

RESEARCH ARTICLE

Performance and remediation potential of *Chrysopogon aciculatus* (Retz.) Trin. grown in nickel-contaminated soils

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Abstract: The performance and remediation potential of *Chrysopogon aciculatus* (Retz.) Trin. grown in nickel contaminated soil was assessed. Six contamination regimes of 50, 100, 150, 200, 250 and 300 mg Ni/kg soil and control were set up in three replicates each. *Chrysopogon aciculatus* established from tillers were allowed to grow six weeks before data collection started. Data were collected weekly on ground cover, chlorophyll index and concentrations, total carotenoids and yield at maturity. Soil Ni concentrations were determined at pre-plant and post-harvest stages. Ground cover was not significantly different among the treatments. Ni contaminated treatments had the highest chlorophyll index at 10 and 12 weeks after planting. Chlorophyll a concentration was highest in 150 mg Ni/kg treatment (T₁₅₀) at 12 WAP. T₁₅₀ also had the least Chlorophyll b concentration at 12 WAP. T₂₀₀ and T₂₅₀ had low total carotenoids at 12 WAP. Dried shoot and root from T₃₀₀ had the highest Ni concentrations. Grass grown in T₂₀₀ accounted for the highest soil Ni uptake (% Ni remediated) with 96.4%. Soil Ni contents were reduced in all treatments. It is concluded that *C. aciculatus* can tolerate Ni contamination in soil and therefore can be used as a turf grass on Ni polluted soils.

Keywords: Contamination, heavy metal, performance, Port Harcourt grass, remediation, tolerance.

INTRODUCTION

Heavy metals are naturally present in the soil but geologic and anthropogenic activities may increase their concentrations to levels that are harmful to both plants and animals (Raskin *et al.*, 1994; Chibuike and Obiora, 2014). Some of these activities include mining and smelting of metals, burning of fossil fuels, use of fertilizers and pesticides in agriculture, production of batteries and other metal products in industries, sewage sludge, and municipal waste disposal (Alloway, 1995; Shen *et al.*, 2000). Heavy metals are elements that exhibit metallic

properties such as ductility, malleability, conductivity, cation stability, and ligand specificity. They are characterized by relatively high density and high relative atomic weight with an atomic number greater than 20 (Raskin *et al.*, 1994). Some of the heavy metals such as Cd, Pb and Hg are toxic to living organisms while heavy metals such as Co, Cu, Fe, Mn, Mo, Ni, V, and Zn are required in trace amounts for proper functioning of the cell (Tchounwou *et al.*, 2012).

Nickel, the 24th most abundant element in the Earth's crust, comprise about 3% of the composition of the earth. As a member of the transition metal series, it is resistant to corrosion by air, water and alkali, but dissolves readily in dilute oxidizing acids (Young, 1995). Nickel is a nutritionally essential trace metal for at least several animal species, micro-organisms and plants, and therefore either deficiency or toxicity symptoms can occur when too little or too much Ni is taken up, respectively. Although Ni is omnipresent and vital for the physiological functioning of cells in many organisms, concentrations in some areas from both anthropogenic release and naturally varying levels may be toxic (McGrath and Zhao, 2003). High Ni concentration in the soil affect germination of seeds, retard root and shoot, decrease branching and luxuriance, deform various plant parts including flower, decrease biomass production, induce leaf spotting, disturb mitotic root tips, and produce Fe deficiency that leads to chlorosis and foliar necrosis. Additionally, excess Ni also affects nutrient absorption by roots, impairs plant metabolism, inhibits photosynthesis and transpiration, and causes ultrastructural modifications (Ahmad and Ashraf, 2011). Soluble Ni compounds are often absorbed by plant roots passively (through a cation transport system) while chelated Ni compound are taken up through active-transport-

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mediated means, using transport proteins such as permeases. Insoluble Ni compounds primarily enter root cells through endocytosis. Once absorbed by roots, Ni is easily transported to shoots via the xylem through the transpiration stream and can accumulate in neonatal parts such as buds, fruits, and seeds (Ahmad and Ashraf, 2011).

Chronic exposure to Ni has been connected with increased risk of lung cancer, cardiovascular disease, neurological deficits, developmental deficits in childhood and high blood pressure (Chervona *et al.*, 2012). Therefore, the cultivation of crops in Ni-contaminated sites is unsafe, as the metal can be re-introduced into the food chain (Ahmad and Ashraf, 2011). Considering the adverse effect of Ni pollution in the environment, the need to for remediation should be emphasized.

Remediation of metal-polluted soils by physical and chemical methods such as solidification, stabilization, soil washing and flushing are often expensive and may render the soil unsuitable for plant growth (Marques *et al.*, 2009). Biological remediation using plants (phytoremediation) and/or micro-organisms is found to be an useful application (Chaney *et al.*, 1997; Glass, 1999). Although heavy metals cannot be degraded during phytoremediation, they are transformed from one organic complex or oxidation state to another. Due to a change in their oxidation state, heavy metals can be transformed into either less toxic, easily volatilized, more water soluble (and thus can be removed through leaching), less water soluble (which allows them to precipitate and become easily removed from the environment) or less bioavailable (Garbisu and Alkorta, 1997; Garbisu and Alkorta, 2003).

Turf grasses have been used in remediating metal-polluted soils (Jadia and Fulekar, 2008; Danh *et al.*, 2009; Zhang *et al.*, 2010; Subhashini and Swamy, 2014) and notable species including *Festuca arundinacea* (tall fescue), *Spartina patens* (salt meadow cord grass), *Eremochloa ophiuroides* (centipede grass), *Buchloe dactyloides* (buffalo grass) and *Cynodon dactylon* (Bermuda grass) have been tested for their metal-remediating potentials (Qu *et al.*, 2003; Rathi *et al.*, 2011).

Chrysopogon aciculatus (Retz.) Trin. is a perennial grass with creeping rhizomes (Paria

and Chattopadhyay, 2005). The plant is commonly called lovegrass (English) and Port Harcourt grass (Nigeria). It is popular for its good turf, especially with low mowing (Royal Botanic Gardens, 2005). It is usually found in sunny, dry, exposed areas such as roadsides, lawns, pasture, river banks and water courses (Noltie, 2000). The grass is fairly drought tolerant (FAO, 2016), persisting almost throughout the year and with great potentials to spread quickly over open areas using its creeping rhizomes. It can tolerate grazing, mowing and trampling by animals (Kabir and Nair, 2009). Ambasta and Rana (2013) identified *C. aciculatus* as a good soil binder that prevents soil erosion and difficult to eradicate when fully established. Oyediji *et al.* (2014) assessed the performance of *C. aciculatus* for turf in a field trial and reported the species' adaptation to low mowing. Documentation on the heavy metal uptake by *C. aciculatus* is scarce. However, Garbisu *et al.* (2002) identified features including rapid growth rate, high biomass, and profuse root system in *C. aciculatus* which is shared by most phytoextractors.

Although new species are added to the list of grasses suitable for phytoremediation, the information on remediation potentials of *Chrysopogon aciculatus* is scarce. The present study aims to bridge this gap by assessing the performance and remediation potential of *Chrysopogon aciculatus* grown in nickel contaminated soil regimes.

MATERIALS AND METHODS

Experimental design and set-up

A pot experiment was conducted in the screen house located in the Botanical Garden, University of Ilorin, Ilorin, Nigeria. Ilorin lies within the southern guinea savanna. The soils are lateritic consisting of three layers classified as Luvisols or Alfisols. The predominant soil in the University estate is loamy sand with dark greyish brown (5YR3/2) colour (Ogunkunle *et al.*, 2016).

The soil used for the experiment was collected from the University Botanical Garden (N 08° 28' 53.3'', E 04° 40' 28.9''). The collected soil was air-dried for 7 days and sieved using a 2 mm mesh to remove the debris. The soil was homogenized before packing into the planting pots. A sample of 7 kg each of the sieved soil were weighed into plastic pots (0.40 m × 0.40 m × 0.25 m, length by width by depth)

perforated (to allow aeration) and underlaid with plastic trays (to collect and return percolates back to the soils). Nickel contamination levels were created using anhydrous nickel chloride (NiCl_2). Six contamination levels, 50, 100, 150, 200, 250 and 300 mg Ni per kg soil were prepared and denoted as T_{50} , T_{100} , T_{150} , T_{200} , T_{250} and T_{300} respectively. The control soils were without Nickel (T_0). All the treatments were arranged in a completely randomized design and replicated three times. The pots were watered for a week with distilled water to allow the metal to fully stabilize prior to planting of the grass.

In each pot, nine 2 cm tillers of *Chrysopogon aciculatus* (Retz.) Trin. (Port Harcourt grass) were planted equidistant to each other. Irrigation to field capacity was carried out every 2 days using distilled water.

Soil chemical analyses

Five replicate soil samples were collected for the determination of pH, organic carbon, total nitrogen, available phosphorus, sodium, potassium, calcium and magnesium. Soil pH of 1:2 soil - 0.01 M CaCl_2 mixtures was measured with electronic digital pH metre (Orion star pH Benchtop® by ThermoScientific). Organic carbon was determined according to the method by Vickery *et al.* (1995). Total nitrogen was determined by macro Kjeldahl method as adopted by Bremner (1965). Available phosphorus was extracted with Bray P2 solution prior to colour development by ascorbic acid and determination by spectroscopy (Bray and Kurtz, 1945). Exchangeable cations (Na, K, Ca and Mg) were extracted from soil using 1 N neutral ammonium acetate and the concentrations were determined using respective wavelengths.

Ground cover measurement

The ground cover of the turf was measured weekly, starting from the sixth week after planting using a 0.16 m² quadrat with a regular 7 × 7 grids. Percentage ground cover was determined by dividing the number of points (quadrat grids) touching grass species by forty-nine and multiplied by 100.

Turf colour/pigment determination

Turf colour in the treatments and control were assessed biweekly starting from sixth week after planting (6 WAP) using the chlorophyll index.

The concentrations of chlorophylls a, b and total carotenoids were also measured. Chlorophyll index was determined using Atleaf chlorophyll meter (FT Green LLC, USA). Chlorophyll a, b and total carotenoids were determined by soaking 25 mg fresh leaf samples in 7 ml of 100% acetone (Analytical grade) for 72 hours and reading the absorbance of the supernatant at wavelengths of 470 nm, 645 nm and 662 nm (Lichtenthaler, 1987). Concentrations (in $\mu\text{g ml}^{-1}$) of chlorophyll a, chlorophyll b, and total carotenoids (xanthophyll + β -carotene) in the leaf extract were determined using the equations:

$$\text{Chlorophyll a} = 11.24A_{662} - 2.04A_{645} \quad (1)$$

$$\text{Chlorophyll b} = 20.13A_{645} - 4.19A_{662} \quad (2)$$

$$\text{Total carotenoids} = 1000A_{470} - (1.9 \text{ Chl. a} - 63.14 \text{ Chl. b})/214 \quad (3)$$

Yield determination

At maturity (13 WAP), shoot and root parts were harvested in each pot separately, washed with distilled water, allowed to drain, before taking biomass measurements. Samples were then oven-dried at 80 °C to constant a weight to obtain dry weights. Shoot-root ratio (SRR) was determined by dividing Shoot dry weight (SDW) with root dry weight (RDW).

Determination of Ni in plant parts

The dried shoot and root materials were then ground to fine powder (using ceramic mortar and pestle) and analyzed for nickel. Soil samples collected from each pot were air-dried, passed through 2 mm sieve and analyzed for nickel. Ni remediated from the soil was determined using the formula:

$$\% \text{ Ni remediated from soil} = \frac{\text{Initial concentration} - \text{Final concentration}}{\text{Initial concentration}} \times 100$$

Concentrations of Ni in shoot, root and soil were determined from nitric acid-perchloric acid digest of 0.5 g samples. The filtrate made to 25 ml with triple distilled water was read at 232.0 nm using atomic absorption spectrophotometer (Buck Scientific Model 210).

Data analysis

Data were analysed using analysis of variance (ANOVA) in SAS 9.1.3 to separate the means. Means were considered significant at $p < 0.05$.

RESULTS

Soil chemical properties

The soil pH was 7.48 ± 0.04 (slightly alkaline). Soil organic carbon and total nitrogen were $0.84 \pm 0.10\%$, $0.44 \pm 0.08\%$, respectively. The available phosphorus was 0.52 ± 0.13 ppm while exchangeable Na, K, Ca and Mg were 122.2 ± 2.22 , 195.1 ± 1.03 , 1248.0 ± 4.17 and 82.41 ± 2.09 ppm, respectively. Ni concentration in native soil was 0.18 ± 0.02 mg kg⁻¹.

Ground coverage

No significant difference ($p > 0.05$) was observed in the percentage ground cover of grass among treatments at 6 - 13 weeks after planting. There was weekly boost in the ground cover, with no consistence among treatments (Figure 1). The results of the present study suggested that the soil Ni contamination up to 300 mg kg⁻¹ had no significant effect on the growth of *C. aciculatus*.

Turf Colour/Pigmentation

Chlorophyll index (CI) for *C. aciculatus* turf in soils contaminated with different concentrations of Ni was not significantly different in all the

weeks after planting (Figure 2a). The CI values indicated that the grass was tolerant to the highest Ni concentration applied (300 mg kg⁻¹). There were significant differences in the foliar chlorophyll a (Chl. a) concentrations at 6 WAP ($p = 0.007$) and 10 WAP ($p = 0.015$). Foliar Chl. a concentrations in turf grown in soils contaminated with Ni were not significantly different from the control (T₀) at 8 WAP ($p = 0.335$) and 12 WAP ($p = 0.105$). (Figure 2b). The concentrations of foliar chlorophyll b (Chl. b) was significantly different for the treatments in the weeks after planting. Turf grown in T₁₅₀ had the significantly highest foliar Chl. b concentration at 6 WAP and 10 WAP but the least at 12 WAP. Turf in T₀, T₂₀₀, T₂₅₀ and T₃₀₀ had low Chl. b concentration at 6, 8 and 10 WAP. Turf in T₀, T₂₀₀ and T₂₅₀ had high concentration of Chl. b and were not significantly different at 12 WAP (Figure 2c). Total carotenoids in the foliage of *C. aciculatus* grown in the Ni-contaminated treatments were not significantly different from the control at 6 WAP ($p = 0.053$) and 8 WAP ($p = 0.174$), except for T₂₀₀ and T₂₅₀. The concentrations of total carotenoids in the turf of the control (T₀) varied significantly ($p = 0.008$) from those in the Ni-contaminated soils (except T₃₀₀) at 10 WAP. There was no significant difference in the concentration of Chl. b at 12 WAP ($p = 0.309$) (Figure 2d).

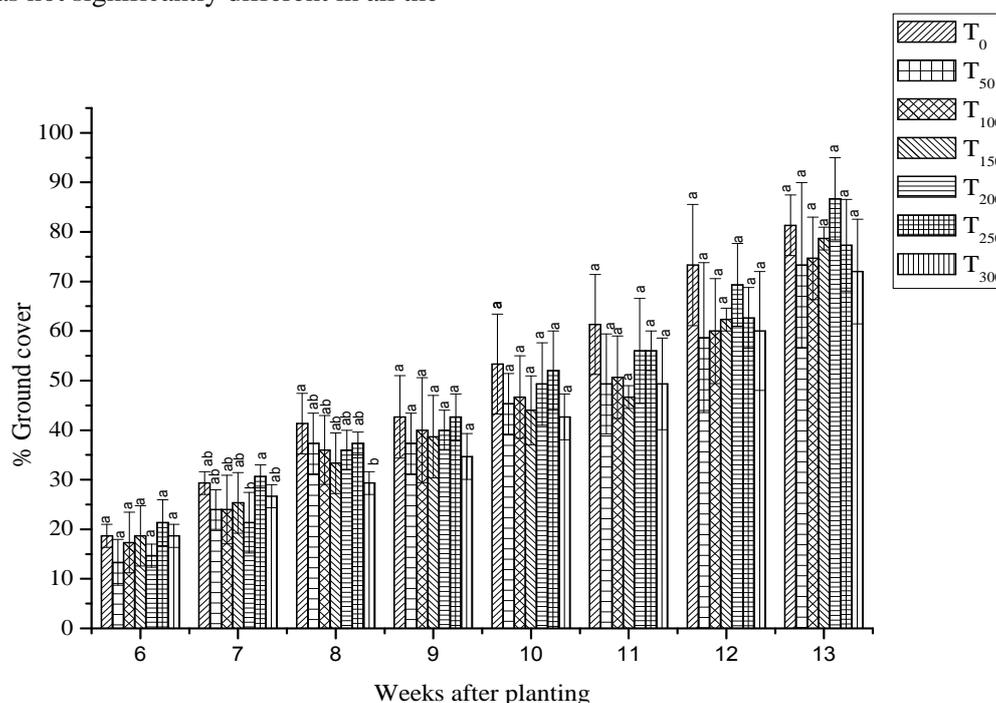


Figure 1: Percentage ground cover of *C. aciculatus* grown in soils contaminated with different concentrations of Ni.

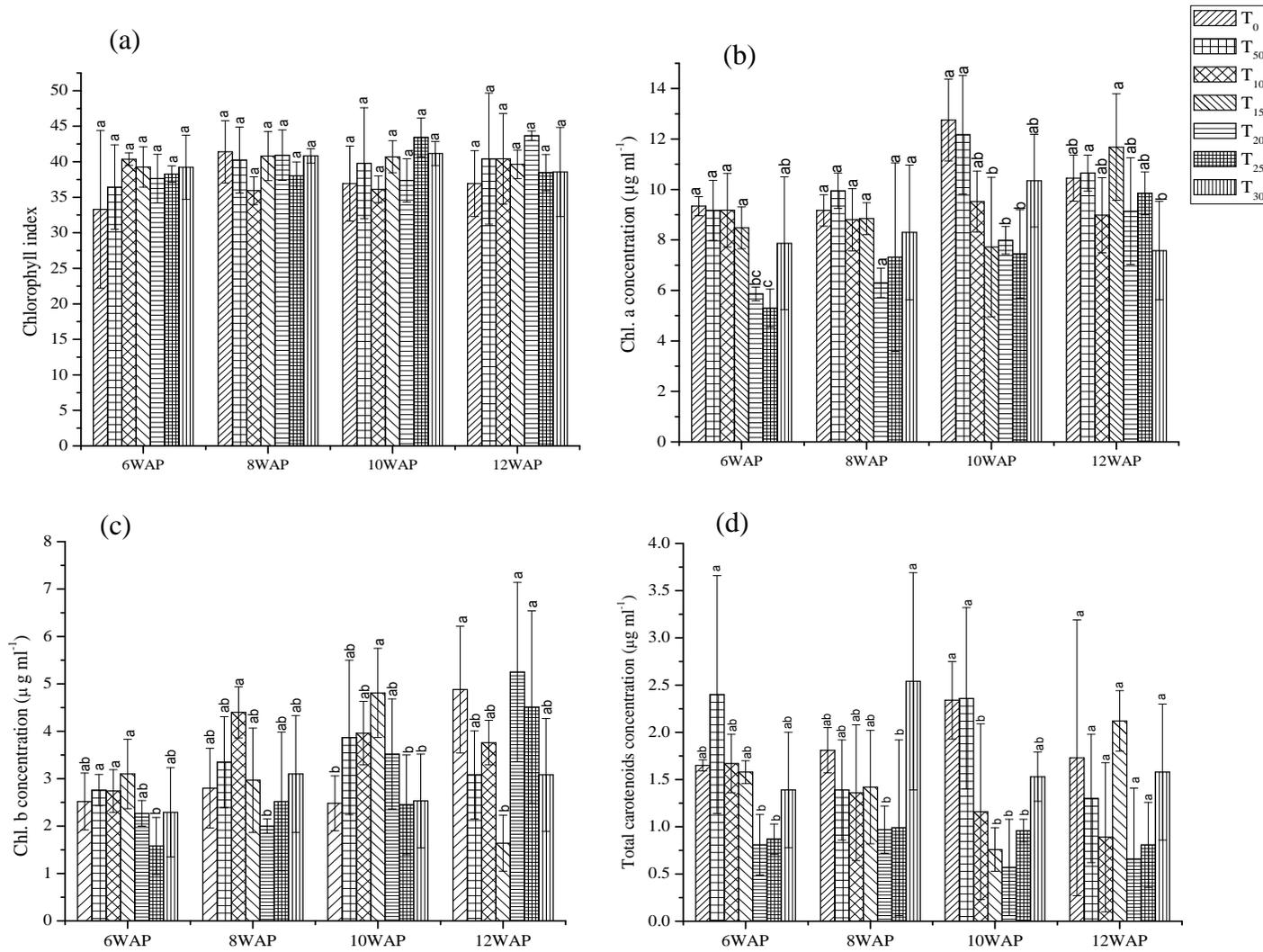


Figure. 2: (a) Chlorophyll index (b) chlorophyll a (c) chlorophyll b and (d) total carotenoids concentrations in *C. aciculatus* grown in soils contaminated with different concentrations of Ni.

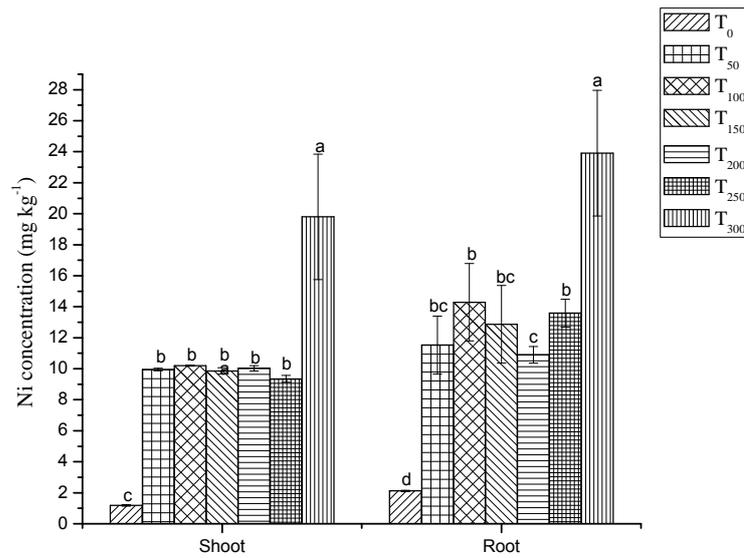


Figure 3: Ni concentrations in shoot and root of *C. aciculatus* grown in soils contaminated with different concentrations of Ni.

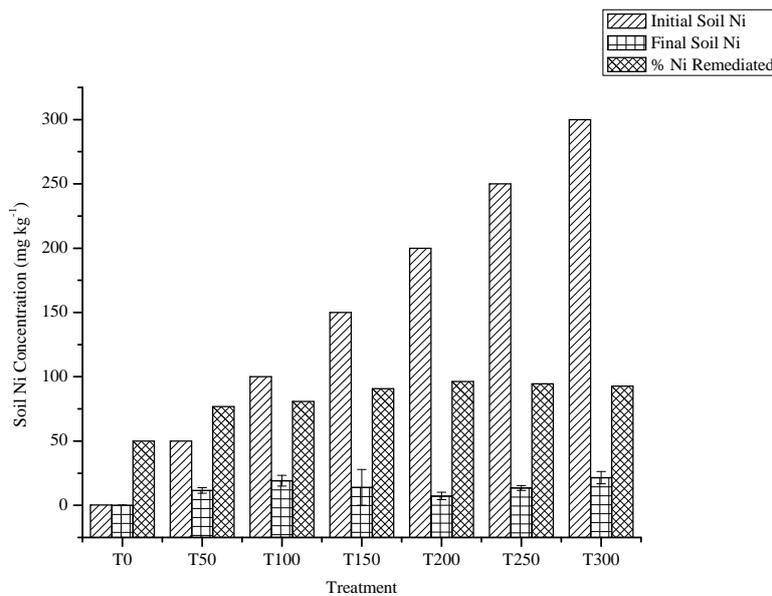


Figure 4: Ni levels in the soil before and after the remediation.

Yield parameters

The shoot fresh weight (SFW) of Ni-contaminated treatments were not statistically different ($p = 0.052$) between treatments but from the control (T₀). The root fresh weight (RFW) of all the Ni-contaminated treatments

were significantly different ($p = 0.004$) from the control. T₀ showed the highest RFW while T₁₅₀ was least. The shoot dry weight (SDW), root dry weight (RDW) and shoot-root ratio (SRR) were also not significantly different for the treatments (Table 1).

Table 1: Yield of *C. aciculatus* grown in soils contaminated with different concentrations of Ni.

Treatment	SFW (g m ⁻²)	RFW (g m ⁻²)	SDW (g m ⁻²)	RDW (g m ⁻²)	SRR
T ₀	334.37±22.86 ^a	273.73±5.21 ^a	107.86±6.10 ^a	113.33±6.13 ^a	0.94 ^a
T ₅₀	291.80±7.45 ^{ab}	184.07±4.19 ^b	88.17±2.67 ^b	100.73±3.95 ^{ab}	0.87 ^a
T ₁₀₀	265.07±31.84 ^b	179.00±23.59 ^b	86.90±9.88 ^b	74.63±6.27 ^b	1.16 ^a
T ₁₅₀	259.30±19.26 ^b	171.93±13.29 ^b	81.23±6.30 ^b	82.00±11.51 ^{ab}	1.00 ^a
T ₂₀₀	267.10±6.19 ^b	188.50±9.37 ^b	87.27±2.90 ^b	87.67±13.08 ^{ab}	0.84 ^a
T ₂₅₀	238.00±17.02 ^b	195.50±8.14 ^b	87.03±4.84 ^b	103.50±10.19 ^{ab}	0.84 ^a
T ₃₀₀	254.43±13.70 ^b	197.70±23.72 ^b	82.70±5.46 ^b	81.33±13.62 ^{ab}	0.99 ^a
p-value	0.052	0.004	0.100	0.127	0.582

SFW – shoot fresh weight, RFW – root fresh weight, SDW – shoot dry weight, RDW – root dry weight, SRR – shoot-root ratio. Means ± SD with the same letter(s) down a column are not significant at $p > 0.05$.

Uptake potential of Ni by *C. aciculatus*

Grass grown in T₃₀₀ showed significantly the highest Ni concentrations in shoot (19.80±4.05 mg kg⁻¹) and root (23.91±4.05 mg kg⁻¹) reflecting the plant's ability to absorb nickel. Concentrations of Ni in shoot and root materials for T₅₀, T₁₀₀, T₁₅₀, T₂₀₀ and T₂₅₀ were not significantly different ($p > 0.05$). The control (T₀) had the least foliar Ni concentrations. Generally, more Ni was stored in the root than shoot (Figure 3). There was reduction in the concentrations of Ni in soils across the treatments at the time of harvest. The highest Ni concentration at the time of harvest was recorded in T₃₀₀ (21.52±4.70 mg kg⁻¹) while the least was in T₀ (0.09±0.02 mg kg⁻¹). *C. aciculatus* grown in T₂₀₀ accounted for the highest soil Ni uptake (% Ni remediated) with 96.4%. Soil Ni concentration decreased by over 75% in all treatments except the control (T₀) (Figure 4).

DISCUSSION

The native soil pH was higher than that reported by FAO (2016) for optimum growth of *C. aciculatus*. Despite the disparity in soil pH, *C. aciculatus* seemed to tolerate the soil pH in the present study. The exchangeable cations in the native soil are relatively high. High soil cation exchange capacity (CEC) has been known to favour nutrient retention in turfs (Bigelow et al., 2001). Soil physicochemical properties also influence metal speciation and consequently, its mobility, bioavailability and toxicity (Irha et al., 2003). The speciation and sorption of Ni complexes in soil is dependent on the soil physicochemical properties. Sherene (2010) reported that heavy metals sorption increase with pH and organic matter composition.

The results of the present study suggested that the soil Ni contamination up to 300 mg kg⁻¹ had no significant effect on the growth of *C. aciculatus*. Brown et al. (1987) reported that Ni plays a vital role in the growth and development of plants and therefore in deficient soils plants metabolize Ni that is present in soils as contaminant. No visual symptoms of Ni toxicity were observed in all treatments. Although heavy metals are toxic at high concentration, their behaviour and fate are governed by a range of physicochemical processes in the soil or sediment system (Sherene, 2010; Olaniran et al., 2013). Plant tolerance to Ni toxicity has been associated with exclusion or influx/transfer of Cu, Fe, and Mn into roots (Yang et al., 1996). Nickel has been reported to produce beneficial effects in oats, wheat and tomato etc. (Brown et al., 1987; Seregin and Kozhevnikova, 2006). The metal has been documented to play an important part in many metabolic processes in plants including redox-processes, stabilization of molecules, components of various enzymes and in the regulation of osmotic pressure (Olaniran et al., 2013).

The results of CI in this study also showed that the grass was tolerant to the highest Ni concentration applied (300 mg kg⁻¹). Zhu et al. (2012) have reported that CI values directly correlated with leaf nitrogen and total chlorophyll contents. Contrary to chlorosis and necrosis reported by Rathor et al. (2014) in plants grown in soils contaminated with high concentration of Ni, *C. aciculatus* showed good health (CI > 35.0) throughout the study period, except in T₀ at 6 WAP. Soil Ni levels showed no direct relationship to concentrations of photosynthetic pigments in *C. aciculatus*. However, relatively higher chlorophyll a

concentrations over chlorophyll b and total carotenoids show that Chl. a contribute more in the photosynthesis. The result of this study is contrary to the findings of Singh and Pandey (2011) where nickel exposure decreased chlorophyll a, b and total chlorophyll contents in lettuce.

Nickel uptake by grass grown in T₃₀₀ was comparable to values reported (24.23 µg g⁻¹ dry of plant) by Syed *et al.* (2010) for *Eichhornia crassipes*. The Ni accumulation levels in shoots and roots of *C. aciculatus* were higher than the values reported by Netty *et al.* (2013) for other plant species such as *Tephrosia* sp., *Melastoma malabathricum*, *Mimosa pigra*, *Celtis occidentalis* and *Sarcotheca celebica*. Lasat (2016) reported that non-accumulators of micronutrients including Ni usually do not accumulate more than 10 mg/kg of the element. The present study showed that *C. aciculatus* is an excellent accumulator of Ni.

CONCLUSION

The results suggest that Ni contamination had no effect on the performance of *C. aciculatus* affirming its ability to endure in Ni contaminated sites. Higher photosynthetic pigments in plants exposed to high Ni concentrations also reflect the ability of the species to tolerate high Ni. Therefore, *C. aciculatus* can be used as a good candidate for remediating Ni contaminated soils. The present research further support the potential use of plants in restoring and remediating contaminated landscapes.

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