



## Growth and some heavy metals accumulation by vetiver grasses in lead-contaminated soil

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### ABSTRACT

Vetiver grass effectiveness for phytoremediation has received wide publicity. Therefore, a pot experiment was conducted to compare metal tolerance and metal uptake of two vetiver grass varieties; *Vetiveria zizanioides* (VZ) and *Vetiveria nigriflora* (VN) on soil collected from an abandoned Exide battery site. A pot experiment with treatments of different levels of pollutant; 100% top soil, 100% polluted soil, 75% top soil + 25% polluted soil, 25% topsoil + 75% polluted soil and 50% topsoil + 50% polluted soil. Each treatment consists of a pot filled with 5kg of the dumpsite soil and top soil mixture. These were planted with two vetiver grass varieties and were replicated three times. The vetiver grass was carefully uprooted twelve weeks after planting and was analyzed for lead concentration. Data collected were subjected to analysis of variance using SAS 2.0 and the means were separated using Least Significance Difference. The results showed that VZ had a better growth performance than VN. Number of tillers and root length were significantly higher for VZ (7.9, 36.8 cm) than VN (4.0, 23.3 cm), respectively. However, VN had higher uptake of lead ( $11017 \text{ mg kg}^{-1}$ ) than VZ ( $9405 \text{ mg kg}^{-1}$ ). In summary, both varieties may be well suited for phytoremediation in tropical lead mine areas, but VN could tolerate higher lead concentration than VZ.

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### Introduction

Soil contamination, a very important environmental problem, has attracted considerable public attention in recent decades (Garbisu and Alkorta, 2011). Metaliferous mining and processing, including the dumping of wastes, often produce severe heavy metal pollution (Baker *et al.*, 1994). Soils contaminated with heavy metal usually lack of long term or permanent cover. Barren soils are more prone to erosion and leaching which further spreads pollutants in the environment (Salt *et al.*, 1995). Establishment of a vegetation cover is thought to be most practical and economical and economical method for restoration of the mined land (Flathman and Lanza, 1998; Pangetal., 2003). However, revegetation of these areas is often difficult and slow due to the extreme soil properties, which include physical conditions, gross lack of nutrients and toxicity from residual heavy metals (Bradshaw, 1997).

The selection of appropriate plant species is very important to ensure a self sustaining vegetation cover (Wong, 2003). Two ecotypes of vetiver grass were selected in the present study, exotic (*Vetiveria zizanioides* (L) Nash) and a local type (*Vetiveria nigriflora* (L) Nash). Vetiver grass is a fast growing plant that tolerates various extreme environments, including pH values between 3.0 and 10.5 and temperature from -10 to 48°C (Dalton *et al.*, 1996; Wong, 2003). It also effectively controls erosion of soil by water (Truong and Baker, 1998). The previous studies have shown that vetiver grass can grow well in soils contaminated with multiple elements at high concentrations such as those found at coal, cadmium, and gold mining sites (Truong and Baker, 1998; Roong tanakiat and Chairaj, 2002) and can accumulate relatively high concentrations of lead (Chantachon *et al.*, 2004).

Phytoextraction is one of the phytoremediation techniques that is adopted to translocate and accumulate metals from contaminated soil in the above ground tissue of higher plants (Kumar *et al.*, 1995; Lasat 2002). Plant tissues harvested following phytoextraction are subjected to composting, compaction, incineration ashing, pyrolysis, or liquid extraction, and then they are sent to land fill site to reduce their harmfulness to human health (Sas Nowosi Eska *et al.*, 2004). The use of plants to remove metals from contaminated soil is a popular and satisfactory strategy because it enables the contaminated sites to be recovered to their natural condition.

The use of vetiver grass (*Vetiveria zizanioides* L.) Roberty, family poaceae,  $2n = 20$ ) in soil and water conservation has been suggested throughout 20<sup>th</sup> century. It is a versatile, fast growing hardy plant having wide climatic tolerance to extreme environments including soil pH ranging from 3.0-10.5 and temperature from 14-55°C. it accumulated  $\text{Pb}^{++}$ ,  $\text{Cd}^{++}$ ,  $\text{Hg}^{++}$ ,  $\text{Ag}^{++}$ ,  $\text{Zn}^{++}$ ,  $\text{N}^{++}$  and  $\text{Cu}^{++}$ , and tolerates many organic poisons making it valuable for decontaminated around mines (Roongtanakiat *et al.*, 2002).

Owing to its unique morphological, physiological and ecological characteristics such as its massive and deep root system, and its tolerance to a wide range of adverse climatic and edaphic conditions, including elevated levels of heavy metals, the interest in this grass has increased in recent years (Truong, 2000, Chen *et al.*, 2000). However, the use of vetiver grass in phytoremediation technology is not widely recognized owing to the lack of detailed investigations of its capacity to absorb contaminants and practical field applications.

The objectives of the present study were (1) to study the growth of the two vetiver grass eco-types; *Vetiveria zizanioides*

and *Vetiveria nigriflora*, and (2) to investigate the lead uptake ability of the two vetiver grasses.

#### Materials and Methods

The experiment which lasted for twelve weeks was carried out at Teaching and Research farm of Ladoke Akintola University of Technology (LAUTECH) Ogbomoso, Oyo State. The site has an elevation of 1147 m above sea level and it is situated at latitude  $8^{\circ}10'N$  and longitude  $4^{\circ}10'E$ . Soil samples used for the experiment were collected from an abandoned Exide battery manufacturing site and soil from land under fallow at the Teaching and Research farm. The two soil samples were air dried and made to pass through 2mm sieve. The soil samples from the battery manufacturing site were subjected to laboratory analysis for some chemical properties and some heavy metals concentrations (Table 1). The experiment was laid out in a randomized block design with three replications. The treatments consisted of soil samples that were thoroughly mixed up from Exide battery site and land under fallow at different ratios which give a total of 5kg per pot. The treatment consists of different levels of pollutants; 100% top soil (control), 100% polluted soil, 75% top soil 25% polluted soil, 25% topsoil + 75% polluted soil, and 50% topsoil + 50% polluted soil. The soil samples were planted with two vetiver grass varieties; *Vetiveria zizanioides* and *Vetiveria nigriflora*. Vetiver grass from pot was uprooted carefully to maintain roots intact at 12 weeks after planting.

#### Growth and metal accumulation

Plant growth was determined as plant height, number of tillers, wet biomass, and root length. To determine Lead accumulation in plants, dried plant samples were ground to a fine powder and sieved through 0.50mm mesh. The concentration of these heavy metals in plants were determined using 0.5kg plant sub-samples digested with  $HNO_3$  (APHA, 1998). The heavy metals were read on the atomic absorption spectrophotometer. The residual heavy metals in soil were determined by the same procedure.

#### Statistical analysis

The data for height, number of tillers, biomass, root length, lead concentration in soil and plants under different treatments were analyzed using the SAS statistical package by analysis of variance to compare the means of different treatments. Where significant F values were obtained, differences between individual means were tested using the least significant different test (LSD) at the  $p \leq 0.05$  significance level.

#### Results

##### Growth performance of *Vetiveria zizanioides* and *V. nigriflora* in polluted soil

The growth performance data (height, number of tillers (NOT) total wet biomass root length) of *V. zizanioides* (VZ) and *V. nigriflora* (VN) are shown in Table 2. Height of the plants significantly decreased with increase in levels of pollutant ( $P \leq 0.05$ ). 75% topsoil (TP) + 25% polluted soil (PS) was significantly highest in plant height than all other treatments. Although, VN had taller plant than VZ but the difference was not significant. However, VN gave significant higher total wet biomass (45.81g) than VZ (12.20g). In terms of NOT, VZ was significantly higher (7.87) than VN (4.00). Also, VZ had longer root than VN although the difference was similar.

##### Lead accumulation in plants

Lead accumulation in the two grasses varieties is presented in Table 3. *V. nigriflora* had higher accumulation of Pb than *V. zizanioides* in 25%TS + 75%PS, although no significant differences were observed among treatments.

##### Residual lead concentration in soil

The residual lead concentration in soil after the removal of the grasses after 12 weeks after planting is presented in Table 4. Pb remnant in soil was higher in soil grown with *V. zizanioides* (48041.00 mg/kg) than *V. nigriflora* (41764.00 mg/kg) although not significant. 100% PS gave significant higher lead content in soil (71983.00 mg/kg) than other treatments.

#### Discussion

The choice of plants is a crucial aspect of the practical application of phytostabilization based techniques on mine-degraded soils (Torroff *et al.*, 2000; Bleeker *et al.* 2002; Freitas *et al.*, 2004; Rizzi *et al.*, 2004). In terms of stabilizing metal contaminated sites, a higher metal concentration in phytoremediating plant is preferred, in order to reduce heavy metals in soil to a tolerable level for hygienic food production (Oshunsanya and Ewetola, 2011). From the present study, at highest application level of pollutant (100% PS), the two varieties *V. nigriflora* and *V. zizanioides* were able to grow well and showed no toxicity symptoms. This indicates that both varieties could tolerate high concentration of heavy metals. However, *V. nigriflora* had higher uptake of heavy metals which shows its superiority as phytoremediator over *V. zizanioides*. This may be as a result of its adaptability to the local ecology than the exotic *V. zizanioides*. The result contradicts some results (Wong 2003; Truong and Bakers. 1998) that showed superiority of *V. zizanioides* over other vetiver grass ecotypes.

The accumulation of heavy metals in plants vis - a - vis reduced its content in the soil. *V. nigriflora* had higher uptake than *V. zizanioides* which eventually reduced lead contents in the soil.

#### Conclusion

Both grass species, *V. zizanioides* and *V. nigriflora* are good choices for phytoremediation of lead mine soil. They show very high tolerance to lead, and more lead could be accumulated in the plants. The application of different levels of lead contaminated soils showed that *V. zizanioides* and *V. nigriflora* could survive under high lead toxicity. In summary, both varieties may be well suited for phytoremediation in tropical lead mine areas, but *V. nigriflora* could tolerate higher lead concentration than *V. zizanioides*.

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**Table 1: Soil chemical properties with some heavy metals concentration of the soil from Exide battery site**

Soil chemical properties with some heavy metals	Concentration
Pb (mg/kg)	133,000.0
Cd (mg/kg)	41.0
Zn (mg/kg)	528.0
Cu (mg/kg)	431.0
O.M (%)	0.8
P (mg/kg)	145.0
K (cmol/kg)	0.6
Ca (cmol/kg)	244.0
N (%)	0.2

Pb- Lead, Cd- Cadmium, Zn- Zinc, Cu- Copper, O.M- Organic matter  
P- Phosphorus, K- Potassium, Ca- Calcium and N- Nitrogen

**Table 2: Height (cm) number of tillers and biomass (g/fresh/pot) of *V. zizanioides* and *V. nigriflora* grown on different levels of soil pollutant from battery waste for 12 weeks.**

	Treatment	<i>V. zizanioides</i>	<i>V. nigriflora</i>	Treatment mean
Height	1(control)	67.33	101.00	84.17ab
	2	75.33	80.00	71.67b
	3	105.00	117.00	111.00a
	4	62.00	98.00	80.00ab
	5	82.33	97.00	89.67ab
	Mean	78.40a	98.60a	
Number of tillers	1	8.67	3.00	5.83a
	2	6.67	4.00	5.33a
	3	8.67	5.00	6.83a
	4	5.67	4.67	5.17a
	5	9.67	3.33	6.50a
	Mean	7.87a	4.00b	
Root length	1	40.67	36.33	38.50a
	2	32.53	16.00	24.27a
	3	39.20	39.17	39.18a
	4	38.33	8.77	23.55a
	5	33.00	16.17	24.58a
	Mean	36.75a	23.29a	
Biomass	1	10.57	24.44	17.41ab
	2	7.68	30.05	18.86b
	3	25.48	67.27	46.38a
	4	4.91	47.51	26.21ab
	5	12.38	59.96	36.17ab
	Mean	12.20b	45.81a	

Mean with different letter in the same row indicate as significant difference at 5% level according to LSD test 1- 100% Topsoil (TS); 2-100% Polluted soil (PS); 3-75%TS + 25%PS; 25%TS+75%PS; 5-50%+50%PS.

**Table 3: Effects of different levels of pollutant and two vetiver grass varieties on lead uptake (mg/kg).**

Treatment	<i>Vetiveria nigriflora</i>	<i>Vetiveria zizanioides</i>	Treatment means
1	865.83	680.50	773.00d
2	14293.00	16770.00	15532.00ab
3	8443.00	4633.33	6538.00dc
4	20793.00	17881.67	19338.00a
5	10686.66	7060.00	8873.00bc
Variety mean	11017.00a	9405.00a	
LSD variety (ns)			
LSD treatment (17.67)			
LSD variety*treatment (ns)			

Mean with different letter indicate significant different at 5% level according to LSD test.1- 100% Topsoil (TS); 2-100% Polluted soil (PS); 3-75%TS + 25%PS; 25%TS+75%PS; 5-50%+50%PS

**Table 4: Residual effects of different levels of pollutant and two vetiver grass varieties on lead concentration (mg/kg) in soil**

Treatment	<i>Vetiveria nigritana</i>	<i>Vetiveria zizanioides</i>	Treatment means
1 (control)	944.17	1022.67	983.00c
2	66633.33	77333.33	71983.00a
3	35540.00	59246.67	47393.00b
4	53260.00	66866.67	60063.00ab
5	52440.00	35734.67	44087.00b
Variety mean	41764.00a	48041a	
LSD variety (ns)			
LSD treatment (17.67)			
LSD variety*treatment (ns)			

Means with different letter indicate significant difference at 5% level according to LSD test. 1- 100% Topsoil (TS); 2-100% Polluted soil (PS); 3- 75% TS + 25%PS; 25%TS+75%PS; 5-50%+50%PS.

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