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Toxicity of Plastics in Soil: Influence on Soil Bacteria, *Aporrectodea Longa,* Nutrient and Phytochemical Properties of *Telfairia Occidentalis*

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Abstract

Problem: Over the years, there has been massive production of plastics which are usually discarded after use resulting in a spate of plastic pollution in the soil environment. The presence of these plastics and its contaminants elicit toxic effects on soil and soil biological sentinels. **Approach:** All Soil samples were collected in Ugbowo, Benin-City, Edo State, Nigeria which was the study area. The various samples collected were analyzed using standard methods. **Findings:** The results from this study revealed various physicochemical parameters and additives present in the test soil. It was observed that the test soil being the plastic composted soil was slightly acidic with a pH value of 5.62±0.05 compared to the control garden soil with a neutral pH 7.92±0.09. The additives detected in the soil were 40.07ng/g bisphenol A, 14.05ng/g di(2-ethylhexyl) phthalate (DEHP), 26.23ng/g dimethylpthalate (DMP) and 7.07ng/g diethylpthalates (DEP). Di-n-butylphthalate (DBP) and butylbenzylphthalate (BBzP) were below the limits of detection and only 3.01ng/g of bisphenol A was detected in the control garden soil. The plastic composted soil had low bacterial count of 0.43×10^2 cfu/g compared to the control soil with 27.0×10^4 cfu/g. A progressive reduction in the percentage survival of the earthworms with increased plastic contaminant concentrations was observed. Treatment B had lowest plastic concentration of 27.27 percentage survival followed by treatment C with 20.93 percentage survival. It was observed that the plastic composted soil served as a medium for the growth and survival of the *Telfairia occidentalis*. **Conclusion:** Polystyrene plastics could be used as manure to grow non-edible plants like flowers. It was observed that children can accumulate higher concentrations of these plastics in their body which is due to their low body weight. This further reveal that young children are vulnerable to the danger of plastic accumulation and toxicity.

Keywords: Plastics, Additives, Toxicity, Biological Sentinels, Soil

Introduction

With the advances in technology and the increase in the global population, plastic materials have been useful in every aspect of life and industries. However, most conventional plastics such as polyvinyl chloride, polyethylene, polyethylene terephthalate polypropylene, and polystyrene are non-biodegradable, and their increasing accumulation in the environment has been a threat to the planet.[1] Plastic wastes are present in soil in various forms with varying solubility and availability. Plastics bioavailability depends on its chemical behavior, soil properties and the individual characteristics of the receptor (organism or plant). Among soil properties pH, cation exchange capacity and redox potential play the most important role.[2] Polyethylene (PE) and Nylon 11 (NY11), polypropylene, polystyrene, polyvinyl chloride and polyethylene terephthalate are non-biodegradable which decreases their chances of entering into microbial cell, therefore they increase and accumulate in the environment with several adverse effects on soil sentinels. [3] Ecotoxicity of plastic waste can be evaluated with respect to their bioavailability and biodegradation. Microbes can only degrade plastics that are available for their metabolism. The constant release of these additives into the environment is a harmful practice. [4] Soil and other living organisms like bacteria are exposed to the released additives from plastic degradation. Additives like Phthalates, organotins, bisphenol A and others have been shown to target nuclear hormone receptor signaling pathways.[5] This release may take place during the service life of the plastics or after their disposal. For example, in landfills, both the landfill compartment and other potential receptors such as sediments represent complex environments with multiple chemical and biological processes occurring concurrently. In landfills, plastics are exposed to an extraction solvent in the form of acidic (pH 5–6) leachates with high ionic strength and neutral or alkaline leachates containing high-molecular-weight organic compounds. The different leachates have not only different potentials to extract and transport, but also different biological populations with the potential to degrade or transform the released additives. Plastics have shown to elicit toxic effects on soil and on several life forms especially in the marine environment where some of these additives from plastic have been found to act as hormone disruptors. [6, 7, 44] Plastic toxicity greatly depends on the following: physical and chemical properties of the plastic wastes, photo, thermal and oxidative degradation, and the nature of the surrounding medium. [8, 9, 10, 11, 12] To overcome all the dangers of plastic toxicity, production of plastics with high degree of degradability is encouraged. [13] However, this present study evaluates the impact of plastics on soil, earthworm, soil bacteria and fluted pumpkin.

Methodology

Collection of Soil Samples

Five (5) soil samples (500g each) were collected from different locations within the Edo State Waste Management Landfill site and one soil sample from a non-plastic contaminated soil at Ekosodin community in Ugbowo, Benin-City, Edo State, Nigeria which served as the control at a depth of 0-10cm with a standard soil auger in plastic bags. In the laboratory, the five soil samples from the landfill site collected earlier were merged together to form a composite sample. The soil samples were homogenized and kept on the laboratory bench to air dry. [14] The waste soil sample collected from the landfill site was used to isolate nitrifying bacterial while a portion of the garden soil was used to formulate specific concentrations of plastic-enriched composted soil with polystyrene plastic granules for earthworm and plant toxicity analysis.

Collection of Plant Seeds

Seeds of *Telfaira occidentalis* (fluted Pumpkin) were collected from Department of Crop Science, University of Benin, and Edo State, Nigeria.

Isolation of *Nitrosomonas* sp.

Winograsky plates were aseptically inoculated with 0.1ml of the appropriate dilution of the soil suspension using spread plate technique. The inoculums were spread over the entire surface of the solid agar with sterile glass rod. All the inoculated Petri dishes were incubated aerobically at room temperature (28°C) for 72h. *Nitrosomonas* sp. was isolated and confirmed. [15]

Collection of Earthworm (*Aporrectodea longa*)

The earthworms were collected from a farm land in Ugbowo Benin-City, Edo State. The worms were collected according to the method described by Spurgeon.[16] They were collected by digging and hand sorting from substances liters and were taken to the labouratory for identification. They were washed with water to remove soil particles and were left to on moist filter paper for voiding. Earthworms were selected based on their maturity (shown by the presence of clitellum) and liveliness (active response when anterior segment is prodded). The physicochemical parameters of the native soil were determined prior to the test. [17]

Soil Analysis

The soil samples were analyzed for the following parameters: pH, total organic carbon (TOC), moisture content and essential minerals: Nitrate, Phosphorus, calcium, magnesium, sulphate, potassium, sodium, and total hydrocarbon content. [18, 47]

G.C. Analysis of Soil Samples and leaves of *Telfairia occidentalis*

The waste soil sample and the garden soil sample were analyzed for the presence of plastic additives. The leaves of *Telfairia occidentalis* were also analyzed using gas chromatogram with mass spectrometry (GC-MS). These analyses were carried out at the National Agency for Food and Drug Administration and Control (NAFDAC) Kaduna, State, Nigeria. [15]

Preparation of Plastic Composted Soil Concentration for Toxicity Analysis

For the determination of the median lethal concentration (LC_{50}), plastic concentrations of 100, 200, 300, 400 and 500 mg/l were formulated by adding (100, 200,300,400 and 500 g) in 1000 ml of Winograsky medium respectively.[19] For the median effective concentration, the following plastic concentrations (20, 40, 60, 80 and 100 mg/l.) were formulated by adding (20, 40, 60, 80 and 100 g) in 1000 ml of Winograsky medium respectively. A control experiment consisting of Winograsky medium only (without the plastic composted soil) was set up. [19]

Nitrifying Bacteria Acute Toxicity Test

Winograsky medium which was fortified with several grams of plastic granules (100, 200, 300, 400 and 500 mg/l) and (20, 40, 60, 80 and 100 mg/l) respectively were inoculated with ten milliliters of bacteria (*Nitrosomonas* sp.) standard inoculum. They were allowed to stand for an hour for growth. One (1) ml of the suspension thereafter was plated from mineral salt media composted with different grams of plastic granules on a non-plastic composted winograsky agar plate. This was done for the all concentrations and repeated for 2, 3 and 4hrs interval. [20, 45] This was followed by nitrite determination from the various plastic composted mineral salt media after 1, 2, 3 and 4hrs which were incubated at room temperature (28+/-2^{o}C) for 24hrs. Percentage nitrite accumulation for *Nitrosomonas* sp. was plotted against the plastic concentration and the median effective concentration (EC_{50}) was determined using the probit analysis. The percentage inhibition of bacteria growth (log survival) was plotted against test concentration and the median lethal concentration (LC_{50}) value was determined using the probit analysis. [21, 46]

Chronic Toxicity of Plastic-Enriched Composting Waste on the Growth and Survival of Earthworm

Four concentrations (50, 100, 150, and 200 mg/4kg) of plastic-enriched composting soil were prepared using polystyrene plastic granules and 20 g of cellulose was added to the soil as food for the earthworms. Blank (control) containing cellulose, water and a non-plastic composted soil was also prepared. The distribution of individual earthworms among the test chambers was randomized. Death, weight loss and behavioral symptoms were the criteria used in this test guideline to evaluate the toxicity of polystyrene plastic on the earthworms. Each test and control chamber were checked for dead or affected earthworms and observations recorded on 7, 14, 21 and 28 days after the beginning of the test. [22] The experiment consisted of five (5) treatments; amounting to an aggregate of 5 experimental buckets. Each bucket is of 4 liters capacity and perforated at the base and set out on the field. The treatments are:

A= (Control), untreated soil
B= (0.83% treatments level)
C= (1.67% treatments level)
D= (2.50% treatments level)
E= (3.33% treatments level)

Effects of Plastic-Enriched Waste on the Growth and Survival of the Plants

Block design with three replicates were used to assess the impact of plastic composting waste on the growth and survival of *Telfairia occidentalis*. Garden soil samples were collected from a farm land and were divided into four (A, B, C, D) 4 kg each and three of them (B, C, D) were further fortified with different concentrations of (100, 150 and 200 g/4 kg) of polystyrene granules to form the plastic-enriched composting soil, while soil sample A served as the control without plastic granules. The plastic type used polystyrene plastic granules. The experiment consisted of four (4) treatments; amounting to an aggregate of 4 experimental buckets. Each bucket is of 4 liters capacity and perforated at the base and set out on the field. The treatments are:

A = Untreated 4kg composite soil sample (Control).
B = (2.5% treatments level)
C = (3.75% treatments level)
D = (5% treatments level)

Each treatment was thoroughly mixed in its allotted bucket at the beginning of the experiment. Percentage germination was observed, the height and stem girth of the plants were measured using a meter rule for eight weeks (Plate 1).

Plant Growth Analysis

Seed viability test was carried out prior to planting of the bean seeds by flotation method. The seeds were grown on the three different concentrations of the plastic-enriched composted waste soil (100, 150 and 200 g/4 kg) which was formulated using polystyrene plastic granules and on the control garden soil sample which a non-plastic composted soil. Plant growth was determined by monitoring the various growth parameters which are germination, height of the plants per week, number of leaves produced by each plant per week, stem girth which is the size or diameter of the stem per week and the weight of the plant per week also. The leaves of *Telfaira occidentalis* grown were harvested after six months, dried for two weeks and then used for the determination of the proximate and phytochemical constituent of the plant. [23]

Proximate and Phytochemical Screening of *Telfairia occidentalis*

The three sets of leaves of *Telfairia occidentalis* were collected from the three different concentration (100, 150 and 200 g/kg) of plastic enriched-composted waste soil and one from the control soil sample was air-dried for two weeks, mashed into powder and sieved. Fifty (50g) of each leaf was dispensed in 500 ml of distilled water in a liter conical flask. The mixture was vigorously stirred intermittently with a magnetic stirrer and then allowed to stand for 48 hrs. It was stirred the second time and filtered through a Whatman filter paper-lined funnel into a conical flask. The filtrate was evaporated at 40^oC with a water bath to obtain the solid crude extract. The same procedure was carried out for ethanol extraction except that the crude solid extract which was obtained by concentrating the filtrate with a rotary evaporator. All extracts obtained was stored in a refrigerator until required for use. The extracts of both leaves were analyzed for its nutrient and phytochemical properties using standard procedures. [24, 25]

Human Health Risk Assessment of Plastic Residues in *Telfairia occidentalis*

To estimate the carcinogenic and non-carcinogenic risk of detected plastics to humans, using two population groups (young children and adults). [26] The per capita consumption of *Telfairia occidentalis* in Nigeria is 285g while body weight for young children was set at 28.1kg and 70 kg for adult population group. [27]

Discussion

The results from this study revealed various physicochemical parameters and plastic additives present in the test soil. It was observed that the test soil being the plastic composted soil was slightly acidic with a pH value of 5.62±0.05 compared to the control garden soil with a neutral pH 7.92±0.09. This acidic pH observed in the test soil could be as a result of the release of metals from the degradation of various additives in the soil. The test soil sample had higher value for electrical conductivity, heavy metals, and Total carbon compared to the control garden soil sample (Table 1). According to Atuanya and Tudararo-Aheroba, [17] who assessed the ecotoxicological effects of petroleum refinery sludge on biological sentinels recorded high concentration of metallic ions in the slugde. It was observed that the plastic composted soil samples had low pH values than the control soil sample which corresponded with previous studies. [28] Ayolagha and Nleremchi, who worked on soil polluted by oil also observed an increase in the metal content of their test soil sample. [29, 47] Bingham *et al.,* also observed such trend while assessing heavy metal pollution in the surface soil region that had undergone intense industrialization. [30]

The following plastic additives were detected in the soil samples: 40.07ng/g bisphenol A, 14.05ng/g di (2-ethylhexyl) phthalate (DEHP), 26.23ng/g dimethylpthalate (DMP) and 7.07ng/g diethylpthalates (DEP). Di-n-butylphthalate (DBP) and butylbenzylphthalate (BBzP) were below the limits of detection and only 3.01ng/g of bisphenol A was detected in the control garden soil (Table 2). The total additive concentration for the test soil was 87.42ng/g and 3.02ng/g for the control garden soil sample. Additives in the soil are constantly released from plastic waste. The presence of these plastic contaminants can adversely affect soil fertility and soil fauna.[31] These additives elicit toxic effects on the soil and soil biological sentinels. Atuanya *et al*., had earlier revealed that nitrification process and other microbial based processes which enhance soil fertility may be hindered in an ecosystem polluted with plastics.[6]

Bacterial growth was observed in both soil samples. However, there was an increased bacterial count from the control soil sample. The growth followed a normal bacterial growth curve with a highest count of 27.0 x 10^4 cfu/g. On the other hand, a retarded growth was observed in the plastic composted soil sample where growth was observed from day 3 with its highest count of 0.43 x 10^2 cfu/g (Figure 1). It was observed that bacterial growth was inhibited in the test soil which was acidic which collaborated with Okpokwasili and Odokuma, who also observed a similar trend in their assessment of the ecotoxicological impact of petroleum refinery oily sludge. [20,32, 33] It was also observed that the toxicity of plastic composted soil on the *Nitrosomonas* sp. depended on the contact time and plastic composted soil concentrations which corroborated with Ibiene and Okpokwasili, who assessed the toxicity of different insecticide concentrations on Nitrifying bacteria. [19, 47] There was a sharp decrease in the EC_{50} values with time (Fig. 1B) while the LC_{50} values increased with time. This shows that at low plastic composted soil concentrations the bacteria were able to adapt and oxidize ammonia which increased with time. Also at higher plastic composted soil concentration, the bacterial growth and metabolism were slowly retarded which is as a result of the inhibition of enzyme activities by the additives in the plastic composting soil. [34, 35]

A progressive reduction in the percentage survival of the earthworms with increased plastic composted soil concentrations was observed (Table 3). Treatment B with the lowest plastic composted soil concentration had 27.27 percentage survival rate followed by treatment C with 20.93 percentage survival. Earthworms did not survive in both treatments D and E with 150g/4kg and 200g/4kg concentration respectively. It was observed that there was a reduction in the weight of the earthworms after they were treated with various concentrations of plastic composted soil except for the control sample. Metals concentration and other soil properties appear to have influenced the high mortality and weight loss of the earthworms in the treated soil which corroborated with VanGester *et al.,* who observed the impact of several toxicants on earthworms. [36] This result suggests that weight loss in earthworms is a more sensitive parameter than survival in assessing pollutant effects on soil. This is so because earthworms were more sensitive and susceptible to metal pollution than any other groups of soil invertebrates. [37] High concentrations of additives and metal contamination can affect the density, viability, growth and sexual reproduction in earthworms. [38] The high concentration of pollutants in the test soil explained the avoidance and mortality as well as the weight loss in the earthworms.

It was observed that the plastic composted soil served as a medium for the growth and survival of the *Telfairia occidentalis*. This means that the plant was able to use the polystyrene plastic as a source of carbon. Three weeks after germination, the treatments with polystyrene plastic granules especially that of the 200g treatment seems to be healthier than the control treatment. This was because leaf coloration of the treatments with plastic granules were greener than the control treatment; there were no wilting of leaves from the treatments with plastic granules as compared to the control treatment where some of the leaves experienced wilting. Although the treatment with plastic granules had the higher percentage growth with respect to height and stem gilt especially the treatment with 200% plastic granules (Table 5, 6, 7 and 8). However, statistically there was no significant difference in the values obtained from the plant growth parameters measured. Nevertheless, it was observed that the plants with the highest concentration of plastic concentrations performed better than the other treatments even that of the control and this indicates that the polystyrene plastic granules present in the soil samples was suitable rather than being toxic to the growth of *Telfairia occidentalis.* Contrarily to the report of Atuanya *et al.,* who evaluated the impact of plastic-enriched composting on soil structure, fertility and growth of maize plant.[18] He observed that the presence of plastic granules in the soil affected the germination, stem girth and the overall growth of *Zea mays*. He noted that the higher the plastic granules in the soil, the more its negative impact on the growth of *Zea mays*. The resistance and survival of *Telfairia occidentalis* in a polystyrene plastic composted soil could be attributed to the plant's ability to metabolize and use polystyrene plastic granules as a source of carbon.

The morphology of the leaves showed that the plant had a healthy growth (Plate 1) which suggests that it is composed of both vitamins and minerals in their right proportion. According to Idris, who observed the presence of vitamins and mineral composition in *Telfairia occidentalis*.[39] The nutritional composition also reveals that the plant was not affected in any way by the presence of additives in the plastic amended soil. Previous studies on the qualitative analysis of *Telfairia occidentalis* revealed the presence of the following phytochemical components: tannins, saponins, alkaloids, flavonoids, phenol, phylate, and oxalate while glycosides, steroids and terpenoids which are in line with the findings in this research. [40, 41, 42,43] The result from the phytochemical constituent of the plant showed no significant difference from that of the control sample which suggests that the plastic had no significant toxic effect on the plant and on its phytochemical constituent (Table 9). The treatments B, C and D composted with 50, 100 and 200g/4 kg respectively had a higher carbohydrate concentration compared to the control (treatment A) (Table 10). The high carbohydrate concentration was as a result of accumulation of carbon from the plastic composted soil. The presence of higher carbohydrate concentration in the plastic amended treatments further shows that polystyrene plastics support the growth and survival of *Telfairia occidentalis.*

This study also revealed some of the degradation by-products of different additives in the plant which include: Methylene chloride, hexane, chloroform, toluene, tetrachloroethylene, chlorobenzene, dichlorobenzene and benzene (Table 4). Most of these compounds are in the degradation pathway of lots of plastic contaminants like Bisphenol A, Polyvinyl chloride (PVC), Phthalates, Organotins, Alkyltins, PCBs and Alkylphenols. The estimated acceptable daily intake (EADI) was used to determine the human health risk of plastic residues in the consumption of *Telfairia occidentalis*. It was observed that the EADI values for young children were higher than that of the adults (Table 11) which suggests that children could accumulate higher concentrations of these plastic contaminants in their body which is due to their low body weight. This analysis further reveals that young children are more prone to the danger of plastic toxicity.

Conclusion

The findings from this research indicate that there is a constant release of plastic contaminants into the soil which adversely affects soil physicochemical properties and poses serious threat to the survival of soil bacteria and earthworms except for the fluted pumpkin which it served as manure. These findings could open the possibility of turning plastic waste to fertilizer and using it to grow non-edible crops like. The human health risk assessment has also shown that children can accumulate more plastics in their body and are more in danger of plastic toxicity than adults due to their relatively low body weight. Plastic waste should be properly managed to reduce the amount of plastic pollution in the environment. Therefore, further research into the production of biodegradable plastics, recycling, recovery and recollection of plastic waste will help to mitigate the impact of plastic toxicity on the environment.

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Results

Table 1: The physicochemical parameters of the plastic composted soil and the control garden soil sample

Parameters	Plastic composted soil	Control garden soil
CL^-, mg/kg	26.89±0.13	16.08±0.08
K^+, mg/kg	7.97±0.02	3.16±0.04
Ca^{2+}, mg/kg	8.95±0.03	2.97±0.05
SO_4^{2-}, mg/kg	4.50±0.01	2.717±0.02
NO_3^-, mg/kg	7.03±0.02	2.44±0.46
PO_4^{3-}, mg/kg	2.13±0.00	0.86±0.01
Na^+, mg/kg	5.58±0.01	1.45±0.02
Zn^{2+}, mg/kg	9.97±0.04	3.26±0.01
Mn^{2+}, mg/kg	5.22±0.01	0.45±0.02
Mg^{2+}, mg/kg	3.02±0.01	0.81±0.01
Fe^{3+}, mg/kg	27.97±0.05	6.88±0.42
Cd^{2+}, mg/kg	4.1±0.02	0.72±0.06
V^{2+}, mg/kg	3.13±0.23	0.47±0.00
Cu^{2+}, mg/kg	5.56±0.06	1.54±0.06
Ni^{2+}, mg/kg	5.35±0.01	2.72±0.02
Pb^{2+}, mg/kg	8.23±0.02	3.89±0.09
Hg^+, mg/kg	<0.001	<0.001
Cr^{6+}, mg/kg	1.26±0.11	0.73±0.23
As, mg/kg	1.11±0.025	0.31±0.025
pH	5.62±0.05	7.92±0.09
EC, us/cm	61.91±0.41	39.85±0.26
TDS, ppm	31.95±0.73	10.92±0.16
T-Carbon, %	5.80±0.58	0.33±0.38
T-Nitrogen, %	3.06±0.01	0.74±0.56

Table 2: Plastic additives detected in the soil samples and their concentration.

Plasticizers	Plastic composted soil	Garden soil sample
DEHP	14.05	NR
DMP	26.23	NR
DEP	7.07	NR
BPA	40.07	3.02
DBP	<LOD	NR
BBzP	<LOD	NR
Total (ng/g)	87.42	3.02

Key: BBzP—Butylbenzylphthalate, DEP—Diethylphthalate, DMP—Dimethylphthalate,
DEHP-Diethylhexylphthalate, BPA---Bisphenol A, DBP—Di-n-butylphthalate
LOD: Limit of detection, NR: Not recovered

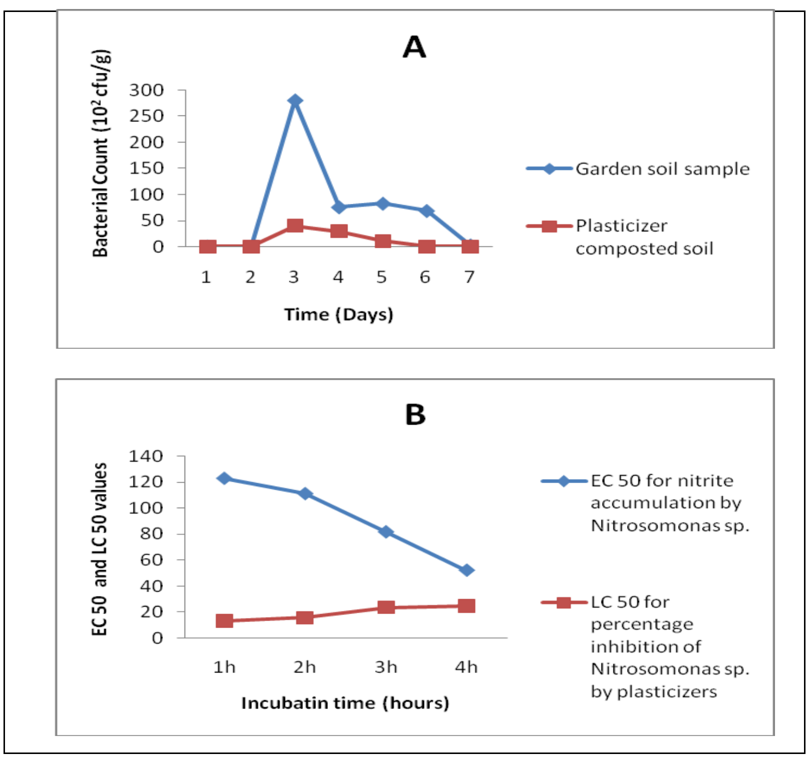

Figure 1: A: Bacterial growth of the control and plastic composted soil samples
B: Bacterial toxicity test showing EC_{50} and LC_{50} values

Table 3: Earthworm growth and survival across various plastic composting soil concentrations

Concentration (g/kg)	Initial weight (g)	Final weight (g)	Weight change	Survival rate (%)
A	0.40±0.05	0.53±0.07	0.13	75.47
B	0.44±0.10	0.12±0.11	-0.32	27.27
C	0.43±0.09	0.09±0.10	-0.34	20.93
D	0.43±0.11	0.00	0.00	0.00
E	0.41±0.06	0.00	0.00	0.00

Key:A= (Control), untreated soilB= (0.83% treatments level)C= (1.67% treatments level) D= (2.50% treatments level) E= (3.33% treatments level)

Plate 1: *Telfairia occidentalis*

Key:

A = Germination of *Telfairia occidentalis*
B = Mid-growth of *Telfairia occidentalis*
C:D = Full growth of *Telfairia occidentalis*

Table 4: Bisphenol A contingents found in the *Telfairia occidentalis* grown both on the plastic composted soil and the garden soil sample

Parameters	Plastic composted soil sample	den soil sample
Methylene	127.05	0.34
Hexane	98.45	0.22
Chloroform	23.56	0.36
Toluene	5.87	0.08
Tetrachloroethylene	1.38	0.02
Chlorobenzene	0.35	0.01
Dichlorobenzene	0.16	0.00
Benzene	0.12	0.00

Table 5: Average stem girth of *Telfairia occidentalis*

Time (weeks)	A	Treatments (cm) B	C	D
3	0.20±0.28	0.30±0.42	0.30±0.42	0.30±0.42
4	0.20±0.28	0.33±0.40	0.33±0.40	0.33±0.40
5	0.21±0.20	0.33±0.48	0.31±0.20	0.33±0.80
6	0.21±0.58	0.34±0.10	0.34±0.00	0.34±0.05
7	0.22±0.38	0.34±0.70	0.34±0.50	0.34±0.58
8	0.22±0.88	0.34±0.90	0.35±0.00	0.35±0.10

Table 6: Height of *Telfairia occidentalis*

	A	Treatments (cm) B	C	D
3	9.33±2.03	12.67±1.86	13.33±0.88	14.33±2.66
4	13.09±0.83	13.5±1.76	14.00±1.83	15.17±1.59
5	13.21±0.20	13.33±0.48	14.31±0.20	15.33±0.80
6	14.51±0.08	14.34±0.10	15.04±0.00	16.34±0.05
7	15.42±0.30	16.40±0.00	16.60±0.00	17.30±0.08
8	16.02±0.81	17.20±0.10	17.48±0.22	19.35±0.10

Table 7: Average number of leaves of *Telfairia occidentalis*

Time (weeks)	A	Treatments (g) B	C	D
3	18.01±2.03	17.67±1.86	20.33±0.88	19.33±2.66
4	43.09±0.83	49.5±1.76	38.00±1.83	45.17±1.59
5	76.21±0.20	70.33±0.48	71.31±0.20	65.33±0.80
6	80.00±0.08	84.34±0.10	86.04±0.00	82.34±0.05
7	112.42±0.30	120.40±0.00	122.60±0.00	123.30±0.08
8	148.02±0.81	147.20±0.10	150.48±0.22	142.35±0.10

Table 8: Phytochemical constituents of *Telfairia occidentalis* across various plastic composting soil concentrations

Phytochemical constituents	A	Treatments B	C	D
Tannins	+	+	+	+
Saponins	+	+	+	+
Flavonoids	+	+	+	+
Steroids	--	--	--	--
Phenol	+	+	+	+
Alkaloids	+	+	+	+
Cynogenic glycosides	--	--	--	--
Terpenoids	--	--	--	--
Oxylate	+	+	+	+
Phylate	+	+	+	+

Table 9: Proximate composition of *Telfairia occidentalis* across various plastic composting soil concentrations

Proximate constituents	A	Treatments (%) B	C	D
Protein	42.60	23.89	21.30	21.80
Ash	9.23	7.66	11.80	7.90
Moisture	8.14	8.69	12.82	8.80
Fiber	25.51	22.29	21.20	26.00
Lipid	11.74	13.60	13.10	13.50
Carbohydrate	2.78	24.34	24.60	22.00

Table 10: Human health risk assessment of plastic residues in *Telfairia occidentalis*

Concentration of plastic residues (g)	EADI Values for *Telfairia occidentalis* (g)	
	Young children	Adults
BPA (0.045)	0.46	0.18
DEHP (0.012)	0.12	0.05
DMP (0.028)	0.28	0.11
DEP (0.005)	0.05	0.02