

PLANTING DENSITY MODULATES GROWTH AND BIOMASS ALLOCATION OF COWPEA (*VIGNA UNGUICULATA* [L.] WALP) IN IRON INDUSTRY EFFLUENT-CONTAMINATED SOIL

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ABSTRACT

Industrialisation and improper effluent discharge practices are major contributors to soil contamination, raising concerns about crop productivity and food safety. This study assessed the influence of iron industry effluent on growth performance and biomass accumulation of *Vigna unguiculata* under varying planting densities. Soil samples from an effluent-contaminated site (ECS) and a non-contaminated control location (CL) were analysed, and cowpea was cultivated in both soils at five densities (1-5 plants/pot). Relevant growth, nodulation, and biomass indices were evaluated. The study revealed significantly higher levels of cadmium, lead, and organic matter in ECS compared to CL. Plants cultivated in CL exhibited higher vegetative and biomass indices and nodulation across all densities. Both effluent contamination and plant density significantly influenced most growth parameters ($p \leq 0.05$), with reduced performance at higher densities, especially in ECS. The study demonstrates that effluent-contaminated soils impair cowpea productivity and that intra-specific competition intensifies stress effects. These findings underscore the need for strict effluent management and density-specific agronomic strategies in polluted agroecosystems.

Keywords: Industrial effluents, heavy metals, pollution, cowpea, yield, nodulation.

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INTRODUCTION

There is tremendous pressure on the soil and environment due to rising industrialisation and effluent discharge. Man's industrial, agricultural, economic, and domestic activities have brought about a shift in the ecological balance (Islam . 2006; Rashid ., 2023). These activities produce wastes which negatively impact the environment, consequently affecting the overall well-being of plants and the soil. Industrialisation, though critical for development, is a major cause of environmental degradation and pollution (Rashid ., 2023). Depending on the source of production, wastewater (treated or untreated) that exits treatment plants, sewers, or industrial outfalls varies in composition. In addition to raising the amount of nutrients in the soil, effluents have the potential to surpass tolerance limits and become hazardous (Mititelu ., 2025).

Soils, as the major sink for industrial effluent heavy metals, may become contaminated, resulting in health hazards through the food chain (McGrath ., 2002; Angon ., 2024). Increased soil pollution due to continuous discharge of heavy metal-contaminated industrial effluents is harmful for crop production globally and poses an environmental threat. These heavy metals, which are not subject to bacterial degradation, accumulate in soils and plants in excess enters the food chain, thereby disrupting animal and plant biological processes and mechanisms (Stolt ., 2006; Jamali ., 2007; Rashid ., 2023). For instance, increased levels of cadmium (Cd) have been reported to alter anatomical and physiological phenomena, causing growth and biomass reduction (Demirevska-Kepova . 2006; Oladele ., 2024; Oladele ., 2025).

Previous studies have also shown that direct or indirect application of industrial effluents could either enhance or reduce the growth and yield of farm crops (Rehman ., 2009; Umbese and Onasanya, 2009; Akinyemi .,

2015). For instance, Akbar . (2007) reported improvement in the growth of *Zea mays* L. when untreated marble industrial wastes were applied at low concentration, whereas at high concentration, the effluents had negative effects. Like all organisms, plants are sensitive both to the deficiency and excessive amounts of some heavy metal ions as essential micronutrients, while the same at higher concentrations are poisonous to their metabolic activities (Akinyemi . 2015).

Cowpea (*V. unguiculata*) is an economically important and nutritionally significant leguminous crop widely cultivated in Nigeria and other parts of sub-Saharan Africa (Enyiukwu ., 2018). Cowpea grain contains about 23-25% protein and serves as an affordable, plant-based alternative to animal protein for low-income Nigerian populations. In rural and suburban settings, cowpea has been shown to contribute 30-36% of daily protein intake in young children and about 26% of adult daily protein requirements, with consumption levels near universal (~98%) in surveyed West African communities (Kuzhkuzha ., 2021; Akissoé ., 2023).

Despite widespread recognition of the detrimental effects of industrial effluents on agricultural systems, few studies have empirically evaluated the direct impact of iron industry effluents on soil physicochemical properties and crop performance in tropical agro-ecosystems. Cowpea, a legume of critical nutritional and economic value in sub-Saharan Africa, is frequently cultivated near industrial zones, yet its physiological and yield responses to heavy metal-laden effluents remain poorly understood. This study integrates soil contamination assessment with density-dependent growth and nodulation dynamics of cowpea, thereby providing novel insights into how effluent-altered soils affect legume productivity and biological nitrogen fixation.

By employing realistic exposure conditions and local cultivars, the findings offer critical evidence to support environmental regulation, effluent pre-treatment and sustainable cropping decisions in peri-industrial landscapes.

Effluent-contaminated soils (ECS) are very common across Nigeria due to the rapid industrial growth, inadequate wastewater treatment facilities, and very poor enforcement of environmental regulations. Empirical evidence shows that in many instances, untreated or partially treated effluents enter the surrounding land and drainage systems, leading to their infiltration into soil used for agriculture by smallholder farmers, particularly located in peri-urban areas (Wuana and Okieimen, 2011; Ihedioha ., 2017). All the industrial activities in the South-west of Nigeria relate to subsistence agriculture; therefore, agricultural products are prone to industrial pollution. Research over the last several years has indicated an increased concentration of heavy metals and a reduction in soil quality on agricultural land found near the iron and steel industries, mechanic villages and industrial zones in this area (Adebayo ., 2022; Adewole ., 2022). Cowpea, a popular food source grown in this area, is typically grown on contaminated soils, threatening both food safety and yields. Although there has been extensive documentation on the existence of ECS in Nigeria, very few studies have focused on how agronomic factors, especially planting density, interact with effluent-induced soil stress to affect crop growth and biomass distribution.

MATERIALS AND METHODS (METHODOLOGY)

Experimental materials

Soil samples were collected randomly from a regrowth forest, specifically between the Department of Botany and Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria, while effluent-contaminated soil samples were collected from the premises of Ife Iron and Steel Nigeria Limited, Fashina, along Ibadan expressway, Ile-Ife, Nigeria (70 29' 42.14" N, 40 28' 38.18" E). Viable seeds of *V. unguiculata* (Ife Brown) were collected from the Osun State Agricultural Development Project office, Osogbo, Osun State, Nigeria. Ife brown was selected because of its broad adaptability, stable yield performance, and frequent use in agronomic and stress-response studies, making it suitable for controlled pot experiments.

Experimental site

Pot experiments were carried out within the screenhouse of Obafemi Awolowo University's Department of Botany in Ile-Ife, Nigeria.

Experimental setup

Each soil sample was thoroughly mixed, air-dried, loosened and placed in 21-cm diameter and 24-cm tall plastic containers that were perforated at the base for effective drainage.

Physicochemical Analysis of Soil

The soil samples were analysed for pH using a pH-meter, particle size distribution using a Bouyoucos hydrometer, and organic matter content, to establish the baseline soil health. The organic matter (OM) was determined by wet combustion with Potassium dichromate ($K_2Cr_2O_7$) (Nelson and Sommers, 1982).

Total nitrogen (N) was determined by the micro-Kjeldahl method (Jackson, 1973; Adejumo, 2025) while potassium (K) was extracted through 1M CH_3COONH_4 and determined by flame photometry (Black, 1965). Available phosphorus (P) content was determined by sodium bicarbonate extraction (Olsen, 1954) while the concentrations of cadmium (Cd), copper (Cu), and lead (Pb) were determined by ICPOES (Inductively Coupled Plasma Optical Emission Spectrometer).

Raising of Seedlings

Seedlings were raised by broadcasting the *V. unguiculata* seeds for 2 weeks in a rectangular container filled with soil for the establishment of a nursery bed. The seedlings were first raised in the nursery to ensure uniform establishment. The emerged seedlings were transferred into perforated pots filled with the soil samples at 1, 2, 3, 4, and 5 plants per pot. Each density had 5 replicates for each soil sample.

Evaluation of Growth and Biomass Parameters

During the growing period, parameters like the number of leaves, leaf length, leaf breadth (leaf area) and shoot length were measured weekly, according to standard procedures (Hunt, 2012; Rueegg ., 2012). At the mid-flowering stage, plant samples were harvested for nodule counting. Pots were gently soaked and soil carefully loosened before uprooting plants to avoid dislodging the nodules. Roots were washed under slow-flowing tap water

to preserve nodules. The number of nodules counted and dry matter yield (leaves, stems, pods, nodules and roots) were determined after oven drying plant samples in a Gallenkamp oven at 80 °C for 46 hours.

The samples were further dried for two more hours and reweighed repeatedly until a constant weight was achieved, establishing complete moisture removal.

Statistical Data Analysis

All results were subjected to analysis of variance (ANOVA) and are presented in graphs and tables. Significant means were separated by Duncan's multiple range test at the 5 % probability level with the SPSS 16.0 statistical software (SPSS Inc., USA) due to the factorial nature of the experiment involving multiple treatment combinations.

RESULTS AND DISCUSSION

Soil Properties

The physicochemical properties of the soil contaminated with the effluent and uncontaminated soil samples (CL) showed that the effluent-contaminated soil (ECS) had higher organic carbon contents than the CL. Clay and silt contents were higher in ECS than the control, while the control soil had a greater percentage of sand particles. The amounts of Nitrogen and available Phosphorus were higher in ECS than in the control soil. The results also show that the concentrations of copper, cadmium and lead were higher in the effluent-contaminated soil. Aside pH, silt and Cu content, the differences between the other physicochemical parameters of the two soils were significant (Table 1).

Table 1: Physicochemical Properties of the Soil

Soil type	pH	Sand	Silt	Clay	OM (mg/kg)	N %	K (mg/kg)	P (mg/kg)	Cu (mg/kg)	Pb (mg/kg)	Cd (mg/kg)
CL	7.60a	81.00a	12.00a	7.00b	0.80a	0.67b	7.12b	6.11b	10.20a	21.91b	2.45b
ECS	7.50a	75.00b	13.50a	11.50a	2.90b	0.92a	17.73a	16.22a	10.30a	22.82a	2.67a

***Means within the same column and with different letters are significantly different ($p \leq 0.05$).**

ECS = Effluent Contaminated soil, CL = Control soil

Vegetative Growth Parameters

Cowpea plants grown in control soil (CL) exhibited better vegetative performance than those in effluent-contaminated soil (ECS) (Figures 1a-3b). Throughout the growth period, mean leaf number, leaf area, and shoot height were consistently greater in CL. By week 9, the pots holding only one plant in CL produced a peak average of 37 leaves, which declined under densities ≥ 4 , indicating intra-specific competition. In contrast, plants grown in ECS produced an average of 21 leaves at a similar density. The result revealed significant main effects of soil type and plant density on leaf number, leaf area, and stem/leaf dry weights ($p \leq 0.05$), while shoot height showed no statistically significant response to soil type ($p \leq 0.05$).

Biomass and Yield Indices

At harvest, plants in CL soil accumulated greater dry biomass: leaf dry weight (4.11g), stem dry weight (3.57g), and root dry weight (1.29g), significantly greater than in ECS (3.48g, 2.85g, and 1.03g, respectively; $p \leq 0.05$). Root nodule numbers were slightly larger in CL (34.20) than in ECS (32.60), though this

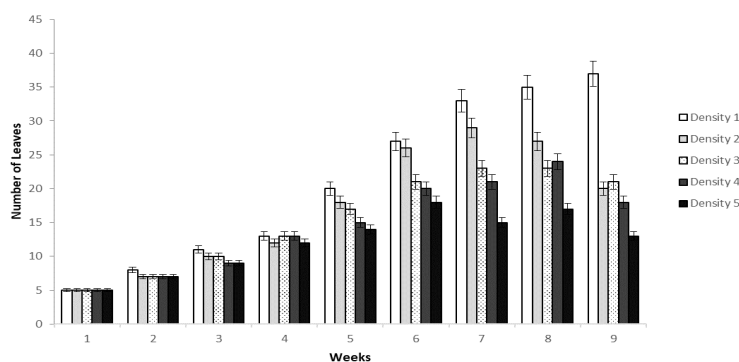
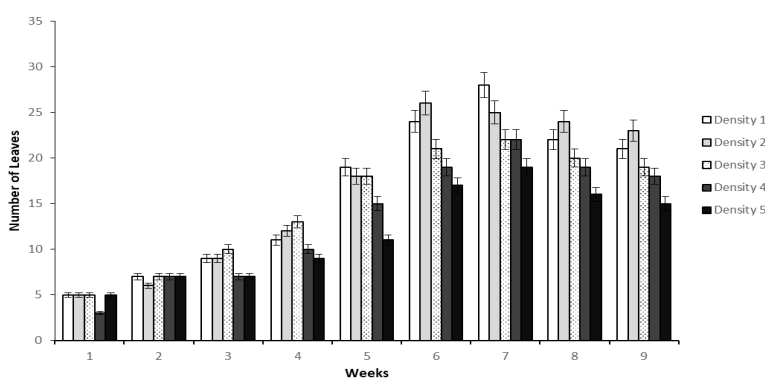
was not statistically significant ($p \leq 0.05$). The average number of pods per plant remained constant (2.00) across soil treatments (Table 2). The single-plant density (1 plant/pot) majorly outperformed denser arrangements in nodulation and dry matter yield significantly under the two soil conditions ($p \leq 0.05$) (Figures 4 and 5).

The result of ANOVA indicates that planting density had a significant influence on the biomass allocation and vegetative traits of cowpea. There is progressive increase in stem, root, and dry weights as planting density increases. On the contrary, leaf and pod numbers showed significant decrease in with increasing planting densities while number of pods and nodulation showed no significant difference across varying planting densities. The result of ANOVA indicates that planting density had a significant influence on the biomass allocation and vegetative traits of cowpea. There is progressive increase in stem and root dry weights as planting density increases. On the contrary, leaf and pod numbers showed significant decrease in with increasing planting densities while number of pods and nodulation showed no significant difference across varying planting densities.

Table 2: Some Growth Characteristics and Yield of *V. unguiculata* at Harvest Under Different Soil Conditions.

Soil type	Stem dry wt. (g)	Root dry wt. (g)	Leaf dry wt. (g)	Leaf number	No. of pods	Root nodules
CL	3.57a	1.29ab	4.11a	88.80a	2.00a	34.20a
ECS	2.85b	1.03a	3.48b	65.42b	2.00a	32.60a

*Means within the same column and with different letters are significantly different at $p \leq 0.05$. ECS = Effluent discharge soil, CL = Control soil

**Fig. 1a:** Weekly leaf number of *V. unguiculata* (CL)**Fig.1b:** Weekly leaf number of *V. unguiculata* (ECS)

Planting Density and Cowpea Growth under Effluent Contamination

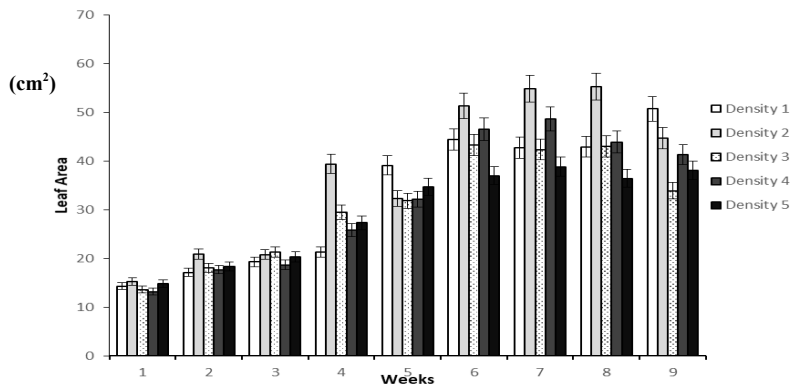


Fig. 2a: Weekly leaf area of *V. unguiculata* (CL).

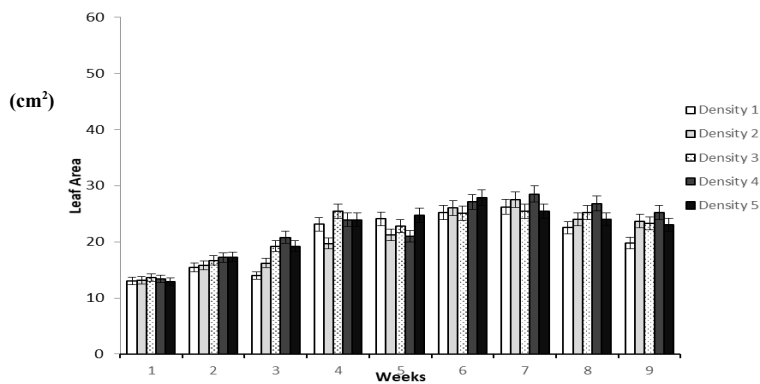


Fig.2b: Weekly leaf area of *V. unguiculata* (ECS).

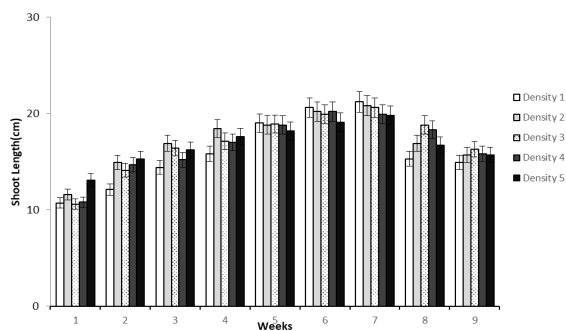


Fig. 3a: Weekly shoot height of *V. unguiculata* (Control)

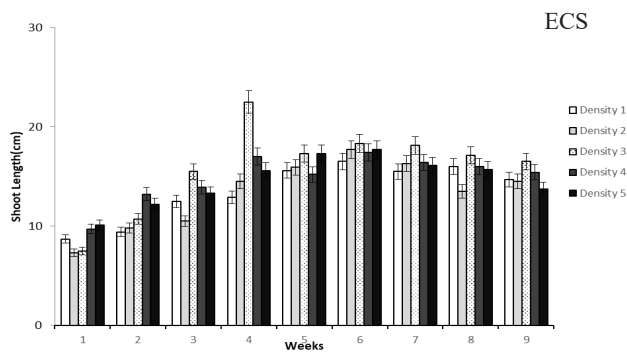


Fig. 3b: Weekly shoot height of *V. unguiculata* (ECS)

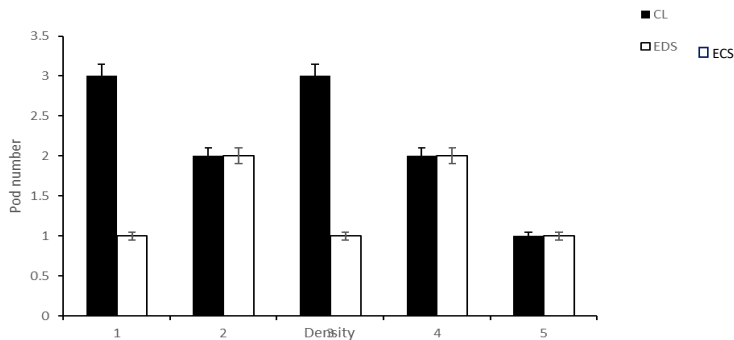


Fig. 4: Number of pods per plant on cowpeas grown on the contaminated and uncontaminated soils
CL = control; ECS= Effluent Contaminated Soil

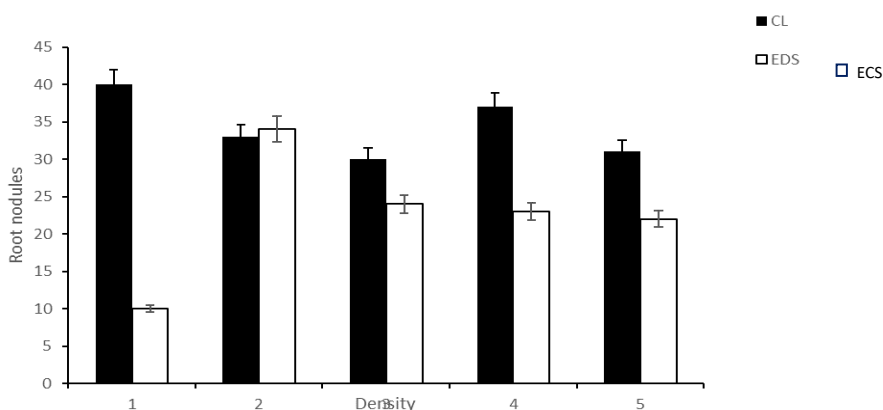


Fig. 5: Number of root nodules on cowpeas grown on the contaminated and uncontaminated soils
CL = control; ECS = Effluent Contaminated Soil

Table 3: Summary of ANOVA Table Showing the Effect of Planting Density on Growth, Biomass, and Yield Parameters of Cowpeas

Planting Density (plants/pot)	Stem dry weight (g)	Root dry weight (g)	Pod dry weight (g)	Leaf dry weight (g)	Pod number	Nodules number	Leaf number
1	2.085 ^B	0.600 ^B	3.785 ^A	2.420 ^B	2.500 ^A	25.00 ^A	39.500 ^A
2	2.915 ^{AB}	1.265 ^{AB}	4.300 ^A	2.865 ^B	2.500 ^A	33.50 ^A	29.500 ^B
3	3.420 ^{AB}	1.120 ^{AB}	4.250 ^A	3.735 ^{AB}	1.000 ^B	27.00 ^A	26.000 ^{BC}
4	3.585 ^{AB}	1.215 ^{AB}	4.945 ^A	4.765 ^A	2.000 ^{AB}	30.00 ^A	22.500 ^{BC}
5	4.065 ^A	1.605 ^A	5.970 ^A	5.195 ^A	1.000 ^B	26.50 ^A	19.500 ^C

*Means within the same column and with different letters are significantly different at $p \leq 0.05$.

Discussion

Chemical analysis indicated that the concentrations of lead, cadmium and copper were higher in effluent-contaminated soil than in the control soil. Reports have shown that soils contaminated with industrial wastes have high metal contents. Dastorania (2008) reported an increase in Lead (Pb) content of soils treated with industrial wastewater.

The study evaluated the response of the growth characteristics of cowpea (*V.*

unguiculata) seedlings to different soil conditions and densities.

The observed reduced shoot height of *V. unguiculata* when grown in effluent-contaminated soil is in line with the findings of Bafna and Rath (2012), who observed a significant decrease in shoot height of *Vigna radiata* in pharmaceutical effluent-contaminated discharged soil.

The reduction in shoot height might be due to the heavy metals present in the effluent discharged into the soil. The result is also supported by Verma and Sharma (2012), who observed a decrease in shoot height and root length of wheat as the flow of dairy and textile wastewater increased.

The effect of seedling density on shoot height showed that the pots with five seedlings had taller shoots than the other seedling densities in both soil types. Results similar to these were obtained by El Naim and Jabereldar (2010), who found that cowpeas produced at higher densities were taller and more sparsely branched. Increasing shoot density tended to increase plant shoot height, which may be due to competition for light, which made the seedlings direct more biomass into stem elongation and branching.

The number of root nodules and pods in the control was greater than in the effluent-contaminated soil. This is in agreement with the work of Aery and Sarkar (1991), who observed a decrease in soybean nodule numbers in soils with high levels of zinc and cadmium. Nodules are active sites of biological nitrogen fixation that supplement the needs of the crop; the reduction of nodule number may be due to the high lead content in the effluent discharged into the soil. Concerning the effect of seedling density on root nodule dry weight, the pots containing only one plant had the greatest root nodule dry weight compared to other seedling densities. Hamad (2004) also reported that plants grown at very high densities per stand produced fewer pods than those at low densities. This observation can be explained by the direct relationship between the individuals engaged in the competition for the limited resources.

The total biomass was smaller in the effluent-contaminated soil than in the agricultural soil, which was in line with the work of Verma and Sharma (2012), who reported that wheat weights decreased as the concentration of

dairy and textile effluent increased. The result also agrees with the work of Sundaramoorthy (2001), who reported a significant reduction in the biomass of groundnut varieties grown on soil exposed to fertiliser factory effluent. Considering the effects of seedling number per pot on biomass production, pots with one seedling had greater biomass than those with higher seedling density, which might be attributed to minimal intra-specific competition due to low planting density.

CONCLUSION

This study showed that the growth and crop yield of *V. unguiculata* grown on soils contaminated with industrial effluents were adversely affected by the elevated concentrations of heavy metals such as cadmium, lead, and copper. The effluents also put the soil at risk of being unable to support healthy crop growth. The modulation effects of planting density underscore the importance of optimising plant population when cowpea is cultivated on effluent-contaminated environments, where resource limitation and stress interactions may amplify competition effects. These findings also emphasise the urgent need for regulatory enforcement of industrial effluent pre-treatment and safety analysis before discharge into agricultural landscapes.

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CONFLICT OF INTEREST

The authors declare none.

AUTHORS' CONTRIBUTIONS

Akinyemi: designed the study, carried out the investigation, data collection, and manuscript writing (original draft).

Adejumo: collected data, analysed and validated data, reviewed and edited the manuscript.

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