FULafia Journal of Science & Technology
ISSN (print) 2449-0954 \ ISSN (online) 2636-4972
https://lafiascijournals.org.ng/index.php/fjst
Sponsored By TERTIARY EDUCATION TRUST FUND
Manuscript => Received: July, 2025; Accepted: Sept, 2025; Published: Sept., 2025 https://doi.org/10.62050/fjst2025.v9n2.627

INFLUENCE OF HEAVY METALS ON THE GROWTH OF TWO COMMON WEEDS *Hyptis suaveolens* AND *Sida acuta* IN LAFIA, NASARAWA STATE

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ABSTRACT
An assessment of the influence of heavy metals on the growth of two common weeds *Hyptis suaveolens* and *Sida acuta* in soils of polluted areas and an unpolluted soil (control) in Lafia, Nasarawa State, Nigeria was carried out. Soil samples were collected in three replicates from an automobile workshop site (AWS), refuse dump site (RDS), and unpolluted soil (UPS) in Lafia. The growth of each plant was monitored in an interval of two weeks for a period of 8 weeks. The heavy metals investigated in the soil include Cu, Pb and Zn using digestion process and atomic Absorption Spectrophotometer (AAS). The heavy metal analysis revealed that AWS recorded highest concentration (0.71 mg/kg) while UPS recorded the least concentration of Pb (0.28 mg/kg) respectively. RDS had the highest concentration of Cu (0.61 mg/kg), while UPS had the highest concentration of Zn (0.19 mg/kg). It was observed that *Hyptis suaveolens* grew better on RDS and the growth parameters were seen to be significantly different from AWS and UPS. The AWS recorded least growth parameters which is likely due to the high level of metal concentration in the soil. Similarly, Sida acuta grew better on RDS and the least in AWS. The use of weeds which are not consumed by humans and animals should be encouraged in phytoremediation in order to prevent the effects of waste- related problems in the environment.

Keywords: Heavy metals, *Hyptis suaveolens*, phytoremediation, *Sida acuta*

INTRODUCTION
Heavy metals are naturally deleterious to non-tolerant plants resulting in several plant defects such as chlorosis, stunted growth, yield reduction, metabolic disorders and poor nutrient uptake (Anoliefo & Vwioko, 1995; Guala *et al*., 2010; Okonokhua *et al*., 2007). Soil is one of the natural resources that provide essential elements and interrelating functions which include; as a store for biodiversity, natural habitat for living organisms, food and biomass production as well as a relatively stable reservoir for the whole ecosystem. However, it can be easily deteriorated by both anthropogenic and natural changes (Department of Environment, Malaysia 2009).

Many polluted soils contain heavy metals like cadmium, lead, zinc, copper, cobalt and chromium. Phytoremediation is a set of ecological strategies that utilize plants, in situ, to promote the breakdown, immobilization, and removal of pollutants from the environment. For plants to be regarded as phytoremediators of these heavy metals, they must be able to grow massively in polluted soils.

American mint (*Hyptis suaveolens* L.) and Wire weed (*Sida acuta*) are very popular weeds which are found in Lafia, Nigeria. These plants are known for their high biomass production in polluted soils, road sides and abandoned sites /vegetation; this makes them potential candidates for removal of pollutants from the soil. Therefore this study is aimed at investigating the influence of heavy metal on the growth and development of the two common weeds *Hyptis suaveolens* and *Sida acuta*.

MATERIALS AND METHODS
Study area
The study area, Lafia is located on latitude 8.48^0 N and longitude 8.52^0 E, North central Nigeria, Nasarawa State (Figure 1). With a population of about 330,712 (NPC, 2012), it is home of abundant food crops that produce important fruits which meet the immediate needs and also serve as the major source of income for the inhabitants. The climate in Lafia is characterized by dry and rainy seasons.

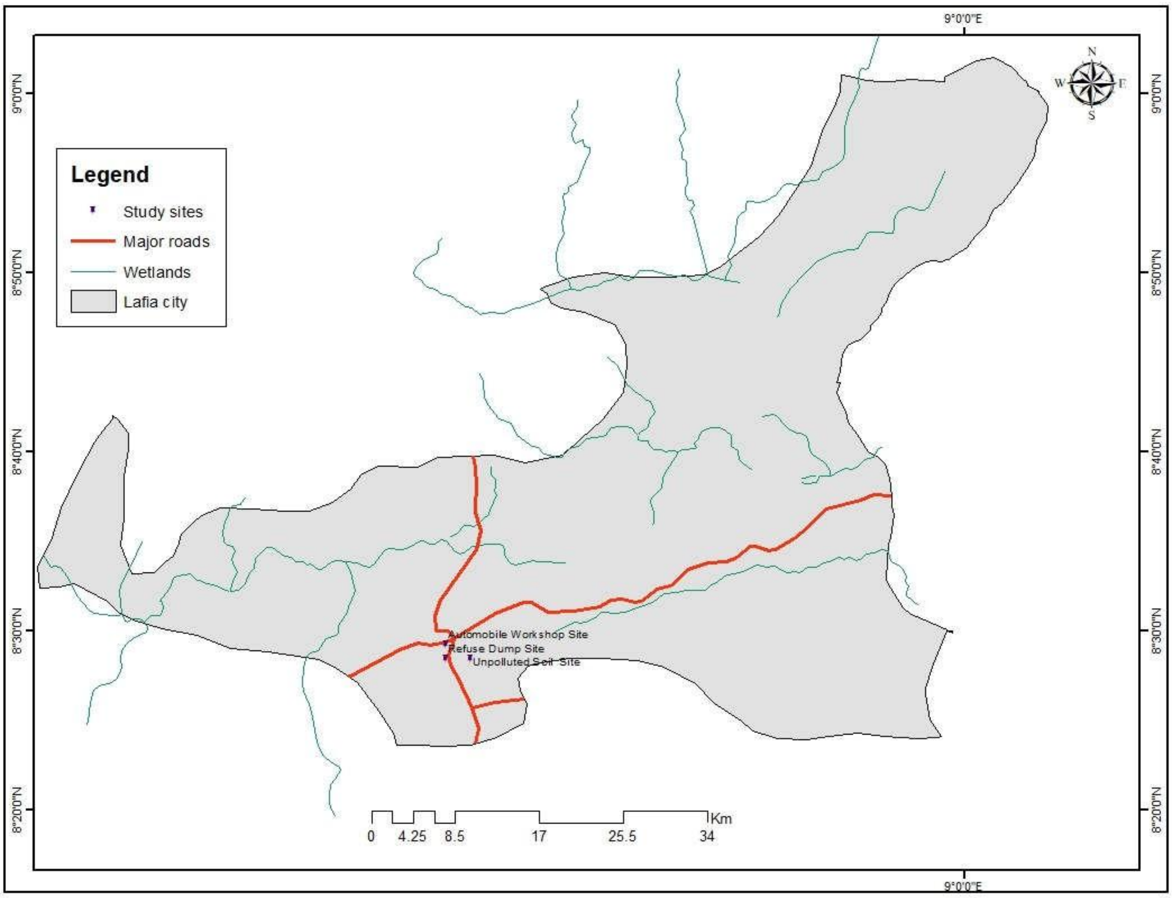

Figure 1: Map of the study site

Soil sampling

Soil samples were collected from three different areas in Lafia, Nasarawa State, Nigeria which included, Automobile workshop (AWS), refuse dump site (RDS) and unpolluted site (UPS) as control. The soil samples were collected in three replicates to give a fair representation of the various sites. The soil samples were collected at the depth of 0–15 cmeach. The soil samples were packaged in black polyethene leather bags and labeled accordingly for easy identification and transported to Muhammadu Buhari TETFund of Excellence (MBTCE) laboratory at Federal University of Lafia. The soil samples were air- dried separately at room temperature in the laboratory (McGrath, 2015).

Soil samples preparation

The soil samples collected from the field were air-dried, gently crushed with porcelain pestleand mortar (to reduce the size of soil lumps so as to increase their surface area for reaction), and sieved through 2 mm sieve mesh prior to analysis, core sampling method was used to collect soils for bulk density and moisture content determination. The soil samples were properly labeled stored in foil papers, for various types of analysis (Zauro, 2017).

Digestion of soil samples

The procedures of Ayodele and Gaya (1998) were used for the pretreatment of the soil samples. 1 g of air-dried soil sample was weighed into a beaker using analytical weighing balance (model: BL-P3/6002) (Eco Star), 20ml aqua-regia mixture was added (i.e., mixture of Hydrochloric acid (HCl) and Nitric acid (HNO_3), ratio 3:1). The mixture was digested on a hot plate at 70^0C till the solution became transparent. The resulting solution was filtered through Whatman filter paper No. 42 into 50 ml volumetric flask, distilled water was added to make up to mark (OAC, 2012). The filtrates were then transferred into 100 ml prewashed sample bottles and were analyzed using atomic absorption spectrophotometer (AAS) (model: AA320N) for elemental analysis at Muhammadu Buhari TETfund Centre for Excellence (MBTCE), Federal University of Lafia. The analysis was done in triplicate and results obtained were recorded.

Analysis of soil samples for heavy metal

The heavy metals concentrations in these samples were determined with the aid of a Buck Scientific model 210VGP Flame Atomic Absorption Spectrophotometer (FAAS). This was operated with a continuous source background correction. Working standards were prepared, after serial dilution of 1000 ppm metal stock solution in each case. Calibration curves were prepared for the elements individually. A blank reading was also taken, and necessary corrections were made during the calculation of concentration of various elements.

Experimental design

The study was a pot experiment conducted in a screenhouse at the Department of Plant Science and Biotechnology Federal University of Lafia. The setup was a Complete Randomized Design (CRD) with three replicate each from each site. Leather pots of 20 and 14 cm in height and diameter respectively were filled with 1.50 kg of air-dried soil samples. The seeds of the weeds which were collected from Federal University of Lafia campus permanent site were planted in the pots individually and allowed to germinate. The pots were watered twice daily (morning and evening) with tap water per pot. The growth parameters including plant height, number of leaves and leaf area of each plant were measured in every two weeks for a period of 8 weeks.

The leaf area was calculated by multiplying the length of leaves (L) and breadth of leaves (B) by a constant (2.325) by Osei-Yeboah *et al*., (1983)

i.e. Leaf Area (LA) = Length of leaf (L) x Breadth of leaf (B) x 2.325

Statistical analysis

The differences in the heavy metals concentrations in the soils between the contaminated sites and control were tested using non-parametric Kruskal-Walis test at $P \leq 0.05$. Differences in the morphological parameters between the soil treatments were determined using one-way ANOVA. The significant means were separated using Duncan Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Pre-experiment analysis of heavy metals in soil

As shown in Table 1, AWS was observed to have the highest Pb (0.71 mg/kg) which is significantly higher than the UPS (0.28 mg/kg), but it is not significantly different from RDS (0.56 mg/kg). RDS treatment was observed to have the highest Cu (0.61 mg/kg) but it is not significantly different from AWS (0.29 mg/kg) and UPS (0.47 mg/kg). UPS was observed to have the highest Zn (0.19 mg/kg) but it is not significantly different from AWS (0.14 mg/kg) and RDS (0.12 mg/kg).

Table 1: Heavy metal concentrations in the soil before the experiment

Treatment	Pb (mg/kg)	Cu (mg/kg)	Zn (mg/kg)
AWS	0.71	0.29	0.14
RDS	0.56	0.61	0.12
UPS	0.28	0.47	0.19

Morphological parameters of the plants after the experiment

The result of the morphological parameters of *Hyptis suaveolens* grown on the different soil types (Table 2) revealed that *Hyptis suaveolens* grown on RDS had the highest number of leaves (35.1) which is significantly different from those of AWS and UPS (13.63 and 17.55) respectively. The highest leaf area of the plant was also recorded at the RDS (56.94 cm^2) which is significantly different from AWS but not different from UPS. Highest shoot height (Table 2) was also recorded at the RDS (31.15 cm) which is significantly different from AWS (16.13 cm) and UPS (16.94 cm). It was observed that there was no significant difference between AWS and UPS in terms of shoot-height. Similarly, RDS (1.11 cm) recorded highest in stem girth and was observed to be significantly different from AWS (0.48 cm) and UPS (0.64 cm). It is however, worthy of note that there was no significant difference in stem girth between AWS and UPS.

Table 2: Growth parameters of *H. suaveolens* grown on different soil treatments

Treatment	Number of leaves	Leaf area (cm^2)	Shoot height (cm)	Stem girth (cm)
AWS	13.63 ± 3.73^a	31.25 ± 11.54^j	16.13 ± 3.54^a	0.48 ± 0.15^j
RDS	35.1 ± 4.33^b	56.94 ± 5.07^k	31.15 ± 1.05^b	1.11 ± 0.05^k
UPS	17.55 ± 2.45^a	35.18 ± 5.49^{jk}	16.94 ± 1.56^a	0.64 ± 0.13^j

Values represent mean ± S.E; Means with same superscript across same column are not significantly different while meanswith different superscript in same column are significantly different.

Table 3: Growth parameters of *S. acuta* grown on different soil treatments

Treatment	Number of leaves	Leaf area (cm^2)	Shoot height (cm)	Stem girth (cm)
AWS	2.07 ± 0.50^a	1.07 ± 0.54^j	2.14 ± 0.55^a	0.06 ± 0.01^j
RDS	3.52 ± 0.89^a	6.66 ± 2.22^k	4.78 ± 1.24^a	0.09 ± 0.02^j
UPS	3.03 ± 0.87^a	3.80 ± 1.88^{jk}	2.35 ± 0.68^a	0.05 ± 0.01^j

Values represent mean ± S.E; Means with same superscript across same column are not significantly different while means with different superscript in same column are significantly different

Sida acuta recorded highest number of leaves of (3.52) at RDS (Table 3) which is not significantly different from AWS and UPS (2.07) and (3.03) respectively. Similarly, RDS recorded highest in leaf area (6.66 cm^2) which is significantly different from AWS (1.07 cm^2) but not significantly different from UPS (3.80 cm^2). However, AWS and UPS are not significantly different from each other. Again, RDS recorded highest in shoot height (4.78 cm) as against AWS (2.14 cm) and UPS (2.35 cm) although means were not significantly different across the treatment. Similarly, in terms of stem girth, RDS recorded highest (0.09 cm) which is not significantly different from the other soil treatments.

The massive growth and development of *Hyptis suaveolens* on these heavy metal contaminated soils clearly shows that the plant is a good candidate for phytoremediation. The accumulation ability of heavy metals in plants is a factor of genetic make-up of the plant species and their efficiency is determined by the plant uptake or soil to plant transfer factors of the metals that are involved (Khan *et al.*, 2008). This was also buttressed by another study where the same plant was found at several other polluted soils in Lafia, Nigeria (Akomolafe & Lawal, 2019). Other studies have also affirmed its phytoremediation ability (Abdulhamid *et al.*, 2017; Sivakuma *et al.*, 2016), hence, its abundance within the study area. In the same vein, *Sida acuta* has been identified as a phytoremediator of heavy metals and as a plant adapted to growing in waste soils (Abidemi *et al.*, 2014; Tripathi & Misra, 2013).

Heavy metal concentrations in plants after the experiment

The result of the concentrations of heavy metals in *H. suaveolens* is presented in Table 4. UPS was observed to have the highest Pb concentration (0.65 mg/Kg) and is significantly different from that of RDS which is the lowest (0.07 mg/Kg). Similarly, the Cu concentration in UPS was observed to be the highest and is significantly different from those of AWS and RDS. However, AWS was found to have the highest concentrations of Zn (0.32 mg/Kg) which is significantly different from those of RDS and UPS.

Table 4: Concentrations of heavy metals in *H. suaveolens* after the experiment

Treatment	Pb (mg/Kg)	Cu (mg/Kg)	Zn (mg/Kg)
AWS	0.44^{b}	0.16^{a}	0.32^{a}
RDS	0.07^{c}	0.14^{a}	0.02^{b}
UPS	0.65^{b}	0.20^{b}	0.15^{c}

Values with same superscript across same column are not significantly different (p>0.05)

Table 5: Concentrations of heavy metals in *S. acuta* after the experiment

Site	Pb (mg/Kg)	Cu (mg/Kg)	Zn (mg/Kg)
AWS	0.10^{a}	0.02^{a}	0.12^{a}
RDS	0.38^{a}	0.32^{b}	0.15^{a}
UPS	0.13^{a}	0.07^{a}	0.27^{a}

Values with same superscript across same column are not significantly different (p>0.05)

The result of the concentration of heavy metals in s. acuta is presented in Table 5. RDS was observed to have the highest Pb concentration (0.38 mg/Kg) but is not significantly different from AWS and UPS. Similarly, the Cu concentration in RDS was observed to have the highest Cu concentration (0.32 mg/Kg) and is significantly different from AWS and UPS, however UPS was found to have the highest Zn concentration (0.27 mg/Kg) but it is not significantly different from AWS and RDS

The growth and massive development of *Hyptis suaveolens* and *Sida acuta* proves their hyperaccumulation potentials. Heavy metal profiles increased with increasing human activities. The concentrations of heavy metals in the soils is in the order of Pb > Cu > Zn (i.e. increasing order of concentration). Although these metals found in soils around the dump sites had a considerable amount, and were seen to be far above USEPA, FEPA and WHO permissibility. Lead was also found to be above the WHO standard maxima of 0.01 mg/kg. Lead accumulates in the upper 8 inches of the soil and is highly immobile. Its contamination is also long-term. Concentration of copper in the soil is within the permissible level. However, Zinc had lowest concentration compared to the rest of the metals, though it is still safe i.e. the values are below the tolerable levels of 90-300 mg/kg recommended by EC (1986), but in very high concentrations may pose danger to consumers of plants around these areas.

CONCLUSION

The study revealed the concentration of Pb to be the highest in the plants and soils. The heavy metal concentrations in both plants and soils at the polluted and control sites range from Pb>Cu>Zn. There were variations in the levels of the heavy metals from the different sampling sites. Phytoremediation using weeds that are not consumed by humans should be encouraged to prevent the effects of waste related problems in the environment.

Conflicts of interest: Authors report no conflicts of interest relating to the publication of this paper.

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