination of Bioconcentration and Translocation Factor

Bioconcentration factor (BCF) and Translocation factor (TF) were calculated using the formula of Yadav *et al* (2009) as:

$$\text{Bioconcentration factor (BCF)} = \frac{\text{Average metal concentration in the whole plant (mg/kg)}}{\text{Metal concentration in soil (mg/kg)}}$$

$$\text{Translocation factor (TF)} = C_{aerial} \times 1/C_{root}$$

C_{aerial} = metal concentration in the aerial part of plant (stem and leaf)

C_{root} = metal concentration in root of plant

Statistical Analysis

The one-way analysis of variance test was used to analyze and compare the data using SPSS 23 software.

RESULTS

Table 1: Physicochemical Properties Of The Soil

Test Parameter	Values
Sand (%)	91±0.33
Silt (%)	1.67±0.33
Clay (%)	7.2±0.0
pH	5.2±0.30
Electrical conductivity	0.08±0.02
Organic matter (%)	6.15±0.14
Total Nitrogen (%)	0.15±0.003
Available Phosphorus (mg/kg)	55.02±5.99
Calcium (cmol/kg)	7.17±0.12
Magnesium (cmol/kg)	2.79±0.15
Sodium (cmol/kg)	0.06±0.006
Potassium (cmol/kg)	0.14±0.006
Exchange acidity (cmol/kg)	1.75±0.05
Effective cation exchange capacity (cmol/kg)	11.90±0.34
Base saturation (%)	75.28±9.80

Mean ± Standard error

Table 2: Physicochemical Properties Of The Soil After Planting

Test Parameter	**Values**
Sand (%)	82.33±0.91
Silt (%)	1.126±0.0047
Clay (%)	3.24±0.049
pH	7.03±0.031
Electrical conductivity	0.063±0.040
Organic matter (%)	4.90±0.127
Total Nitrogen (%)	0.103±0.012
Available Phosphorus (mg/kg)	63.13±2.52
Calcium (cmol/kg)	0.02±0.082
Magnesium (cmol/kg)	0.17±0.0049
Sodium (cmol/kg)	2.09±0.131
Potassium (cmol/kg)	9.85±0.439
Exchange acidity (cmol/kg)	1.73±0.021
Effective cation exchange capacity (cmol/kg)	13.72±0.189
Base saturation (%)	77.69±1.750

Mean ± Standard error

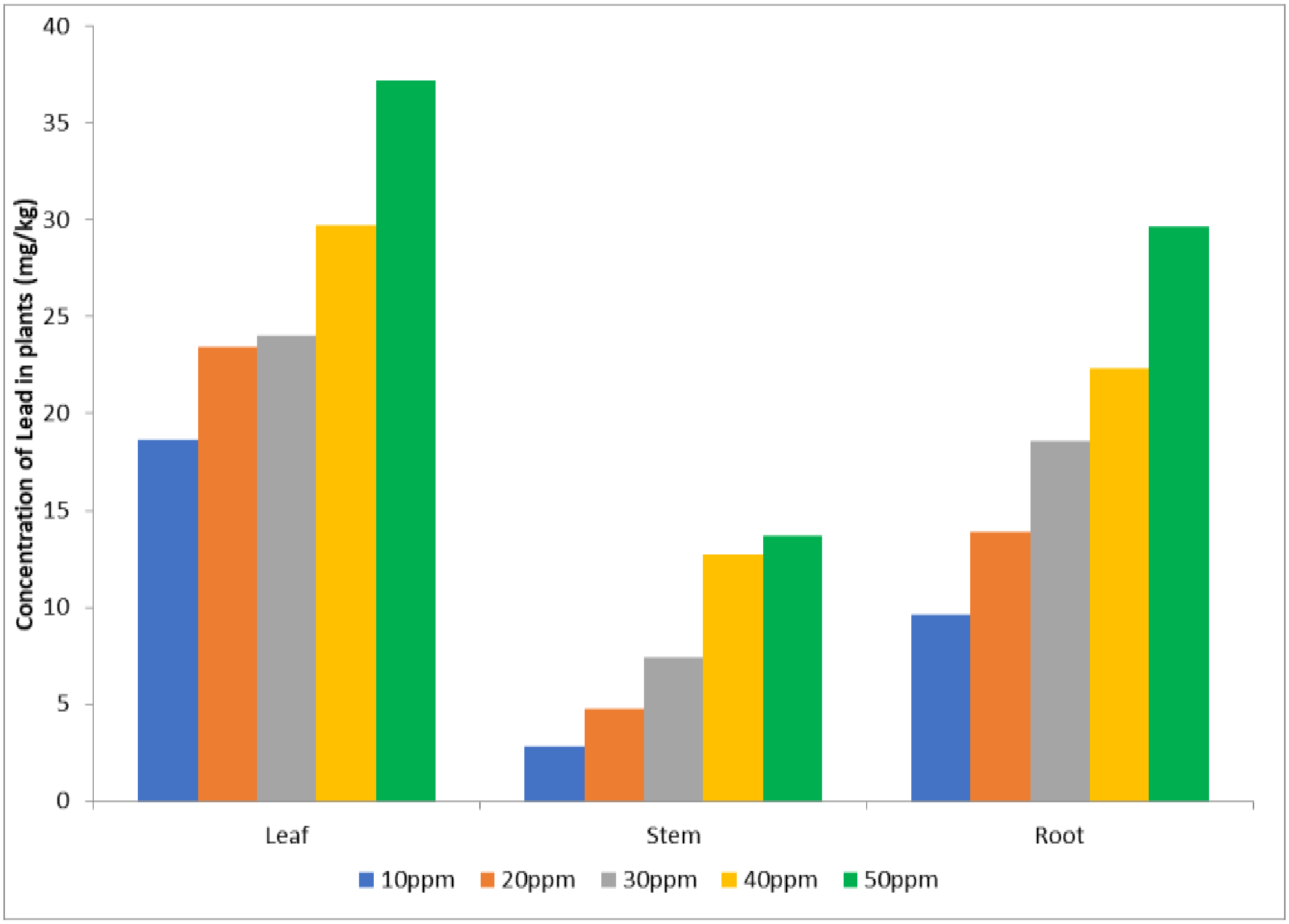

Figure 1. Concentrations of lead in stems, roots and leaves of *Abelmoschus esculentus*

Table 3: Bioconcentration factor (BCF) and translocation factor (TF) of lead in *Abelmoschus esculentus*

Treatment	BCF	TF(LEAVES)	TF(STEM)
10ppm	1.03	1.94	0.29
20ppm	0.70	1.69	0.34
30ppm	0.56	1.29	0.4
40ppm	0.54	1.32	0.56
50ppm	0.54	1.25	0.46

DISCUSSION

Table 1 shows the physicochemical properties of the soil before planting. Table 2 shows the physicochemical properties of soil after 7 weeks of phytoextraction studies. The high pH level of the soil is generally within the range of soil established by FEPA (1991). The pH of the soil after plant harvest (7.03) was higher than the pH of the uncontaminated soil (5.03) probably, this may be due to the presence of lead as a contaminant in the polluted soil.

Soil pH affects heavy metal sorption, as well as the solubility and hydrolysis of metal hydroxides, carbonates, and phosphates, as well as ion–pair formation and solubility of organic matter, as well as the surface charge of Fe, Mn, and Al-oxides, organic matter, and clay edges (Tokalioglu *et al*.,2006) These findings suggest that soil variables such as pH, clay, and organic matter influence metal uptake (Jung *et al*.,1996).

The unpolluted soil before planting had higher organic matter (6.15%) than the polluted soil which had 4.90%. There was a slight decrease in nitrogen content while available phosphorous was higher in the polluted soil (63.13%) than the unpolluted (55.02%). The unpolluted soil before planting had 0.06, 0.14, 2.79 and 7.17cmol/kg of Na^+, K^+, Mg^{2+} and Ca^{2+}, respectively, while 0.02, 0.16, 2.09 and 9.85 cmol/kg were observed for Na^+, K^+, Mg^{2+} and Ca^{2+}, respectively in the polluted soil after harvesting the plants. When compared to other cations, Ca^{2+} had the highest value of 7.17 and 9.85cmol/kg for unpolluted and polluted soil, respectively. Also, the effective cations exchange capacity (ECEC) was seen to be higher in polluted soils, (13.72cmol/kg) than in unpolluted (11.91 cmol/kg). The difference (increase or decrease in the soil properties) observed might be due to the lead added in the soil. Lead (when added to soil) can change soil properties, according to Ryser and Sauder (2006), and data from studies on the toxic effect of heavy metals on soils have been used to establish the concentrations at which heavy metals affect biological soil processes for regulatory purposes, according to Giller *et al* (1998). Metal bioavailability in soil is a dynamic process that is influenced by a variety of chemical, biological, and environmental variables.

Lead was not detected in minute quantities in unpolluted soil used for the experiment but 7 weeks after lead pollution, Pb concentration was recorded and determined. The concentration of lead recorded in the soil after 7 weeks of phytoextraction was less than the concentration of lead introduced into the soil in their respective treatments. It was found that <0.0001, <0.0001, <0.0194, < 0.0001 and <0.0001mg/kg of residual lead were detected in the soil treated with 10, 20, 30, 40, 50ppm of lead, respectively. This indicated that large proportion of lead was removed from the soil which could be traced to phytoextraction potential of the plant used. It could also be possible that some of the lead might have escaped into the atmosphere. USEPA (2000) reported that heavy metals (when mopped up by plants) have the ability to escape into the atmosphere which could be in line with this finding. There was no significant difference in the different concentrations of lead used in the study ($p>0.05$).

In lead contamination soils planted with *Abelmoschus esculentus,* the concentrations of lead after 7 weeks for stem compartment were 2.790, 4.742, 7.440, 12.700 and 13.690 mg/kg, roots were 9.610, 13.847, 18.561, 22.351 and 29.630 mg/kg and leaves were 18.662, 23.464, 24.051, 29.698 and 37.191mg/kg observed at 10,20,30,40 and 50ppm respectively.

The results indicate that *Abelmoschus esculentus* mopped up substantial concentration of lead in the above ground biomass compared to concentrations in the roots. The results also showed that, at the end of 7weeks period the leaves had the highest concentration of lead followed by the roots and stems in that order.

Huang and Cunningham (1996) and Blaylock *et al* (1997) found that plants can remove between 180 and 530kg/ha of Pb/year, making remediation of sites contaminated with up to 2,500mg/kg possible in fewer than 10years. This implies that about 250mg/kg can be removed in a year at an average of 21mg/kg in 4weeks. The value is far lower than Pb concentrations observed at the end of the sampling period (7 weeks) in the *Abelmoschus esculentus.* This indicates that this plant is effective in mopping up the Pb from contaminated soil. Therefore, this plant has a potential to accumulate heavy metals and may be selectively used for phytoextraction of metal contaminated soils. According to emerging technology for the phytoremediation of metal in soil (ETPMS, 1997) phytoextraction is the ability of plants to absorb, concentrate, and precipitate toxic metals from contaminated soils, into the above ground biomass (shoots, leaves, stems and seeds).

The highest BCF was recorded in soil polluted with 10ppm lead. This might be due to the fact that at moderately low concentration of lead in the soil, plants tend to accumulate more metals than higher concentrations (Benzarti *et al.*, 2005). The lowest was recorded in soil polluted with 40 and 50ppm. The highest TF in leaves and stems was recorded in soil polluted with 10ppm. The ability of phytoremediation has commonly been characterized by a TF (Baker, 1981), which is defined as the ratio of the metal concentration in the shoots to that in the roots. Plants with TF values >1 are classified as high efficiency plants for metal translocation from roots to shoots. All the concentration of Pb in leaves showed TF values >1 indicating that the plants could be classified as high efficient plant for metal translocation from the roots to the leaves. Wei and Chen (2008) suggested that plant species with TF values >1 actively take up metals from the soil and accumulate them in their above ground parts, therefore they could be good phytoremediators.

CONCLUSION

Heavy metal pollution has been a major source of concern in recent decades due to the health risks that they provide to humans and other species when they accumulate within a biological system. It is clear that phytoextraction has the potential to help restore equilibrium to a stressed ecosystem, but caution is advised. The ability of *Abelmoschus esculentus* to repair Pb-contaminated soil was established in this study. After 7 weeks of cleanup, this plant had the largest concentration of Pb in its leaves. However, planting *Abelmoschus esculentus* in a lead-contaminated soil without a thorough investigation of the soil for Pb contamination could offer a significant risk to those who harvest the plant for consumption, as this plant has been reported to collect a significant amount of Pb.

REFERENCES

Baker, A. J. M. (1981). Accumulators and excluder-strategies in the response of plants to heavy metals. *Journal of Plant Nutrition*, 3:643–654.

Benzarti, S., Mohri, S. and Ono, Y. (2008). Plant response to heavy metal toxicity: comparative study between the Hyperaccumulator Thlaspi caerulescens (ecotype ganges) and nonaccumulator plants: lettuce, radish, and alfalfa. *Environmental Toxicology*, 23(5):607–616.

Blaylock, M. J., Salt,D. E., Dushenkov S., (1997).Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. *Environmental Science and Technology*, 31(3) 860–865.

Emerging Technologies for the Phytoremediation of Metals in Soils, ETPMS. (1997) Interstate Technology and Regulatory Cooperation Work Group(ITRC), http://www.itrcweb.org.

Federal environmental protection agency (FEPA), (1991).Guidelines and standards for Industrial effluent, gaseous emissions and hazardous waste management in Nigeria.

Gaur A. and Adholeya, A. (2004). Prospects of arbuscularmycorrhizal fungi in phytoremediation of heavy metal contaminated soils. *Current Science*, 86(4):528–534.

Giahi, M., Rakhshaee, R. and Pourahmad, A. (2009). Studying effect of cell wall's carboxyl-carboxylate ratio change of Lemna minor to remove heavy metals from aqueous solution. *Journal of Hazardous Materials*,163(1):165–173.

Giller, K. E., Witter, E., and Mcgrath, S. P. (1998). Toxicity of heavy metals to microorganisms and microbial processes in agricultural soils: a review. *Soil Biology* and *Biochemistry*, 30(10-11):1389–1414.

Huang J. W. and Cunningham, S. D. (1996). Lead phytoextraction: species variation in lead uptake and translocation. *New Phytologist*, 134 (1): 75–84.

Ijah, U. J. J. and Ndana, M. (2003). Stimulated biodegradation of crude oil in soil amended with periwinkle shells. *The Environmentalist* 23:249-254.

Jung, M. C. and Thornton, I. (1996). Heavy metal contamination of soils and plants in the vicinity of alead-zinc mine, Korea. *Applied Geochemistry*, 11(1-2): 53–59.

Kurukote, J., Cho-Ruk, K. Supprung, P. and Vetayasuporn, S. (2006). Perennial plants in the phytoremediation of lead-contaminated soils. *Biotechnology*, 5(1): 1–4.

Laiho, J. V. P., Shtangeeva, I., Kahelin, H. and Gobran, G. R. (2004). Phytoremediation of metal-contaminated soils. Symposia Papers Presented Before the Division of Environmental Chemistry. American Chemical Society, Anaheim, Calif, USA, , http://ersdprojects.science.doe.gov/workshop_pdfs/california_2004/p050.pdf.

Majid, A. and Argue, S. (2001). Remediation of heavy metal contaminated solid wastes usi agglomeration techniques. *Minerals Engineering,* 14(11):1513–1525.

Negri, M. C. Hinchman, R. R. and Gatliff, E. G. (1995). *Phytoremediation: using green plants to clean up contaminated soil, groundwater, and wastewater*. Argonne National Laboratory Hinchman, Applied Natural Sciences, Inc.

Ryser, P.and Sauder, W. R. (2006). Effects of heavy-metal-contaminated soil on growth, phenology and biomass turnover of *Hieraciumpiloselloides*. *Environmental Pollution*, 140(1):52–61.

Tokalioğlu, S. Kartal, Ş. and Gültekin, A. (2006) Investigation of heavy-metal uptake by vegetables growing in contaminated soils using the modified BCR sequential extraction method. *International Journal of Environmental Analytical Chemistry*, 86(6):417-430.

U.S. Environmental Protection Agency, USEPA. (2000). Introduction to Phytoremediation, National Risk Management Research Laboratory, EPA/600/R-99/107. http://www.clu-in.org/download/remed/introphyto.pdf.

Wei, C. Y. and Chen, T. B. (2006). Arsenic accumulation by two brake ferns growing on an arsenic mine and their potential in phytoremediation. *Chemosphere*, 63(6): 1048–1053.

Yadav, S. K., Juwarkar, A. A., Kumar, G. P., Thawale, P. R., Singh, S. K. and Chakrabarti, T. (2009). Bioaccumulation and phyto-translocation of arsenic, chromium and zinc by *Jatropha curcas L.*: impact of dairy sludge and biofertilizer. *Bioresource Technology*, 100(20): 4616–4622.