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The phytoextraction of Cu and Zn by elephant grass (*Pennisetum purpureum*) from tropical soil 21 years after amendment with industrial waste containing heavy metals

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Abstract. Increased soil heavy metal concentrations are suggested to cause roots to work harder. This research was to study the growth and phytoextraction behaviors of elephant grass in soil amended with industrial waste. Soil samples were obtained from an experimental field treated with a heavy metal containing waste at 0, 15 and 60 Mg ha⁻¹, CaCO₃ at 0 and 5 Mg ha⁻¹, and compost at 0 and 5 Mg ha⁻¹. Soil samples were planted with elephant grass, 8 weeks after which the soil samples were analyzed for Cu and Zn. Plant roots and shoots were harvested and weighed for their dry-masses and analyzed for Cu and Zn. The results demonstrate that the Root/Shoot increased and show good correlations with the increase in soil Cu or Zn. The plant Cu or Zn increased with the increase in soil Cu or Zn but decreased with liming. Plant Cu and Zn in roots and the whole plants as well as their TFs were well correlated with soil Cu and Zn. These observations confirm that the root/shoot growth and Cu and Zn absorption by elephant grass are governed by soil Cu and Zn and elephant grass is a Cu and Zn phytoextractor.

1. Introduction

Heavy metal is one of the most important toxic substances that may affect plant growth and production. Research results show that some plants are significantly depressed by elevated concentrations of heavy metals in soils [1,2]. However, several food and weed plants may adapt to extreme concentrations of soil heavy metals by absorbing and accumulating more heavy metals, more frequently more in their roots than in shoots [2-4,6]. It was suggested that plant roots show higher internal tolerance to high heavy metals than do plant shoots and also produce some exudates such as low molecular organic acids [5]. These plants may in some cases be utilized as good agents to clean up soils contaminated by heavy metals [5,7-12], particularly those growing fast with high biomass production and high absorbing capacity with respect to heavy metals [13,14]. Some of these plants may include amaranth, sunflowers, cabbage, chickpea, willows, broccoli, lettuce, and spinach, and several weeds like elephant grass [13-17].

Elephant grass or Napier grass (*Pennisetum purpureum*) is among weed plants reported to have the above characteristics [13,14]. This weed plant has been investigated for phytostabilization, phytoextraction, and phytoremediation of heavy metal contaminated soils [2,6,15,16]. The method of phytostabilization, phytoextraction and phytoremediation uses plants to extract, neutralize, accumulate, and reduce contaminants from the soil, water or air [18]. For example, [13] report that elephant grass was a very good extractor for Zn, better than for Cr, Cu, and Pb. [2] shows that EDTA enhanced the



phytoextraction of Pb, Cd, Cu, and Zn by elephant grass. [19] suggests that the plant species used in phytoremediation should have a rapid growth potential and free of pests and diseases, must be competitive to less desirable species, must be adaptable to local soils and climatic conditions, and also must be able to grow in infertile soils.

Further studies on the importance of this weed plant for phytoremediation of heavy metal contaminated soils are needed, particularly in tropical soils. This research was aimed to study the root/shoot growth and heavy metal phytoextraction behaviors of elephant grass in tropical soil of Ultisol in South Lampung Indonesia about 21 years after amended with high Cu- and Zn-containing industrial waste.

2. Materials and methods

This research was a plastic house experiment. Soil samples were taken from an experimental field set in July 1998 located in the village of Sidosari Natar South Lampung Indonesia (5°20'14.1"S 105°14'39.2"E) (Figure 1). The experimental design and treatment were previously reported [1,20]. The soil in the field was factorially treated with industrial waste at 0, 15, and 60 Mg ha⁻¹, lime at 0 and 5 Mg ha⁻¹, and organic compost at 0 and 5 Mg ha⁻¹, replicated 3 times. The soil was an Ultisol with textural properties 41.2% sand, 26.0% silt, and 32.8% clay (Textural Class was Sandy Clay Loam); pH 5.11, organic C content 1.28 g kg⁻¹, and Cu and Zn 1.28 and 1.60 mg kg⁻¹, respectively [1,20]. The waste was an industrial waste of metal-spoon industry. The waste contained high concentrations of Cu and Zn and was obtained from PT Star Metal Wares Jakarta. Some chemical properties of the industrial waste were pH 7.30, Pb 2.44 mg kg⁻¹, Cd 0.12 mg kg⁻¹, Cu 754 mg kg⁻¹, and Zn 44.6 mg kg⁻¹ [1,21]. The lime was calcite (CaCO₃). The organic compost was made of the cassava (*Mannihot utilisima*) leaf as described in [1].

For purpose of this research, soil samples were collected only from 6 experimental plots, i.e. Control Plots (no Industrial Waste, with or no Lime, and no Organic Compost), low heavy metal plots (with 15 Mg Industrial Waste ha⁻¹, with or no Lime, and no Organic Compost), and high metal plots (with 60 Mg Industrial Waste ha⁻¹, with or no Lime, and no Organic Compost) (Table 1). Plots with 5 Mg Organic Compost ha⁻¹ was not used because the effect of organic compost was not observed at >20 years after amendment. Soil samples were compositely collected using auger at 0–20 cm from 5 sampling sites in each plot measuring 400 cm × 450 cm. Soil samples of the same treatments were thoroughly mixed after air drying and sieving to 2 mm before being used in this experiment.

Five kg of air-dry (oven-dry equivalent, 24 hours 105°C) soil sample put into a plastic pot was used as the planting medium for elephant grass seedlings. Three similar seedlings were planted in each medium after the planting medium was moistened using tap water to approximately 40%. The growth of elephant grass was maintained and observed for 8 weeks.

At the end of the 8-week growth period, the weed plants were harvested. Plant shoots were cut right at the surface of the planting medium. Roots were carefully separated from soil mass using tap water. The plant biomasses were then oven-dried at 60°C for 3 × 24 hours and weighed for their dry-biomasses and analyzed for their contents of Cu and Zn using the method of wet-ashing involving the use of the iCE 3,000 Atomic Absorption Spectrophotometer (AAS). Soil sample was also taken from each planting medium and analyzed for Cu and Zn concentrations using the DTPA method [22]

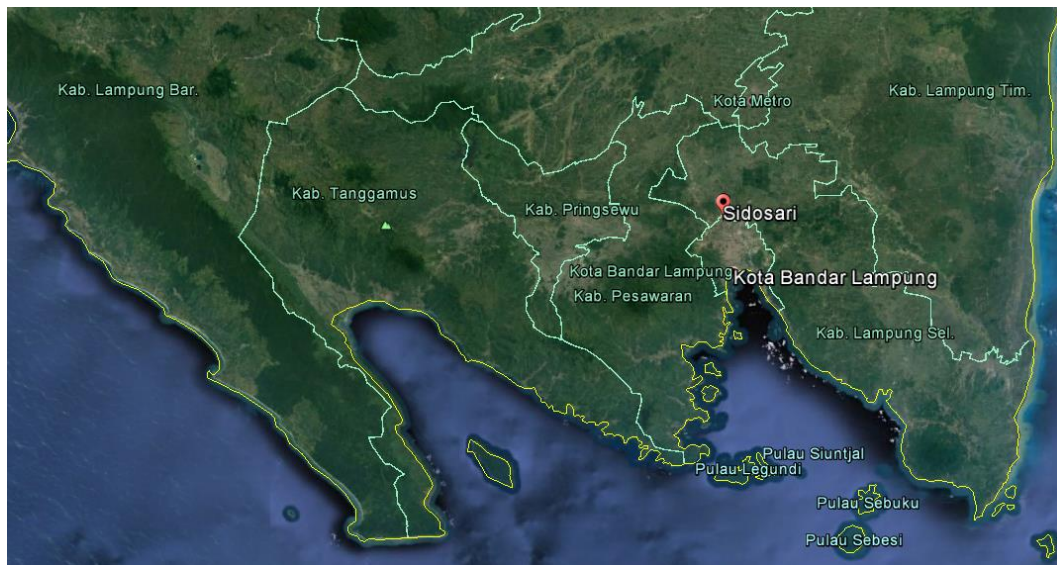


Figure 1. The google location of the experimental plot in Sidosari set up in 1998.

Table 1. The soil samples used in this experiment.

Soil sample ^a	Treatment in July 1998		
	Industrial waste ^b	Lime ^c	Organic compost ^d
 Mg ha ⁻¹		
Unlimed Control	0	0	0
Limed Control	0	5	0
Unlimed Low Heavy Metals	15	0	0
Limed Low Heavy Metals	15	5	0
Unlimed High Heavy Metals	60	0	0
Limed High Heavy Metals	60	5	0

^aSampled in July 2019; ^bMetal-Spoon Industry, ^cCaCO₃, ^dCassava-Leaf Compost

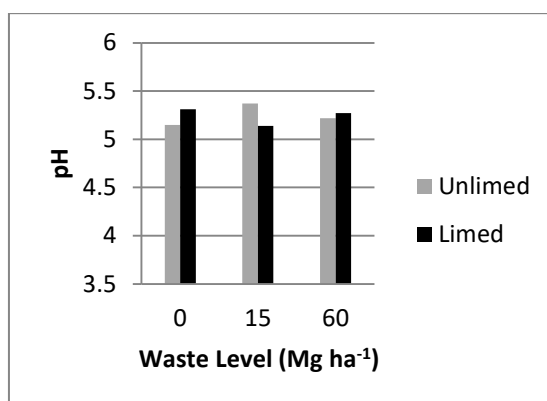
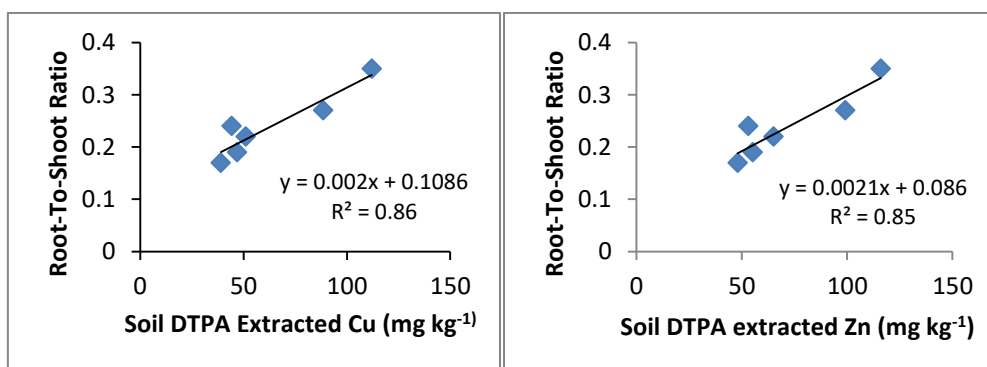
3. Results and discussion

The plant biomasses (roots and shoots) of elephant grass weighed after 8 weeks are presented in Table 2. Shown by the Root-To-Shoot Ratio, it is clear that the weights of plant roots are much lower than those of plant shoots in accordance with that reported by [13]. The growth of roots was significantly affected by industrial waste and/or lime. In general, plant root weight increased significantly in the presence of industrial waste and decreased slightly by lime treatment. This phenomenon clearly shows that the increase in the soil Cu and Zn concentration due to waste treatment may have stimulated the more intensive growth of plant roots. Lime treatment, even though did not affect the soil pH after 21 years (Figure 2), may have lowered the concentrations of soil Cu and Zn, and thereby, lowered the stimulation effect on the growth of plant roots (Table 2). The stimulation of Cu or Zn on the growth of plant roots is clearly demonstrated by good and positive correlations between Root-To-Shoot Ratios and the soil concentration of Cu ($R^2 = 0.86$) or Zn ($R^2 = 0.85$) (Figure 3), that show that the growth of the plant root was stimulated by the increase in Cu and/or Zn concentration in soils.

Table 2. The biomass properties of *Penisetum purpureum* grown in soil 21 years after amendment with industrial waste containing heavy metals and lime.

Amendment		Plant Parts			
Industrial waste	Lime (CaCO ₃)	Roots	Shoots ^a	Roots/shoots	
Mg ha ⁻¹		g polybag ⁻¹			
0	0	1.90	9.45	0.19	
	5	3.57	12.4	0.27	
15	0	4.02	17.7	0.24	
	5	2.66	15.8	0.17	
60	0	4.49	12.6	0.35	
	5	2.75	12.9	0.22	
LSD 5%		1.86		1.95	

^aNo interaction between industrial waste X Lime.

**Figure 2.** The reaction (pH) of soil 21 years after amendment with industrial waste and lime (lime level 5 Mg CaCO₃ ha⁻¹).**Figure 3.** The relationship between the the Root-to-Shoot Ratio and the soil DTPA extracted Cu and Zn.

The growth of plant roots is very important for elephant grass to adjust to the high concentrations of Cu and Zn and probably of other heavy metals. Higher root weight may cause higher root cation exchange capacity (CEC) that may retain more heavy metal cations in the surface of plants roots so that less heavy metals may move to plant shoots. Higher soil CEC may then lower the stimulation on the growth of plant roots. Higher CEC can be attained by increasing soil pH [23,24]. Therefore, the lime treatment was observed to slightly decrease the Root-To-Shoot Ratio (Table 2). Plant roots also produce some exudates such as low molecular organic acids that may chelate heavy metal cations in soil solution and lower heavy metal effects on plants [2,5].

The effect of lime and its interaction with industrial waste on soil Cu and Zn is shown in Table 3 and Table 4. The effect of lime is significant on soil Cu and Zn when industrial waste was also given. In general, the higher soil Cu and Zn caused by the industrial waste treatment was lowered by lime treatment 21 years ago, particularly at high waste levels (Table 4). Lime may theoretically increase the process of precipitation and/or adsorption of heavy metal cations in soil at high pH [24]. Liming is of course not the single factor causing the decrease in Cu and Zn concentration. The decrease in heavy metal concentrations is also probably attributed to metal movement through several physical, chemical, and biological mechanisms for the last 21 years.

Table 3. Analysis of variance of the DTPA-extracted Cu and Zn in soil 21 years after amendment with industrial waste and lime.

Amendment	DTPA extracted Cu	DTPA Extracted Zn
Industrial Waste	*	*
Lime	*	Ns
Industrial Waste and Lime	*	*

*Significant, ns Non-Significant

Table 4. The DTPA-extracted Cu and Zn in soil 21 years after amendment with industrial waste and lime.

Amendment		DTPA-extracted Heavy Metals	
Industrial waste	Lime	Cu	Zn
Mg kg ⁻¹		mg kg ⁻¹	
0	0	46.9	55.2
	5	88.6	99.1
15	0	44.3	53.1
	5	39.0	48.1
60	0	112	116
	5	51.1	65.2
LSD 5%		13.0	13.8

The treatment of industrial waste and lime also significantly affected the accumulation of Zn in roots and shoots and Zn translocation factor but not the accumulation of Cu in root and shoot and Cu translocation factor (Table 5 and Table 6). Translocation factor is the ratio of absorbed heavy metal in shoot and that in root [17,25]. As those of the soil Cu and Zn, the accumulation of Cu and Zn in plant roots and plant shoots increased with industrial waste treatment, more significantly in shoots. The accumulations were higher in shoots than in roots (Table 6). Unlike that suggested by [4] and [3], plants exposed to metals retained a major portion of metal in the shoots, which do not have a higher tolerance than roots for high internal metal concentration. This data clearly show that elephant grass is a Cu and Zn phytoextractor (TF > 1.00).

Table 5. Analysis of variance of heavy metal absorption by *Pennisetum purpureum* from contaminated soils 21 years after amendment with industrial waste and lime.

Amendment	Absorbed Cu			Absorbed Zn		
	Roots	Shoots	^a TF	Root	Shoots	^a TF
Industrial Waste	ns	ns	ns	*	ns	*
Lime	ns	ns	ns	ns	ns	*
Industrial Waste and Lime	ns	ns	ns	*	*	*

^aTF (Translocation Factor) = Absorbed Metal in Shoots/Absorbed Metal in Roots

Table 6. The absorption of Cu and Zn by *Penisetum purpureum* from soils 21 years after amendment with industrial waste and lime.

Amendment		Absorbed Cu ^a			Absorbed Zn		
Industrial waste	Lime	Roots	Shoots	TF ^b	Roots	Shoots	TF ^b
Mg ha ⁻¹		□g polybag ⁻¹			□g polybag ⁻¹		
0	0	27.7	55.5	2.00	12.2	93.8	7.69
	5	84.6	75.7	0.89	65.3	184	2.94
15	0	38.8	60.7	1.56	34.7	88.7	2.50
	5	28.8	63.3	2.17	27.0	120	4.35
60	0	115	86.4	0.75	88.6	110	1.23
	5	36.9	73.8	2.00	35.1	77.7	2.17
LSD 5%					32.9	56.5	6.67

^aNo interaction of Industrial Waste X Lime; ^bTF (Translocation Factor) = Absorbed Metal in Shoots/Absorbed Metal in Roots

In addition to be affected by industrial waste, the metal accumulations in plant biomasses were also lowered by lime treatment, well-correlated with their concentrations in soil. Table 7 shows that the correlation coefficient between the Cu and Zn accumulation in roots and the soil Cu and Zn are all relatively high (0.98 and 0.90, respectively); and those for metal in the whole plants are both 1.00 for Cu and Zn, respectively; and those of metal translocation factors are 0.95 and 0.98 for Cu and Zn, respectively. These data indicate that Cu and Zn in soils govern their accumulation in roots than that in shoot or total in roots and shoots but not that in shoot alone.

Table 7. The correlation coefficients between the plant accumulation and the soil DTPA-extracted Cu and Zn.

Element	Plant accumulation			TF ^a
	Root	Shoot	All plant parts	
Soil Cu	0.98	0.76	1.00	0.95
Soil Zn	0.90	0.25	1.00	0.58

^aTF (Translocation Factor) = Absorbed Metal in Shoots/Absorbed Metal in Roots

4. Conclusions

The Root-To-Shoot Ratios increased and show good correlations with the increase in the soil DTPA-extracted Cu or Zn, showing that the plant biomasses were distributed greater in plant roots at higher concentration of heavy metals to cope with their toxic environment but lower in plant roots with soil liming. The plant accumulation of Cu or Zn increased with the increase in the soil DTPA-extracted Cu or Zn resulted from waste treatments but decreased with lime treatment. Plant accumulation of Cu and Zn in roots and the whole plant roots and shoots as well as their translocation factors (in general TF > 1.00) are well correlated with their respective concentrations in soil ($r^2 > 0.90$). These observations confirm that the root/shoot growth and heavy metal absorption by elephant grass are governed by the concentrations of heavy metals in soils and elephant grass is a Cu and Zn phytoextractor.

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