Nig J. Biotech. Vol. 35 No. 2 (2018): 43-49
ISSN: 0189 1731
Available online at
http://www.ajol.info/index.php/njb/index
and www.biotechsocietynigeria.org
DOI: https://dx.doi.org/10.4314/njb.v35i2.6

NIGERIAN JOURNAL OF BIOTECHNOLOGY

Phyto-extraction ability of *Digitaria exilis* (Fonio) to heavy metals

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Abstract

Among the different contaminants in the environment, heavy metals are unique due to the fact that they cannot be broken down to non-toxic forms. The research was conducted to determine the remediation potential of *Digitaria exilis* and to identify the varieties that can tolerate heavy metals contamination. Soil was collected in an Automobile mechanic workshop, Government Reserved Area (GRA) Ado-Ekiti. Five kilograms (5 kg) of soils collected was air dried, sieved and filled into plastic buckets. The experiment was a completely randomized designed with three replicates. Three different accessions of *Digitaria exilis* namely; Jakah D Iburua (JAK), Dinat D Iburua (DID) and Jiw D Iburau (JIW) were planted. Soils were watered till field moisture capacity. Plant height was determined every two weeks after planting. Soil analysis was carried out to determine the physical and chemical properties before and after the experiment. Heavy metals including Cd, Fe, Zn, Pb, As and Cu were determined in the plant roots and shoots using Atomic Absorption Spectrophotometer (AAS). Accession JAK Bio-accumulated heavy-metals in their root than shoot, consequently, significant high plant height was recorded. However, shoot of accession JAK accumulated 22 % Pb, 46.78% Cu and 37.5% Cd and 20.3% Zn than others whereas accession DID accumulated As than accession JAK. Therefore, accession JAK demonstrated highest phyto-extraction of heavy-metals potentials at the root. Variety JIW has the lowest plant height, root and shoots heavy metals accumulation. Although heavy metals contaminated soil affected the growth of *D. exilis*, nevertheless accession JAK has the highest productivity under this condition and can be further recommended in phyto-extraction of heavy-metals in polluted sites.

Keywords: Phyto-extraction, *Digitaria exilis* and Heavy-metals
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Introduction

The need for economic growth especially in developing countries has led to increase in different industries which emit heavy metals (Ahmadpour et. al., 2012). Soils may become contaminated by the increasing accumulation of heavy metals and metalloids through emissions from the rapidly expanding industrial areas (Wuana & Okieimen, 2011; Ahmadpour et. al. 2012). Disposal of high metal wastes, leaded gasoline and paints, land application of fertilizers, animal manures, sewage sludge, pesticides, wastewater irrigation, coal combustion residues, spillage of petrochemicals, and atmospheric deposition are the various activities leading to contamination of our soils (Sahibin et. al., 2002; Jadia & Fulekar, 2009; Wuana & Okieimen, 2011). The heavy metal contents of these stated pollutants cannot be oxidized by microbes and chemically degraded as in the case of organic contaminants (Ahmadpour et. al. 2012). The average half-life of these metals is as high as 20 years (Ruiz et. al., 2009).

Humans and ecosystem in general, are at the receiving end of the risks and hazards posed by heavy metal contamination of soil (McLaughlin et. al. 2000; Yoon et. al., 2006), with more than 10 million people's health under threat in many countries (Ahmadpour et. al. 2012). It has become necessary to reduce the concentrations of these heavy metals in the environment to a safe level in a sustainable and cost-effective way. Current conventional methods to remediate heavy metal-contaminated soil and water, such as ex situ excavation, landfill of the top contaminated soils (Zhou & Song, 2004), detoxification (Ghosh & Singh, 2005), and physico-chemical remediation, are expensive (Danh et. al., 2009), time consuming and labor exhaustive.

The use of plant to control the concentrations of these metals has proved effective, both on land and in water bodies, through the process of phytoremediation and rhizo-filtration respectively. It is a low cost, long term, environmentally and aesthetically friendly method of immobilizing/stabilizing, degrading, transferring, removing, or detoxifying contaminants, including metals, pesticides, hydrocarbons, and chlorinated solvents (Susarla et. al., 2002; Jadia & Fulekar, 2008; Zhang et. al., 2010). Several hyper-accumulators have been found by several workers, they include *Thlaspica erulescens* (Pence et. al., 2000), *Sedum alfredi* (Yang et. al., 2004), *Arabidopsis halleri* (Bert et. al., 2003), *Brassica campestris* (Glick, 2003) and *Pistia stratiotes* (Moodley et al., 2007; Abubakar et. al. 2014).

Fonio (*Digitaria exilis* and *Digitaria iburua*) is probably the oldest African cereal. For thousands of years, West Africans have cultivated it across the dry savannas. Indeed, it was once their major food. Even though few other people have ever heard of it, this crop still remains important in areas scattered from Cape Verde to Lake Chad. Therefore, this study aimed at investigating the phytoremediation ability of fonio in soil contaminated with petroleum derivatives in order to understand its mechanism of remediation and to identify the varieties which can tolerate heavy metals contamination.

Materials and Methods

Soil preparation and Planting

The experiment was conducted at Federal University Oye-Ekiti screen house. Contaminated soil from an automobile mechanic workshop (AUMS) along with agricultural field as control soil were collected in Ado-Ekiti and air dried for 3 days which were sieved using a 2 mm mesh to remove debris. Five kilogram (5 kg) air dried and sieved soil was weighed into each plastic buckets with perforated at the base to drain water and increase soil aeration. The contaminated soil was watered immediately for a week to allow homogeneous mixture of the oil.

Three different varieties of *Digitaria exilis* (Dinat D Iburua (DID), Jakah D Iburua (JAK) and Jiw D Iburua (JIW)) were planted on the plastic bucket each having three (3) replicate each (2 contaminated, 1 control) which were then watered. Growth was observed on the third day of planting. Growth parameters were accessed second and third week after planting. Plants were watered regularly and the buckets were maintained weed free throughout the experiment.

Measurement of Growth parameters

The plant heights were measured weekly. The experiment was terminated 4 weeks after planting. Thereafter, both shoots and roots were analyzed at Center for Energy, Obafemi Awolowo University, Ile-Ife, Osun, Nigeria (OAU). The plants were dried using an oven at a temperature of 60^0C to constant weight.

Soil and Plant Chemical Analysis

One gram (1g) of the dried soil sample was accurately weighed into a teflon beaker and 20ml Hydrofluoric acid was carefully added. The beaker was heated until the sample was completely digested, at near dryness, 20ml double distilled water was carefully added through the side and allowed to cool, filtered and the filtrate made up to 100ml in a standard flask. The dried plant samples were each ground and weighed (0.20g) into a beaker, 15ml freshly weighed (0.20g) into a beaker, 15ml freshly prepared aqua-regia was added. The beaker was allowed to stand for 24hrs after which it was heated to near dryness. 20ml double distilled water was then added and heated until a clearer digested was obtained; this was then allowed to cool and filtered. The filtrate was made up to 50ml in a standard flask.

Quantitative data were subjected to one way analysis of variance at P=0.05 and differences among means were separated using Duncan Multiple range test.

Results

The properties of the soil used for the experiment are stated in Table 1. The soil used was sandy loam, alkaline with exchangeable acidity of 2.5mg/k and low organic compound of 9.0g/Kg (Table 1). The values of heavy metals such as Cd, Fe, Zn, Pb and Cu ranged from 3.00-45.00.

Table 1: Soil Physio-Chemical Properties of the Automobile Mechanic Soil

Property	Soil
pH (1:1 soil water)	5.5
Exchangeable acidity (mg k^{-1})	2.5
Organic compounds (g kg−1)	9.0
Available nitrogen (%)	1.04
Available phosphorous (ppm)	39.42
Cd (mg/l)	5.40
Fe (mg/l)	39.25
Zn (mg/l)	25.2
Pb (mg/l)	3.40
Cu (mg/l)	41.0
Mg (mg/l)	35.4
K (mg/l)	35.2
Ca (mg/l)	18.2
Sand	84
Clay	7
Silt	9
Soil Textural Class	Sandy loam

Plant Growth

Fonio were cultivated on the automobile mechanic soil for four weeks. The plants showed symptoms of stunted growth for the period of exposure. There was an increase in plant height over the four weeks for both AUMS and CNT. Plants cultivated on AUMS had no significant differences in plant height at 2 weeks after planting (WAP) and 4 WAP for the three accessions. Accession JAK had the highest plant height of 1.6 cm and 25.2 cm at 4WAP for AUMS and CNT respectively (Figure 1).

Heavy Metals Accumulation

The concentration of heavy-metals found in the shoot of fonio accessions ranges from 0.5 mg/l - 27.25mg/l. Accession JAK absorbed Pb, Cd, Cu, Zn and Fe higher than other accessions in AUMS and CNT. However for Arsenic, accession DID accumulated higher than the other accessions in the shoot. Accession JAK accumulated 22% Pb higher than DID and JIW. JAK also absorbed 50% of Cd higher than JIW and 37.5% greater than DID. Copper accumulated in the shoot of JAK was 46.78% higher than accession DID and 36.67% greater than JIW. Iron (Fe) uptake in the shoot of accession JAK is significantly higher than other accessions. Shoots of accession JAK absorbed Zinc (Zn) of about 20.3% greater than DID and 13% higher than JIW. Accession JIW significantly accumulated Cu and Zn at the shoot than DID accession. Accumulation of Cd, Pb and Fe at the shoots of Fonio ranged as follows: JAK>DID>JIW (Table 2).

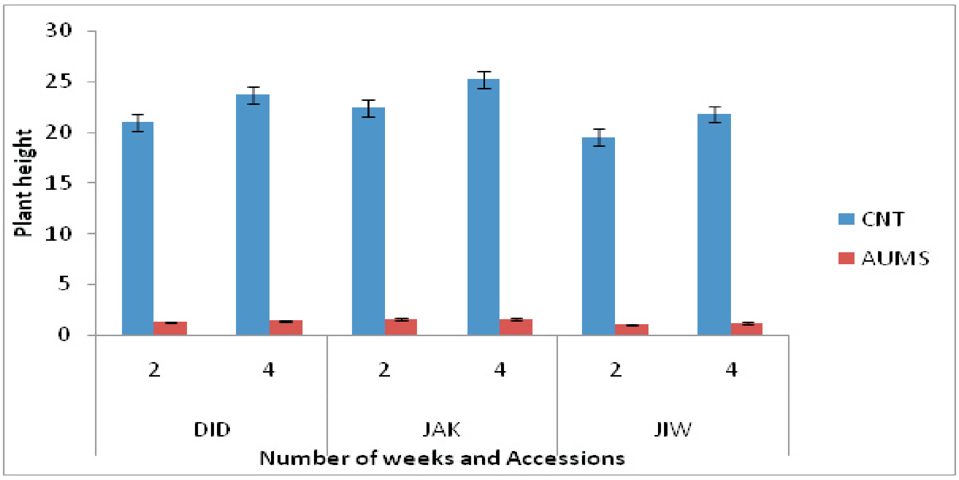

Figure 1: Plant Height (cm) of Digitaria exilis **Cultivated on Mechanic Automobile Soil**

Table 2: Absorption Concentrations (mg/l) of Heavy-Metals in the Shoot

Acc	Treat	Cd	Fe	Zn	Pb	Cu	As
DID	AUTS	2.25^{b}	19.50^{b}	13.75	1.25^{b}	14.50	2.25^{a}
	CNT	0.75^{e}	13.50^{e}	3.50^{e}	0.50^{d}	9.50^{e}	0.50^{d}
JAK	AUTS	3.50^{a}	22.25^{a}	17.25^{a}	2.00^{a}	27.25^{a}	1.50^{b}
	CNT	1.00^{d}	12.00^{f}	5.50^{d}	0.75^{c}	14.25^{c}	0.50^{d}
JIW	AUTS	1.75^{c}	17.25^{c}	15.00^{b}	1.00^{bc}	17.25^{b}	2.00^{a}
	CNT	0.75^{e}	14.00^{d}	5.00^{d}	0.50^{d}	10.00^{d}	1.00^{c}

Table 3: Concentration (mg/l) of Heavy-Metals Uptake by Fonio

Acc	Treat	Cd	Fe	Zn	Pb	Cu	As
DID	AUTS	2.75^{b}	31.00^{b}	19.75^{b}	2.25^{b}	35.75^{b}	4.00^{b}
	CNT	1.00^{d}	13.75^{e}	5.75^{e}	0.75^{e}	11.75^{e}	0.75^{f}
JAK	AUTS	3.50^{a}	38.60^{a}	25.25^{a}	3.25^{a}	38.75^{a}	5.00^{a}
	CNT	1.00^{d}	15.00^{d}	7.25^{d}	1.25	14.75^{d}	1.50^{d}
JIW	AUTS	1.75^{c}	21.75	16.50^{c}	1.50^{c}	22.75	3.50^{c}
	CNT	0.75^{e}	12.25^{f}	4.25^{f}	0.75^{e}	11.25^{e}	1.00^{e}
Acc	Treat	Cd	Fe	Zn	Pb	Cu	As
DID	AUTS	2.75^{b}	31.00^{b}	19.75^{b}	2.25^{b}	35.75^{b}	4.00^{b}
	CNT	1.00^{cd}	13.75^{d}	5.75^{e}	0.75^{e}	11.75^{e}	0.75^{f}
JAK	AUTS	3.50^{a}	38.60^{a}	25.25^{a}	3.25^{a}	38.75^{a}	5.00^{a}
	CNT	1.00^{cd}	15.00^{e}	7.25^{d}	1.25^{d}	14.75^{d}	1.50^{d}
JIW	AUTS	1.75^{c}	21.75	16.50^{c}	1.50^{c}	22.75	3.50^{c}
	CNT	0.75^{d}	12.25^{f}	4.25^{f}	0.75^{f}	11.25^{e}	1.00^{de}

Heavy metals were highly concentrated at root of accession JAK than any other accessions in both AUTS and CNT. JAK absorbed above 50% Cd and Pb at the root more than JIW and JAK accumulated 21.4% Cd and 31% Pb higher than accession DID. There is no significant difference in absorption of Cu at the root of JAK and DID whereas accession JAK absorbed 41% Cu higher than accession JIW at the root. Zinc concentration at the root of JAK plant on AUMS is 44% greater than zinc at the root of JIW and 34.65% higher than zinc concentration at the root of DID. The absorption level of heavy-metals at the root of Fonio ranged as follows: JAK>DID>JIW (Table 3).

The bioaccumulation factors of accession JAK at the root and shoot were significantly higher than other accessions. Bio-concentration factor of Cd and Pb at the root and shoot of JIW was twice that in JAK. There were no significant differences in bio-concentration factors of Cu, Zn and Fe at shoot level among the accessions. However, roots of JAK accounted for below 40% bio-concentration higher than JIW (Table 4).

Table 4: Bio-concentration Factors of Different Accessions of Fonio

Accessions	Treatments	Cd		Pb		Cu		Zn		Fe	
		Sht	Rt	Sht	Rt	Sht	Rt	Sht	Rt	Sht	Rt
DID	AUMS	0.32	0.51	0.37	0.66	0.35	0.92	0.55	0.78	0.49	0.79
JAK	AUMS	0.42	0.65	0.59	0.96	0.66	0.95	0.68	1.00	0.57	0.98
JIW	AUMS	0.28	0.32	0.29	0.44	0.42	0.55	0.59	0.65	0.44	0.55

As shown in Table 5, the transfer factor of all the accessions ranged from 0.00 - 0.90. Accessions JAK and DID had their transfer factors between 0.00 - 0.65 whereas, JIW had its transfer factor of Zn to be 0.91.

Table 5: Transfer factors of Different Accessions of Fonio under Automobile Mechanic Soil (AUMS)

Accessions	Treatments	Cd	Pb	Cu	Zn	Fe
DID	AUMS	0.64	0.56	0.38	0.69	0.63
JAK	AUMS	0.64	0.62	0.70	0.68	0.58
JIW	AUMS	0.86	0.67	0.76	0.91	0.79

Discussion

The accumulation and concentration of these heavy metals in the roots of the three accessions studied is a further indication that they are good candidates for phyto-extraction. This is supported by USEPA (2000) that heavy metals such as Pb, Cd, Cu, Ni, Zn, and Cr are primarily retained within the roots and they can only be removed by rhizo-filtration. According to Jadia and Fulekar (2009), rhizo-filtration is beneficial in disallowing heavy-metals from being translocated to the shoots. Therefore, the Fonio accessions studied, being non-hyperaccumulators could be used for this purpose. Raskin and Ensley (2000) reported that terrestrial plants with fibrous and longer root systems are good candidates of rhizo-filtration because of increase in the amount of root area. Consequently, the higher heavy metal concentrations in the roots of the fonio accessions studied could be as a result of the large surface area of their fibrous root systems being monocots.

As reported by Oloyede et al. (2013), bio-concentration factor (BF) defined as the concentration ratio of heavy metal in the plant to that in the soil is used to measure the effectiveness of a plant in concentrating heavy metal into its biomass. The highest heavy metals bio-concentration factors observed in both roots and shoots of accessions JAK compared to the other accessions shows that it was more effective in accumulating heavy metals in its biomass. Also the highest bio-concentration factors which are observed in the roots than shoots of all the accessions means that fonio is shoots of all the accessions means that fonio is better in accumulating heavy metals to its roots than shoots. This means that these fonio accessions can effectively serve two purposes i.e. as food crops and as remediators of heavy metal contaminated sites. This is because the bulk of the heavy metals are concentrated in the roots of these plants and not the shoot which is consumed by humans.

Transfer factor (TF) which is the heavy metal concentration ratio between shoot and root biomass and is used to measure the effectiveness of a plant in translocating heavy metal from root to the shoot. If the transfer factor is below 1 in any of the treatments, it means that the plant is poor in translocating that particular heavy metal to its shoots in that treatment (Oloyede et. al., 2013). The lower transfer factors observed in accessions JAK and DID indicate that they could not transfer the heavy metals effectively to their shoots. This is unlike of accession JIW whose transfer factor is much higher in Zn. Hence JIW could translocate Zn more effectively to the shoot.

Conclusion

This research showed that the Fonio accessions vary in their respective abilities to absorb and translocate each of the heavy metals studied. However, their mechanism of remediating heavy metals is by accumulating them in the roots while only few concentrations are in the shoots. Therefore the choice of fonio accession to be used for remediation will depend on the type of heavy metals involved. Accession JAK is thereby recommended for remediation purposes due to its better performance in accumulating all the heavy metals.

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