

THE SURVIVAL OF FOUR TROPICAL PLANTS ON SOILS ARTIFICIALLY POLLUTED WITH TOXIC LEVELS OF ZINC

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Abstract: The rise in the incidence of heavy metal pollution in soils has been linked to increased anthropogenic activities which include mining and smelting operations, industrialization, agricultural processes and disposal of heavy metal-containing compounds. Heavy metal polluted sites encourage the depletion of soil nutrients and productivity, erosion, desertification and the developments of brown field lands. Phytoremediation is a technique of reclaiming unproductive and polluted soils by the use of plants which can either hyper-accumulate the heavy metals from the soils to their shoots or convert the heavy metals to forms which are less available or less toxic to plants. This present study was aimed at screening for plants that can grow in soils artificially contaminated with toxic levels of zinc. *Sida acuta*, *Axonopus compressus*, *Andropogon gayanus* and *Cyperus difformis* were grown on soils polluted with 0, 1150 and 2300 mg Zn kg⁻¹ soil for two weeks. The result showed that *Sida acuta*, *Axonopus compressus* and *Andropogon gayanus* tolerated the 1150 mg Zn kg⁻¹ soil level while only the *Axonopus compressus* survived the 2300 mg Zn kg⁻¹ soil level during the two weeks observation period. Our result reveals the great potential of using *Axonopus compressus* in reclaiming soils heavily polluted with zinc.

Keywords: Phytostabilization, Phytoremediation, Zinc tolerance, Zinc pollution, Heavy metals, *Axonopus compressus*.

Introduction

Zinc is a metallic element widely distributed in nature, making up about 0.02% of the earth crust (Irwin, 1997). It occurs naturally as ores of zinc salts commonest of which include, zinc sulphide, zinc carbonates and zinc oxide. Zinc is present in relatively low and non-toxic amount in most agricultural land but occur as rich deposit in certain areas. In low quantities, zinc is beneficial to plants and animals serving as essential micronutrients. It functions in gene expression, DNA Stabilization, and also as an important cofactor to several metalloenzymes (Frassinetti *et al.*, 2006). It is also reported to function in immune system

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(Rink and Haase, 2007) and control of oxidative stress (Zhou *et al.*, 2002). Zinc becomes toxic or lethal when plants and animals are exposed to high levels of it.

The need to utilize zinc for the production of brass, non corrosive alloys, white pigments, fertilizers, fungicides and drugs has stimulated human interest in mining zinc from sites of natural rich deposits of zinc ores (Eisler, 1993). These anthropogenic activities of man, in addition to erosion, leaching and other agricultural and industrial practices eventually distributes zinc from sites of rich deposits to agricultural lands and water bodies, hereby exposing plants and animals to zinc toxicity. Areas around zinc-lead mines have been reported to have very high levels of zinc (Struckhoff, Stroh and Grabner 2013). When exposed to suitable environmental conditions such as acidic rain, some of the zinc is leached into underground waters (Gibson and Pollard, 1988). Areas heavily polluted with zinc are often barren with very scanty or no vegetation. Such areas are easily prone to erosion which results in the distribution of toxic levels of zinc to other agricultural land and water bodies, hereby making such lands and water bodies toxic to living organism (Struckhoff *et al.*, 2013). Continuous use of zinc containing pesticides, fertilizers, and municipal biosolids on agricultural lands results in the eventual accumulation of zinc in such lands hereby making the land less productive to plants and potential source of zinc pollution to nearby water bodies or underground water (Fernández-Luqueño *et al.*, 2013). Improper management of industrial zinc-containing wastes also provides an additional route for the zinc pollution of the environment (Fernández-Luqueño *et al.*, 2013).

Several techniques have been used recently to reclaim lands polluted with zinc and other heavy metals. Some of such techniques includes the excavation and deposition of the top soils of polluted areas in special land fill sites (Conder, Lanno, and Basta 2001; Jing, He, and Yang 2007); the overlaying of polluted area with good soils (Neilson, Artiola, and Maier 2003); adding soil amendment agents to polluted soils hereby keeping the polluting metal in a stable form that are not readily leached, eroded or absorbable by plants (Lone *et al.*, 2008); the insitu chemical treatments and washing of polluted top soils (Neilson *et al.*, 2003) and more recently, bioremediation, which involves the use of living organism to extract or stabilize the polluting metals (Lone *et al.*, 2008).

Phytoremediation is the use of plants to absorb (phytoextraction, phytovolatilization, rhizofiltration etc) or stabilize (phytostabilization) polluting metals and other contaminants from soils or water (Lombi *et al.*, 2001). Phytoremediation has been reported to be a cheaper,

efficient, and environmental friendly way to reclaim lands polluted with zinc and other heavy metal (Jadia and Fulekar, 2008).

A good candidate plant for phytoremediation must be able to accumulate, tolerate and/or possess the capacity to detoxify high metal concentration in their tissue (Jadia and Fulekar, 2008). Several plants that can tolerate and accumulate high levels of metal in their tissues have already been identified, however, more efforts are still ongoing to identify more efficient phytoremediative plant species (USDA, 2000).

This work was aimed at assessing the potential of four common tropical plants to survive in soils artificially spiked with toxic concentrations of zinc sulphate solution. Such plants that can tolerate toxic levels of zinc would have a great potential of being used for phytoremediating soils heavily polluted with zinc (Langer et al, 2009).

Materials and methods

Soil preparation

Dry loam soils were collected within a depth of 5-20cm layer of the top soil from an uncultivated portion of a farmland within Nnamdi Azikiwe University, Awka, Nigeria. The soils were sieved through a stainless-steel 2 mm mesh sieve. The pH of the soil was determined with a suntex pH meter from a 1:2 suspension of the sieved soil with distilled water. The soils (2kg each) were bagged in black plastic bags, with depth and diameter of 15cm by 12cm respectively.

Plant materials

The plants (*Sida acuta*, *Axonopus compressus*, *Andropogon gayanus* and *Cyperus difformis*) were randomly sampled from an uncultivated farmland site of Nnamdi Azikiwe University Awka. Nine healthy plants of each species with height that spans between 15cm -40cm were carefully uprooted from their natural habitat within the study area. They were transplanted in the morning hours into already prepared bags of soil, one plant per bag. The potted plants were watered adequately for two weeks after the transplant to allow for acclimatization of the transplants to their new environment.

Experimental setup

After two weeks of growing on the untreated pots of soil, each of the potted plants was divided into three (3) groups. Group one, two and three contains 0, 1150 and 2300 mg zinc kg⁻¹ of soil respectively. The zinc was introduced in the form of an aqueous zinc sulphate solution. The potted plants in each of the treatment groups were saturated with sufficient amount of distilled water to facilitate the equilibration of the zinc salt across each pot. Each

of the treatment groups were done in triplicate. The pots were arranged in a completely randomized manner in a green house under continuous aeration. The plants were watered adequately throughout the additional two weeks observational period.

Assessment of toxicity and percentage survival

The plants were observed physically for survival and evidence of zinc induced toxicity. Zinc induced toxicity is detected by the presence of yellow and reddish coloration on the leaves. A plant is considered to have a 0% survival if all the 3 replicates died and a 100% survival if all the three replicates survived during the two weeks period of observation.

Results and Discussion

The value obtained for the soil pH was 5.05. Metals are usually more bioavailable for absorption by plants at low pH. It has also been reported that the bioavailability of metals immediately after spiking as metal salt is usually very high (Langer et al., 2009). These facts suggest that the applied zinc salts solutions were readily bio-available to the plants.

The amount of zinc applied to the soil was such that it is within the range often detected around many zinc mines sites. Igwe *et al* (2014) reported that a Pb/Zn mine site in Nigeria contains between 75–1878 mg Zn kg⁻¹ soil. Gibson and Pollard (1988) detected between 103-16139 mg Zn kg⁻¹ soil around picher oklahoma mine site. Struckhoff *et al* (2013), reported up to 2300 mg Zn kg⁻¹ soil around mine wastes. Mico *et al* (2006), detected between 33.4-80.7 mg Zn kg⁻¹ soil in agricultural land in Spain, while Vanita *et al* (2014), reported a zinc level of 73-320 mg Zn kg⁻¹ soil in agricultural land in India. Some sensitive plants have been reported to die when exposed to zinc levels above 100 mg kg⁻¹ soil (Eisler, 1993; Langer *et al.*, 2009).

The observed physical features of the plants in the different treatment groups are presented in table 1. The result showed that *Axonopus compressus* looks healthy when exposed to 1150 mg Zn kg⁻¹ soil whereas the other plants exhibited mild to intense leaves chlorosis at the same concentration of zinc. *Cyperus difformis* at 1150 and 2300 mg Zn kg⁻¹ soil levels were observed to wither after few days of exposure. Chlorosis and red pigments were observed on the leaves of *Axonopus compressus* exposed to the 2300 mg Zn kg⁻¹ soil. Chlorosis of leaves is an indication of zinc toxicity (Reichman, 2002). Chlorosis and discoloration of leaves has been linked to mineral deficiency by the plant due to the excessive presence of one or more mineral salt (Prasad, 2013). Production of anthocyanins is a mechanism of zinc tolerance in plants exposed to toxic levels of zinc; this often results in reddish coloration on leaves (Reichman, 2002). Though the heavy metal concentrations in the leaves of the plants were not

investigated, the observed leaf chlorosis is suggestive that some of the contaminating metals have been translocated to the shoots of the plants (Soudek *et al.*, 2014).

The percentage survival of the different plants grown on 0, 1150 and 2300 mg Zn kg⁻¹ soil is presented in table 2. The result shows that *Cyperus difformis* has 0% survival at both the 1150 and 2300 mg Zn kg⁻¹ soil. *Axonopus compressus* has a 100% survival at 1150 and 2300 mg Zn kg⁻¹ soil. Both *Andropogon gayanus* and *Sida acuta* has above 60% survival at the 1150 mg Zn kg⁻¹ soil but with no survival at the 2300 mg Zn kg⁻¹ soil. The observed survival of the *Axonopus compressus* which was exposed to the 2300 mg Zn kg⁻¹ soil and the observed reddish coloration on its leaves suggests the possibility of an innate mechanism of zinc tolerance in *Axonopus compressus*. MacFarlane and Burchett (2002) reported 100% mortality in mangrove *avicennia marina* plants that were exposed to 1000 mg kg⁻¹ sediment zinc concentration. *Andropogon virginicus* has been previously reported to have a high tolerance for zinc (Gibson and Pollard, 1988). Good phytostabilizing plants should tolerate high levels of metals and immobilize them via root uptake (Matthews, Moran, and Otte 2004). The increased metal tolerant capabilities of *Sida acuta*, *Andropogon gayanus* and *Axonopus compressus* makes them suitable candidates for re-colonization and stabilization of sites contaminated with zinc (USDA, 2000).

In conclusion, this study has shown that *Axonopus compressus*, *Sida acuta*, and *Andropogon gayanus* can survive up to 2300, 1150 and 1150 mg of zinc contamination per kg of soil respectively. This ability reveals the great potential of using them in stabilizing and re-colonizing soils heavily polluted with zinc. However, further studies would be necessary to ascertain the metal accumulating capabilities of the studied plant species as well as their mechanism of resistance to toxic levels of zinc. Furthermore, this study presents a simple, quick and cost-effective procedure for primary screening of plants for phytoremedial potential.

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Tables**Table 1: Observed physical features of the plant**

| | <i>Axonopus compressus</i> | <i>Andropogon gayanus</i> | <i>Sida acuta</i> | <i>Cyperus difformis</i> |
|--|--------------------------------|---------------------------|------------------------|--------------------------|
| Control (0 mg Zn kg ⁻¹ soil) | healthy | healthy | healthy | healthy |
| Group 2 (1150 mg Zn kg ⁻¹ soil) | healthy | Slight Leaf chlorosis | Slight leaf chlorosis | Withered |
| Group 3 (2300 mg Zn kg ⁻¹ soil) | Leaf chlorosis, reddish leaves | Intense leaf chlorosis | Intense leaf chlorosis | Withered |

Table 2: Percentage survival of the plants

| | <i>Axonopus compressus</i> | <i>Andropogon gayanus</i> | <i>Sida acuta</i> | <i>Cyperus difformis</i> |
|--|----------------------------|---------------------------|-------------------|--------------------------|
| Control (0 mg Zn kg ⁻¹ soil) | 100% | 100% | 100% | 100% |
| Group 2 (1150 mg Zn kg ⁻¹ soil) | 100% | 67% | 100% | 0% |
| Group 3 (2300 mg Zn kg ⁻¹ soil) | 100% | 0% | 0% | 0% |