



COMPARATIVE ASSESSMENT OF ETHANOL LEAF EXTRACTS OF *CHROMOLAENA ODORATA*, *AZADIRACHTA INDICA* AND *PERSEA AMERICANA* FOR HEAVY METAL REMEDIATION IN LANDFILL LEACHATE

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ABSTRACT

Landfill leachate constitutes a major environmental challenge because it contains elevated concentrations of heavy metals capable of contaminating surface water, groundwater, and surrounding ecosystems. The present study evaluated the effectiveness of ethanol leaf extracts of *Persea americana*, *Chromolaena odorata*, and *Azadirachta indica*, applied individually and in different combinations, for the removal of heavy metals from landfill leachate. Fresh leaves of the three plant species were collected, air-dried, pulverized, and extracted with ethanol using standard extraction procedures. The extracts were applied to landfill leachate at concentrations of 1, 2, and 3 mg/mL as single, binary, and ternary treatments. Heavy metal concentrations before and after treatment were determined using Atomic Absorption Spectrophotometry (AAS), and the data were analyzed using analysis of variance (ANOVA) at a 95% confidence level. The results shows significant ($p < 0.05$) reductions in the concentrations of iron (Fe), manganese (Mn), copper (Cu), cobalt (Co), zinc (Zn), lead (Pb), silver (Ag), and nickel (Ni) following treatment with the plant extracts. The heavy metal removal efficiency generally increased with extract concentration and was consistently higher in binary and ternary extract combinations than in single-plant treatments. Among all treatments, the combined extract of *P. americana* + *C. odorata* + *A. indica* at 3 mg/L exhibited the highest remediation efficiency, producing the lowest residual concentrations for most of the metals investigated. Copper and zinc concentrations were reduced to levels below the World Health Organization (WHO) drinking water guidelines, while manganese and lead approached acceptable limits in several combined treatments. Iron concentrations were substantially reduced to values approaching aesthetic guideline limits. However, nickel concentrations remained above the WHO permissible limit in all treatments despite marked reductions, indicating that additional polishing treatment would be required before potable use. This study demonstrates that ethanol leaf extracts of *Persea americana*, *Chromolaena odorata*, and *Azadirachta indica*, particularly when applied in combination, possess environmentally friendly, and sustainable properties for heavy metal remediation of landfill leachate.

KEYWORDS: Landfill leachate; Heavy metal remediation; Plant-based treatment; Wastewater treatment; *Chromolaena odorata*.

INTRODUCTION

Landfill leachate is one of the most environmentally hazardous liquid wastes generated from municipal solid waste disposal sites worldwide.

It is produced when rainwater percolates through solid waste deposits, dissolving organic and inorganic constituents generated during waste decomposition and transporting them into the surrounding environment.

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The physicochemical composition of leachate varies considerably depending on landfill age, waste composition, climatic conditions, moisture content, landfill design, and waste stabilization processes. Mature landfill leachates are generally characterized by elevated concentrations of dissolved organic carbon, ammonia, chlorides, sulfates, xenobiotic organic compounds, pathogenic microorganisms, and toxic heavy metals, making them a major source of environmental pollution if discharged without adequate treatment (Kjeldsen et al., 2002; Renou et al., 2008; Christensen et al., 2001; Mor et al., 2006; Aziz et al., 2010).

Heavy metals represent one of the most hazardous constituents of landfill leachate because they are non-biodegradable, persistent, and capable of accumulating within environmental compartments through bioaccumulation and biomagnification. Unlike many organic pollutants that can be degraded by microbial activity, heavy metals remain chemically stable for extended periods, thereby posing long-term ecological and public health risks. Common heavy metals detected in landfill leachates include lead (Pb), cadmium (Cd), chromium (Cr), copper (Cu), zinc (Zn), nickel (Ni), iron (Fe), manganese (Mn), arsenic (As), and mercury (Hg), many of which frequently exceed the permissible discharge limits established by the World Health Organization (WHO), the United States Environmental Protection Agency (USEPA), and other environmental regulatory agencies (Kjeldsen et al., 2002; Fu and Wang, 2011; Tchounwou et al., 2012; Jaishankar et al., 2014; Kumar et al., 2019). The environmental implications of heavy metal contamination are profound because these contaminants readily migrate from landfill sites into groundwater, rivers, agricultural soils, and food chains. Long-term exposure to contaminated water and food has been associated with numerous health disorders including nephrotoxicity, hepatotoxicity, neurotoxicity, immunotoxicity, endocrine disruption, developmental abnormalities, reproductive dysfunction, mutagenesis, and carcinogenesis (Alloway, 2013; Jaishankar et al., 2014; Briffa et al., 2020). Lead exposure has been linked with neurological impairment, reduced cognitive development in children, hypertension, and kidney disease, whereas cadmium accumulates primarily in the kidneys and bones, causing renal dysfunction and skeletal disorders. Hexavalent chromium (Cr^{6+}) is a recognized human carcinogen capable of inducing oxidative stress and DNA damage, while mercury affects the central nervous system and impairs neurological development. Likewise, excessive arsenic exposure has been associated with skin lesions, cardiovascular diseases, diabetes mellitus, and various forms of cancer (Tchounwou et al., 2012; Jaishankar et al., 2014; WHO, 2017). The increasing occurrence of heavy metals in aquatic ecosystems

has become a major environmental concern due to rapid industrialization, urbanization, mining activities, agricultural intensification, indiscriminate waste disposal, and inadequate landfill management. In many developing countries, uncontrolled municipal solid waste disposal has significantly increased the risk of groundwater contamination by landfill leachates, threatening potable water resources and ecosystem sustainability (Mor et al., 2006; Naveen et al., 2017). Consequently, there is a growing demand for sustainable technologies capable of effectively removing heavy metals from contaminated wastewater before environmental discharge. Several conventional technologies have been developed for heavy metal removal from industrial wastewater and landfill leachate. These include chemical precipitation, coagulation-flocculation, membrane filtration, reverse osmosis, ion exchange, electrochemical treatment, solvent extraction, electrodialysis, and adsorption using activated carbon. Although these techniques often achieve high removal efficiencies, their practical application is frequently limited by high capital and operational costs, excessive energy consumption, generation of toxic sludge, membrane fouling, complex maintenance requirements, and poor performance at low metal concentrations (Fu and Wang, 2011; Barakat, 2011; Crini and Lichtfouse, 2019). These limitations have stimulated considerable research interest in environmentally friendly and economically viable alternatives for wastewater remediation. Among the emerging technologies, biosorption has become one of the most promising approaches for heavy metal removal because it employs naturally occurring biological materials capable of binding metallic ions through passive physicochemical interactions. Biosorption involves adsorption, ion exchange, chelation, complexation, precipitation, and electrostatic attraction between dissolved metal ions and functional groups present on biological materials (Vijayaraghavan and Yun, 2008; Ali et al., 2021). Compared with conventional treatment methods, biosorption offers numerous advantages, including simplicity of operation, low cost, high efficiency, minimal sludge production, regeneration potential, and environmental sustainability (Crini and Lichtfouse, 2019; Ghosh et al., 2022). Plant-derived biosorbents have attracted increasing scientific interest because of their abundance, renewability, biodegradability, and high affinity for heavy metals. Plant tissues contain cellulose, lignin, hemicellulose, pectin, proteins, and various secondary metabolites possessing hydroxyl, carboxyl, amino, sulfhydryl, phosphate, methoxy, and phenolic functional groups capable of binding metallic ions. These natural materials have demonstrated remarkable adsorption capacities for lead, cadmium, chromium, copper, zinc, nickel, and other toxic metals from aqueous solutions (Demirbas, 2008; Ali et al., 2021; Ghosh et al., 2022).

The efficiency of plant extracts in heavy metal remediation depends largely on the quantity and accessibility of their active functional groups. Hydroxyl, amino, carboxyl, phosphate, carbonyl, sulfhydryl, and phenolic groups readily interact with dissolved metal ions through electrostatic attraction, ion exchange, chelation, coordination, and surface complexation, thereby facilitating efficient removal from contaminated aqueous media (Demirbas, 2008; Vijayaraghavan and Yun, 2008; Ali et al., 2021). However, plant-derived biosorbents are biodegradable, renewable, locally available, environmentally compatible, and economically sustainable, making them attractive alternatives to synthetic adsorbents. Nigeria continues to experience increasing environmental challenges associated with rapid urbanization, population growth, industrial development, and inadequate municipal waste management systems. These challenges have contributed to increased production of landfill leachates capable of contaminating surrounding water bodies with toxic heavy metals. The exploration of locally available plant materials as natural biosorbents therefore represents a practical strategy for developing low-cost and sustainable wastewater treatment technologies suitable for resource-limited environments (Nwosu et al., 2017; Naveen et al., 2017). Although several studies have investigated heavy metal adsorption using agricultural residues and plant biomasses, relatively few have comparatively evaluated the effectiveness of ethanol leaf extracts of *Chromolaena odorata*, *Azadirachta indica*, and *Persea americana* in the remediation of landfill leachate. Comparative assessment of these indigenous plant species is expected to identify the most efficient biosorbent while expanding current knowledge on plant-based remediation technologies. Therefore, this study was undertaken to evaluate the efficiency of ethanol leaf extracts of *Chromolaena odorata*, *Azadirachta indica*, and *Persea americana* in the removal of heavy metals from landfill leachate. The findings are expected to contribute to the development of environmentally friendly, cost-effective, and sustainable technologies for the remediation of heavy metal-contaminated leachates while promoting the utilization of locally available plant resources in environmental pollution control.

MATERIALS AND METHODS

This study was conducted using leachate samples collected from the Lemna dumpsite located within Calabar Municipality, Cross River State, Nigeria. Calabar is a rapidly growing Urban area characterized by increased waste generation and inadequate waste management practices. The Lemna dumpsite serves as a major disposal site for municipal solid waste and poses environmental risks due to uncontrolled leachate generation. Laboratory analyses were

carried out at the Ministry of Science and Technology Laboratory, Uyo, Akwa Ibom State, for heavy metal analysis.

Leachate collection

Leachate samples were collected from active seepage points at the Lemna dumpsite using clean, sterile plastic containers. The geographical coordinates of the dumpsite are approximately latitude 04°56.922' N and longitude 008°20.974' E. The pH of the leachate at the point of collection was 5.2. The samples were tightly sealed, labeled, and transported in an ice-packed container to the laboratory for analysis to minimize physicochemical and microbial alterations prior to treatment.

Collection and processing of plant materials

Fresh leaves of *Persea americana*, *Chromolaena odorata*, and *Azadirachta indica* were collected from the Botanical garden of the University of Calabar. Fresh leaves were collected and washed thoroughly with distilled water to remove dirt and contaminants. The leaves were air-dried at room temperature (25–30°C) for 10 days until constant weight was obtained. The dried leaves were ground into fine powder using a mechanical grinder. The powdered sample was stored in airtight containers until extraction.

Soxhlet extraction procedure for the collected leaves

The extraction was done by using 100 g of powdered leaf sample that was accurately weighed using an analytical balance. The sample was placed into a Whatman filter paper thimble. The thimble was inserted into the Soxhlet extraction chamber. About 200 mL of ethanol was poured into a 250 mL round-bottom flask. The Soxhlet apparatus was assembled consisting of: round-bottom flask, Soxhlet extractor and condenser. The setup was placed on a heating mantle, and the solvent was heated to reflux. The solvent evaporated, condensed in the condenser, and filled the extraction chamber containing the plant material. Once the chamber reached a certain level, the solvent automatically siphoned back into the flask, carrying extracted compounds. The extraction was allowed to proceed for 6–8 hours (approximately 10–15 cycles) until the solvent in the siphon tube became colorless. After extraction, the apparatus was allowed to cool before dismantling. The solvent containing the extract was concentrated by evaporating on a water bath at 40–50°C. The concentrated crude extract was transferred to a clean, pre-weighed container. The extract was dried in a desiccator to remove residual solvent.

Experimental design

The experimental design adopted for this study was a 4 × 7 factorial arrangement in a Completely Randomized Design (CRD). Factor A consisted of the plant extracts (in single and combined forms), while Factor B represented extract concentrations (0 mg/mL, 1 mg/mL, 2mg/mL, and 3mg/mL).

Treatment of leachate samples

A total of 84 sterile sample bottles were used for the treatment experiment. One hundred milligram (100mg) of leachate was dispensed into each bottle. The bottles were grouped according to extract type and concentration. The leachate samples were treated with varying concentrations of the ethanol leaf extracts (0mg/mL, 1mg/mL, 2mg/mL, and 3mg/mL). Untreated leachate served as the control. The treated samples were gently shaken and allowed to stand at room temperature for 42 days to facilitate coagulation and interaction between the leachate contaminants and plant extracts. The Jar Test procedure was performed according to George et al. (2018).

Heavy metal analysis

Heavy metal analysis was carried out to determine the concentrations of iron (Fe), copper (Cu), zinc (Zn), manganese (Mn), lead (Pb), nickel (Ni), and cobalt (Co) in the leachate samples before and after treatment. The analyses were conducted using Atomic Absorption Spectrophotometry (AAS) at the Ministry of Science and Technology Laboratory, Uyo, Akwa Ibom State, following standard analytical procedures (Agbor et al., 2026).

Sample digestion

Prior to metal analysis, leachate samples were digested to ensure complete dissolution of metals and removal of organic interferences. A 50 mL aliquot of each leachate sample was measured into a digestion flask, and 5 mL of concentrated nitric acid (HNO_3) was added. The mixture was heated gently on a hot plate until the volume was reduced to approximately 10 mL and a clear solution was obtained. The digested sample was allowed to cool, filtered if necessary, and diluted to 50 mL with distilled water.

Determination of iron (Fe)

Iron concentration was determined using the 1,10-phenanthroline colorimetric method (Agbor et al., 2026). One milliliter of the digested sample was pipetted into a 25 mL volumetric flask. One milliliter of 10% hydroxylamine hydrochloride was added to reduce Fe^{3+} to Fe^{2+} , followed by 1 mL of 0.1% 1, 10-phenanthroline and 1 mL of ammonium acetate buffer. The solution was diluted to volume with distilled water and allowed to stand for one hour. Absorbance was measured at 510 nm using a spectrophotometer.

Determination of copper (Cu)

Copper was determined using the bis-cyclohexanone oxalyldihydrazone method (Agbor et al., 2026). Five milliliters of the digested sample was pipetted into a 100 mL volumetric flask. Ammonium citrate solution and neutral red indicator were added, and the solution was neutralized with sodium hydroxide. Borate buffer and bis-cyclohexanone oxalyldihydrazone reagent were added, and the mixture was diluted to volume. Absorbance was measured at 595 nm.

Determination of manganese (Mn)

Manganese was analyzed using the potassium periodate oxidation method. Five milliliters of the digested sample was transferred into a 25 mL volumetric flask, followed by the addition of phosphoric acid and potassium periodate. The mixture was heated in an oven at 100 °C for two hours to develop a purple coloration. After cooling, the solution was diluted to volume, and absorbance was measured at 545 nm.

Determination of zinc (Zn)

Zinc concentration was determined by EDTA titrimetric method (Agbor et al., 2026). Five milliliters of the digested sample was transferred into a conical flask, followed by the addition of ammonium buffer (pH 10) and Eriochrome Black T indicator. The solution was titrated against standardized EDTA solution until the color changed from wine red to blue. Zinc concentration was calculated based on the volume of EDTA used.

Determination of lead (Pb)

Lead was determined using the sodium sulfide colorimetric method (Agbor et al., 2026). Five milliliters of the digested sample was pipetted into a 25 mL volumetric flask. Potassium cyanide solution, ammonia, and sodium sulfide were added sequentially. The solution was diluted to volume, and absorbance was measured at 430 nm.

Determination of nickel (Ni)

Nickel concentration was determined using the dimethylglyoxime method (Agbor et al., 2026). Five milliliters of the digested sample was transferred into a 25 mL volumetric flask. Ammonium citrate, iodine solution, and dimethylglyoxime reagent were added. The mixture was allowed to stand for 20 minutes for color development, and absorbance was measured at 530 nm. All measurements were carried out in triplicate, and mean values were recorded to ensure analytical accuracy and reliability.

Statistical analysis

Data generated from the study were subjected to two-way analysis of variance (ANOVA) appropriate for a factorial Completely Randomised Design to evaluate the effects of extract type, extract concentration, and their interaction on the physicochemical characteristics of the treated landfill leachate. Where significant differences were observed ($p < 0.05$), treatment means were separated using the Least Significant Difference (LSD) test at the 5% probability level. All statistical analyses were performed using appropriate statistical software, and results were presented as mean $\pm$ standard deviation.

RESULTS AND DISCUSSION**Heavy metal concentrations in treated leachate**

The effects of plant extracts on the heavy metal composition of landfill leachate are presented in Table 1. Treatment of the leachate samples with *Persea americana* (PA), *Chromolaena odorata* (CO),

Azadirachta indica (Al), and their combinations resulted in significant reductions ($p < 0.05$) in the concentrations of all analyzed heavy metals when compared with the untreated control. The comparative efficacy of individual and combined plant extracts *Persea americana*, *Chromolaena odorata*, and *Azadirachta indica* on the concentrations of selected heavy metals (Fe, Mn, Cu, Co, Zn, Pb, Ag, and Ni) in landfill leachate samples equally established significant differences among the treatments. The percentage reduction of the heavy metals reduction is as presented in Fig. 1-7.

Iron (Fe)

Table 1 showed that the untreated leachate recorded the highest iron concentration (7.86 mg/L), which was significantly higher than all treated samples. Application of plant extracts at 1–3mg/mL led to remarkable reductions in Fe levels, with values ranging from 0.96 to 0.30 mg/L. The lowest Fe concentration was observed in the PA+CO+Al treatment at 3mg/mL, indicating enhanced removal efficiency through extract combination. Differences exceeding the LSD value (0.12 mg/L) confirm the statistical significance of these reductions. Table 2 revealed that Iron concentrations varied significantly across treatments, ranging from 2.32 ± 0.14 mg/L to 2.60 ± 0.21 mg/L. Among single-plant treatments, *C. odorata* recorded the highest Fe concentration (2.60 ± 0.21 mg/L), whereas *A. indica* showed a comparatively lower value (2.50 ± 0.09 mg/L). Notably, combined treatments resulted in a progressive reduction in Fe concentration. The ternary combination (*P. americana* + *C. odorata* + *A. indica*) exhibited the lowest Fe level (2.32 ± 0.14 mg/L), which was significantly different from all single-plant treatments, indicating a synergistic effect in iron removal efficiency.

Manganese (Mn)

Manganese concentration significantly decreased from 1.34 mg/L in the control to between 0.72 and 0.115 mg/L in treated samples. Although numerical reductions increased with extract concentration, many treatments shared similar superscript letters, suggesting no significant difference among several high-dose treatments. Nonetheless, the lowest Mn value was recorded in the PA+CO+Al (3mg/mL) treatment (Table 1). Manganese concentrations ranged from 0.46 ± 0.09 mg/L to 0.812 ± 0.08 mg/L. The highest Mn concentration was observed in leachate treated with *P. americana* alone. In contrast, the ternary plant extract combination recorded the lowest Mn concentration, significantly lower than both individual and binary treatments. This suggests enhanced Mn sequestration when multiple phytochemicals act concurrently (Table 2). Most treatments at higher doses (especially combinations) fall within and below the WHO guidelines, but some lower-dose treatments may exceed the limit.

Higher extract concentrations improve compliance for Mn; lower doses may remain above safe levels.

Copper (Cu)

The result as presented in Table 1 shows that copper levels were significantly reduced in all treated samples relative to the control (0.745 mg/L). Treated samples ranged from 0.64 to 0.19 mg/L, with combination treatments consistently recording lower Cu concentrations than single-plant extracts. The lowest Cu concentration was observed in PA+Al and PA+CO+Al treatments at higher extract concentrations. In Table 2 copper levels showed significant variation among treatments. *C. odorata* and *P. americana* exhibited the highest Cu concentrations (0.543 ± 0.06 mg/L and 0.504 ± 0.06 mg/L, respectively), whereas *A. indica* and the ternary combination reduced Cu levels to 0.341 ± 0.08 mg/L and 0.38 ± 0.05 mg/L, respectively. Binary combinations showed intermediate effects, indicating partial additive interactions. Cu concentrations after treatment (0.19–0.64 mg/L) are below the WHO guideline across all treatments.

Cobalt (Co)

Cobalt concentration in the untreated leachate (0.864 mg/L) was significantly higher than all treated samples. Most 0.2mg/L and 0.3mg/L treatments recorded extremely low Co concentrations (≤ 0.03 mg/L), sharing similar statistical groupings. This indicates high sensitivity of cobalt to plant-based treatment, even at moderate extract volumes (Table 1). In Table 2 the Cobalt concentrations ranged narrowly between 0.25 ± 0.04 mg/L and 0.326 ± 0.09 mg/L. Single-plant extracts generally recorded higher Co concentrations, while combined treatments, particularly those involving *A. indica*, significantly reduced Co levels. The lowest Co concentrations were observed in binary and ternary treatments (0.25 mg/L), suggesting that combined extracts are more effective in cobalt immobilization.

Zinc (Zn)

The result as presented in Table 1 shows that the zinc concentration declined significantly from 1.84 mg/L in the control to values ranging between 0.80 and 0.26 mg/L in treated samples. Combination treatments, particularly PA+CO+Al, produced significantly lower Zn concentrations compared to single-extract treatments, especially at 3 mg/mL dosage. Zinc concentrations were highest in leachate treated with *P. americana* alone (0.935 ± 0.06 mg/L) and lowest in the ternary extract treatment (0.69 ± 0.08 mg/L). All combined treatments showed significantly lower Zn concentrations compared to individual plant extracts, reinforcing the advantage of multi-plant extract systems for zinc removal (Table 2). The Zn values (as low as 0.26–0.80 mg/L) are well below the WHO guideline.

Lead (Pb)

Lead concentration showed a pronounced and significant decrease from 1.23 mg/mL in the control to ≤ 0.02 mg/L in most 2mg/mL and 3mg/mL treatments. Differences among higher-dose treatments were not statistically significant, indicating that effective Pb removal was achieved across all plant extract combinations (Table 1). Lead concentrations demonstrated clear treatment-dependent reductions. *C. odorata* alone recorded the highest Pb concentration (0.419 ± 0.08 mg/mL), while all binary and ternary combinations significantly reduced Pb levels to approximately 0.33–0.35 mg/L. The absence of significant differences among combined treatments suggests that even dual-plant systems are sufficient for effective Pb reduction. Best treatments (2–3mg/mL) met WHO levels, while lower-dose treatments exceed the guideline in some cases.

Silver (Ag)

It was observed in Table 1 that the silver concentration decreased significantly from 1.80 mg/L in the control to ≤ 0.06 mg/L in all treated samples. However, no significant differences were observed among the various treatments, suggesting that Ag removal was effective but not concentration-dependent. While,

Table 2 showed that the silver concentrations varied from 0.46 ± 0.04 mg/L to 0.61 ± 0.06 mg/L. The *P. americana* + *A. indica* combination uniquely resulted in the highest Ag concentration, indicating possible antagonistic interactions for Ag removal in this specific pairing. In contrast, the ternary combination produced one of the lowest Ag concentrations, highlighting the importance of extract diversity in mitigating metal-specific interactions.

Nickel (Ni)

Nickel concentration declined significantly from 2.43 mg/L in the control to between 0.94 and 0.18 mg/L in treated samples. The lowest Ni concentrations were consistently recorded in combined extract treatments at 3 mg/ml, particularly PA+AI and PA+CO+AI, indicating a synergistic effect (Table 1). Nickel concentrations ranged from 0.80 ± 0.06 mg/L to 1.093 ± 0.02 mg/L. *P. americana* alone resulted in the highest Ni concentration, while all combined treatments significantly lowered Ni levels. The ternary treatment achieved one of the lowest Ni concentrations (0.80 ± 0.06 mg/L), confirming superior remediation performance relative to single-plant extracts. Even after treatment, Ni concentrations down to about 0.18 mg/L still 9× higher than the WHO limit. Ni remains above safe drinking standards in all treatments, indicating additional removal efforts are needed.

Table 1: Heavy metal content of the treated leachate samples from landfill site

Plant extract	Conc. (mg/ml)/ratio	Fe (mg/L)	Mn (mg/L)	Cu (mg/L)	Co (mg/L)	Zn (mg/L)	Pb (mg/L)	Ag (mg/L)	Ni (mg/L)
	Control	7.86 ^a ±0.04	1.34 ^a ±0.06	0.745 ^a ±0.01	0.864 ^a ±0.02	1.840 ^a ±0.08	1.23 ^a ±0.03	1.8 ^a ±0.02	2.43 ^a ±0.07
<i>Persea americana</i>	1	0.93 ^b ±0.02	0.72 ^b ±0.04	0.484 ^c ±0.02	0.360 ^b ±0.01	0.801 ^b ±0.03	0.24 ^c ±0.01	0.04 ^b ±0.01	0.94 ^b ±0.02
	2	0.72 ^b ±0.01	0.65 ^b ±0.02	0.401 ^c ±0.03	0.018 ^e ±0.01	0.628 ^c ±0.02	0.036 ^d ±0.02	0.04 ^b ±0.01	0.62 ^d ±0.02
	3	0.68 ^b ±0.03	0.541 ^c ±0.01	0.384 ^c ±0.02	0.016 ^e ±0.01	0.472 ^d ±0.03	0.021 ^d ±0.01	0.01 ^b ±0.00	0.38 ^e ±0.01
	1	0.96 ^b ±0.07	0.426 ^c ±0.03	0.640 ^b ±0.02	0.40 ^b ±0.02	0.743 ^b ±0.01	0.33 ^b ±0.02	0.06 ^b ±0.01	0.84 ^c ±0.03
<i>Chromolaena odorata</i>	2	0.82 ^b ±0.03	0.343 ^c ±0.01	0.472 ^c ±0.04	0.026 ^e ±0.01	0.523 ^d ±0.02	0.074 ^d ±0.01	0.06 ^b ±0.02	0.36 ^e ±0.04
	3	0.74 ^b ±0.01	0.311 ^c ±0.04	0.318 ^c ±0.02	0.015 ^e ±0.01	0.374 ^d ±0.02	0.042 ^d ±0.01	0.02 ^b ±0.00	0.28 ^e ±0.01
	1	0.90 ^b ±0.04	0.462 ^c ±0.05	0.224 ^c ±0.01	0.20 ^c ±0.02	0.483 ^d ±0.05	0.20 ^c ±0.01	0.04 ^b ±0.01	0.68 ^d ±0.03
<i>Azadirachta indica</i>	2	0.80 ^b ±0.04	0.413 ^c ±0.08	0.204 ^c ±0.03	0.018 ^e ±0.01	0.401 ^d ±0.02	0.063 ^d ±0.01	0.02 ^b ±0.00	0.27 ^e ±0.01
	3	0.43 ^c ±0.02	0.384 ^c ±0.02	0.194 ^c ±0.04	0.018 ^e ±0.01	0.328 ^d ±0.03	0.018 ^d ±0.01	0.01 ^b ±0.00	0.28 ^e ±0.02
PA+ CO	1 (1:1)	0.81 ^b ±0.03	0.347 ^c ±0.03	0.573 ^b ±0.06	0.162 ^c ±0.02	0.476 ^d ±0.01	0.038 ^d ±0.01	0.04 ^b ±0.01	0.34 ^e ±0.03
	2 (1:1)	0.64 ^b ±0.01	0.306 ^c ±0.02	0.407 ^c ±0.02	0.032 ^e ±0.01	0.402 ^d ±0.02	0.022 ^d ±0.01	0.02 ^b ±0.01	0.24 ^e ±0.02
	3 (1:1)	0.40 ^c ±0.05	0.218 ^c ±0.04	0.312 ^c ±0.01	0.024 ^e ±0.01	0.316 ^d ±0.03	0.020 ^d ±0.01	0.02 ^b ±0.00	0.20 ^e ±0.03
PA + AI	1 (1:1)	0.80 ^b ±0.02	0.346 ^c ±0.06	0.214 ^c ±0.03	0.105 ^d ±0.02	0.384 ^d ±0.04	0.094 ^d ±0.02	0.01 ^b ±0.00	0.38 ^e ±0.04
	2 (1:1)	0.42 ^c ±0.01	0.296 ^c ±0.03	0.190 ^c ±0.02	0.032 ^e ±0.01	0.321 ^d ±0.02	0.063 ^d ±0.01	0.01 ^b ±0.00	0.24 ^e ±0.01
	3 (1:1)	0.36 ^c ±0.04	0.124 ^c ±0.02	0.208 ^c ±0.03	0.014 ^e ±0.01	0.276 ^d ±0.04	0.027 ^d ±0.01	0.01 ^b ±0.00	0.18 ^e ±0.01
CO + AI	1 (1:1)	0.84 ^b ±0.07	0.418 ^c ±0.04	0.326 ^c ±0.02	0.098 ^d ±0.01	0.374 ^d ±0.04	0.068 ^d ±0.01	0.03 ^b ±0.01	0.40 ^e ±0.02
	2 (1:1)	0.64 ^b ±0.05	0.302 ^c ±0.03	0.320 ^c ±0.01	0.018 ^e ±0.01	0.321 ^d ±0.02	0.018 ^d ±0.01	0.02 ^b ±0.01	0.27 ^e ±0.03
	3 (1:1)	0.58 ^b ±0.02	0.183 ^c ±0.01	0.285 ^c ±0.02	0.016 ^e ±0.01	0.274 ^d ±0.03	0.016 ^d ±0.01	0.01 ^b ±0.00	0.22 ^e ±0.02
PA+CO+AI	1 (1:1:1)	0.76 ^b ±0.01	0.211 ^c ±0.02	0.314 ^c ±0.04	0.092 ^d ±0.01	0.368 ^d ±0.02	0.043 ^d ±0.01	0.01 ^b ±0.00	0.29 ^e ±0.01
	2(1:1:1)	0.36 ^c ±0.02	0.163 ^c ±0.03	0.262 ^c ±0.02	0.017 ^e ±0.01	0.311 ^d ±0.02	0.019 ^d ±0.01	0.01 ^b ±0.00	0.26 ^e ±0.02
	3(1:1:1)	0.30 ^c ±0.01	0.115 ^c ±0.01	0.205 ^c ±0.02	0.018 ^e ±0.01	0.260 ^d ±0.03	0.016 ^d ±0.01	0.01 ^b ±0.00	0.20 ^e ±0.01
LSD		0.12	0.05	0.08	0.04	0.08	0.04	0.02	0.05

Mean with the same superscript along the vertical array indicates no significant difference (P<0.05)

Table 2: Comparison of the efficacy of the different treatment on the heavy metals in leachate samples from the landfill sites

Plant extract	Fe (mg/L)	Mn (mg/L)	Cu (mg/L)	Co (mg/L)	Zn (mg/L)	Pb (mg/L)	Ag (mg/L)	Ni (mg/L)
<i>Persea americana</i>	2.55 ^b ±0.13	0.812 ^a ±0.08	0.504 ^a ±0.06	0.315 ^a ±0.12	0.935 ^a ±0.06	0.382 ^b ±0.10	0.473 ^b ±0.08	1.093 ^a ±0.02
<i>Chromolaena odorata</i>	2.60 ^a ±0.21	0.61 ^b ±0.07	0.543 ^a ±0.06	0.326 ^a ±0.09	0.87 ^b ±0.10	0.419 ^a ±0.08	0.485 ^b ±0.05	0.978 ^b ±0.07
<i>Azadirachta indica</i>	2.50 ^c ±0.09	0.65 ^b ±0.06	0.341 ^b ±0.08	0.275 ^b ±0.04	0.763 ^c ±0.08	0.378 ^b ±0.06	0.468 ^b ±0.09	0.915 ^b ±0.10
<i>P. americana</i> + <i>C. odorata</i>	2.43 ^d ±0.12	0.55 ^b ±0.08	0.51 ^a ±0.10	0.27 ^b ±0.05	0.79 ^c ±0.04	0.33 ^c ±0.05	0.47 ^b ±0.03	0.80 ^c ±0.08
<i>P. americana</i> + <i>A. indica</i>	2.36 ^e ±0.09	0.53 ^b ±0.06	0.34 ^b ±0.07	0.25 ^b ±0.05	0.71 ^c ±0.08	0.35 ^c ±0.08	0.61 ^a ±0.06	0.81 ^c ±0.04
<i>C. odorata</i> + <i>A. indica</i>	2.48 ^c ±0.06	0.56 ^b ±0.05	0.42 ^b ±0.06	0.25 ^b ±0.04	0.70 ^c ±0.05	0.33 ^c ±0.02	0.47 ^b ±0.03	0.83 ^c ±0.05
<i>P. americana</i> + <i>C. odorata</i> + <i>A. indica</i>	2.32 ^f ±0.14	0.46 ^c ±0.09	0.38 ^b ±0.05	0.25 ^b ±0.06	0.69 ^c ±0.08	0.33 ^c ±0.05	0.46 ^b ±0.04	0.80 ^c ±0.06
LSD	0.03	0.05	0.04	0.02	0.04	0.02	0.11	0.07

Mean with the same superscript along the vertical array indicates no significant difference (P<0.05)

Table 3: Comparison of Treated Leachate Heavy Metal Concentrations with WHO Drinking Water Standards.

Metal	WHO Guideline (mg/L)	Range Treatment (mg/L)	After Compliance Status	Interpretation
Iron (Fe)	Not based* health-	0.30 – 0.96	Partial	Significant reduction achieved; higher-dose combination treatments approach acceptable aesthetic limits
Manganese (Mn)	0.5	0.115 – 0.72	Partial	High-dose and combined extracts generally meet WHO guideline; some lower doses exceed limit
Copper (Cu)	2.0	0.19 – 0.64	Compliant	All treated samples fall well below WHO guideline
Cobalt (Co)	Not specified	0.014 – 0.40	Not evaluated	WHO guideline not available; concentrations substantially reduced
Zinc (Zn)	3.0	0.26 – 0.80	Compliant	All treated samples meet WHO guideline
Lead (Pb)	0.01	0.016 – 0.24	Partial	Significant reduction achieved; some treatments still marginally exceed guideline
Silver (Ag)	Not specified	0.01 – 0.06	Not evaluated	No WHO guideline; concentrations markedly reduced
Nickel (Ni)	0.02	0.18 – 0.94	Non-compliant	Levels remain above WHO guideline in all treatments

*WHO does not provide a health-based guideline for iron, but high concentrations may affect taste, color, and acceptability of water

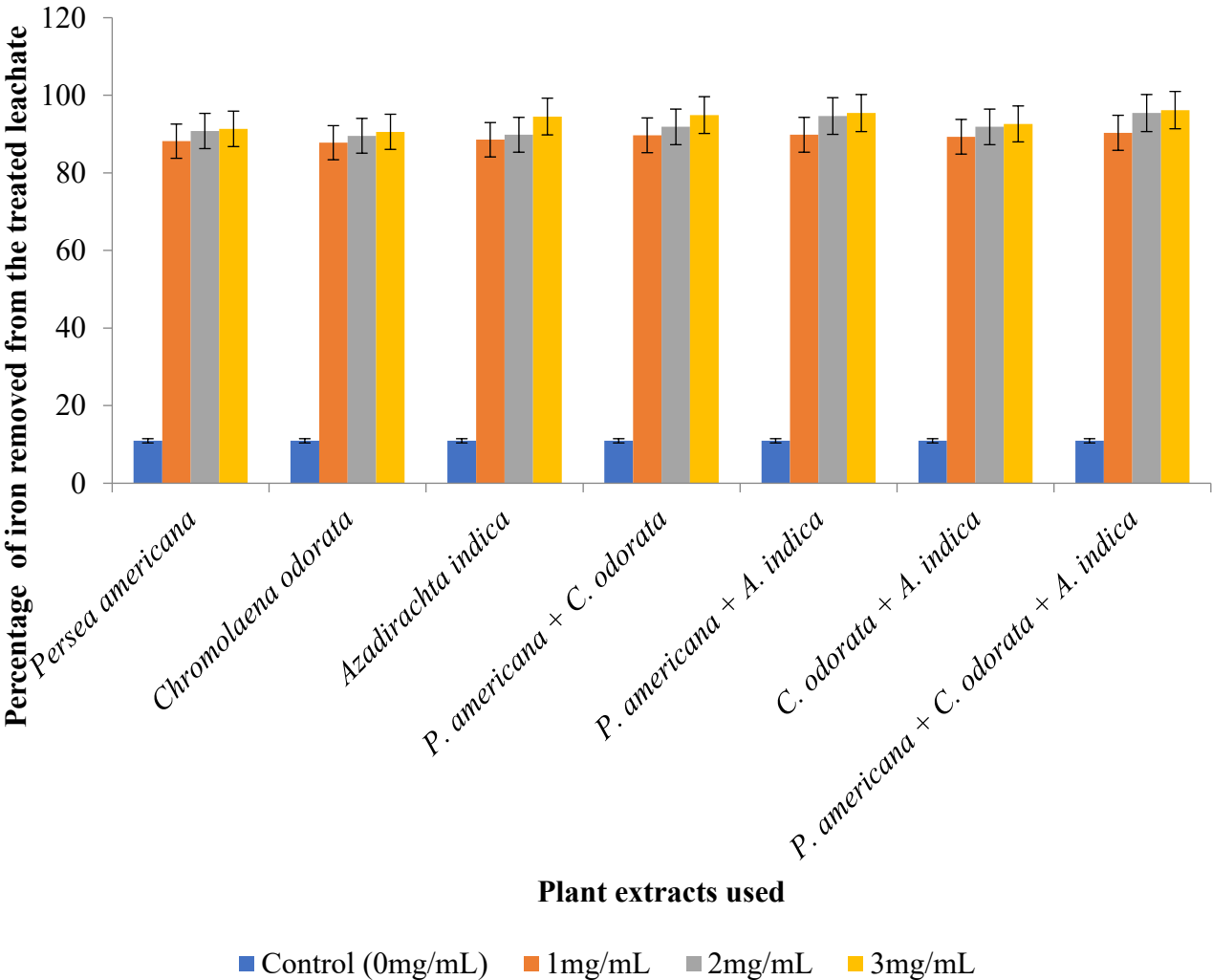


Figure 1: Percentage removal of iron from landfill leachate following treatment with ethanol leaf extracts.

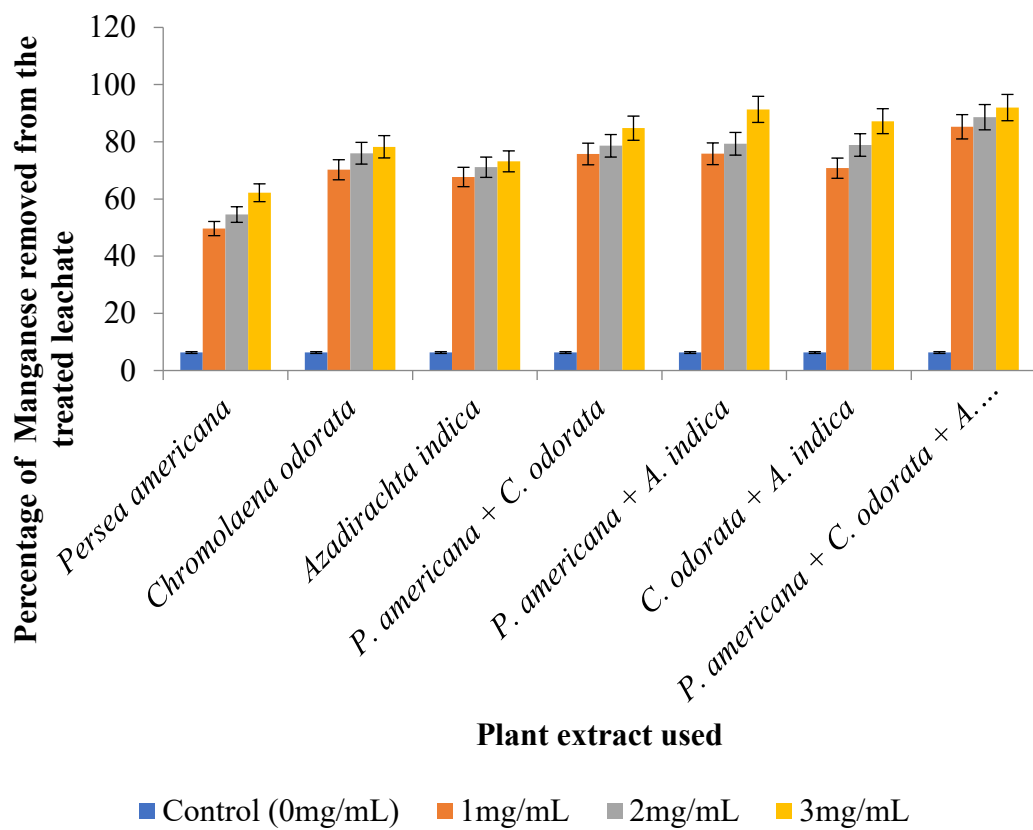


Figure 2: Percentage removal of Mn from landfill leachate following treatment with ethanol leaf extracts.

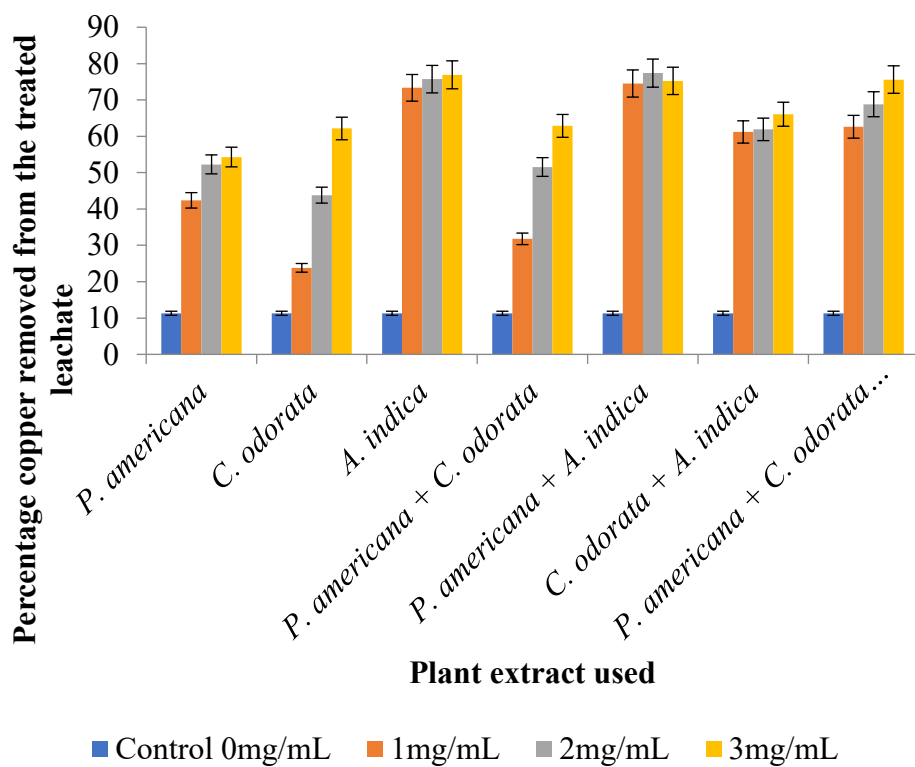


Figure 3: Percentage removal of Cu from landfill leachate following treatment with ethanol leaf extracts.

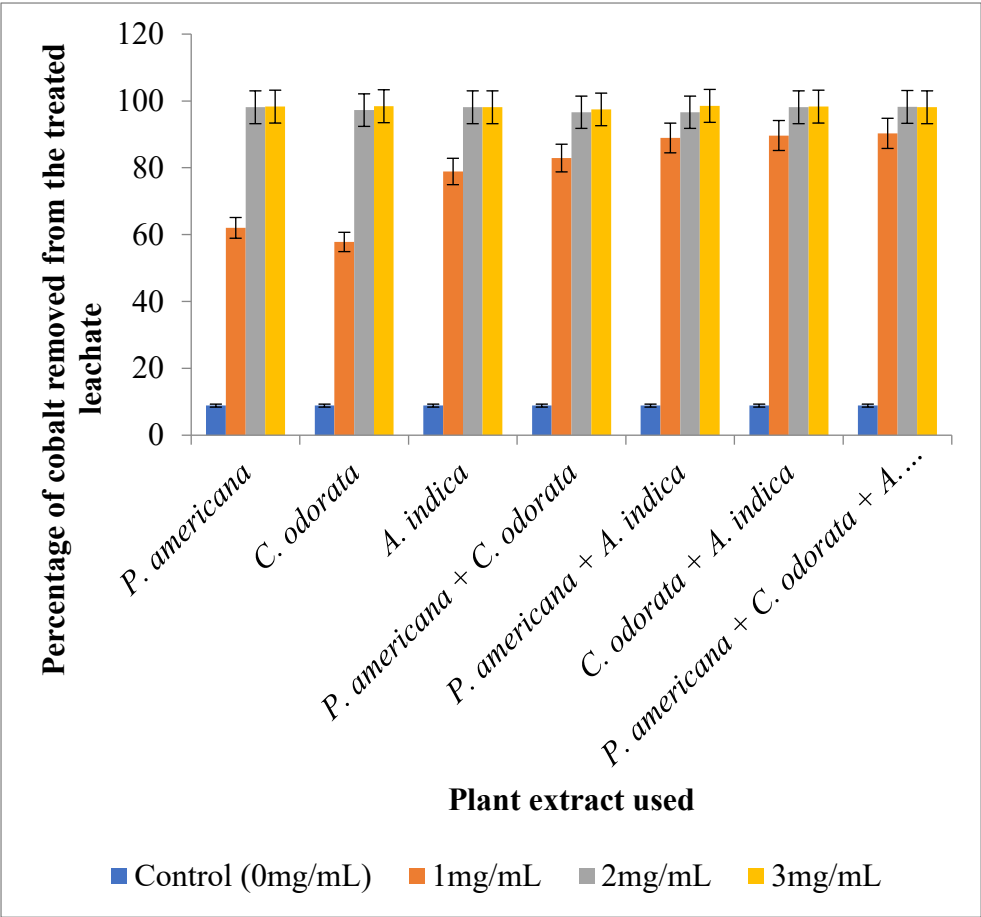


Figure 4: Percentage removal of Co from landfill leachate following treatment with ethanol leaf extracts.

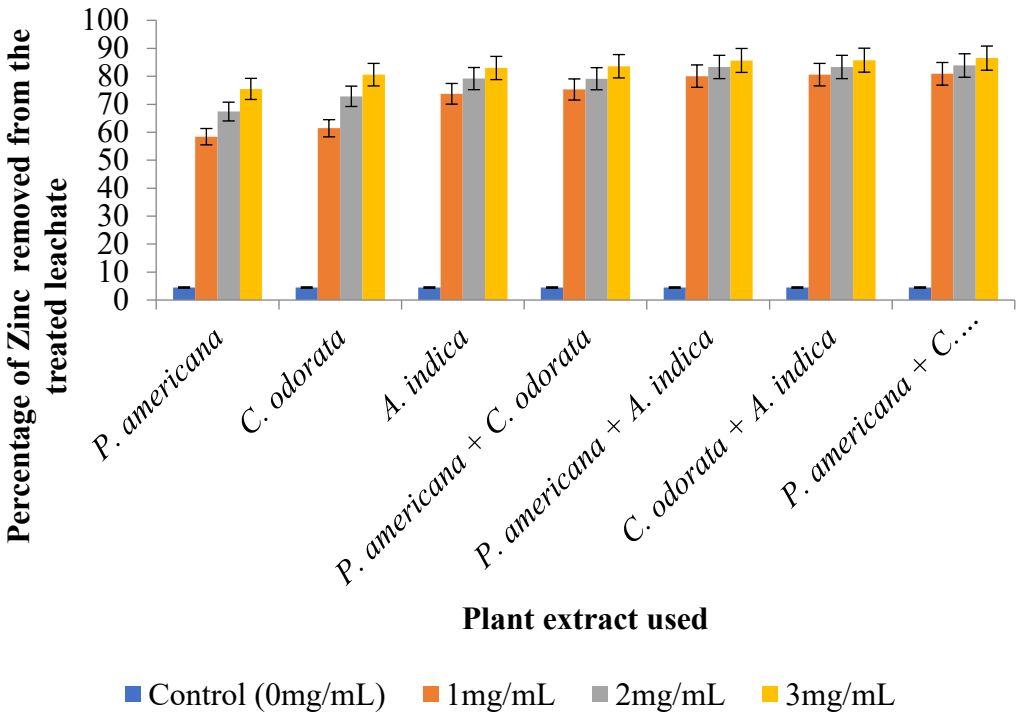


Figure 5 Percentage removal of Zn from landfill leachate following treatment with ethanol leaf extracts.

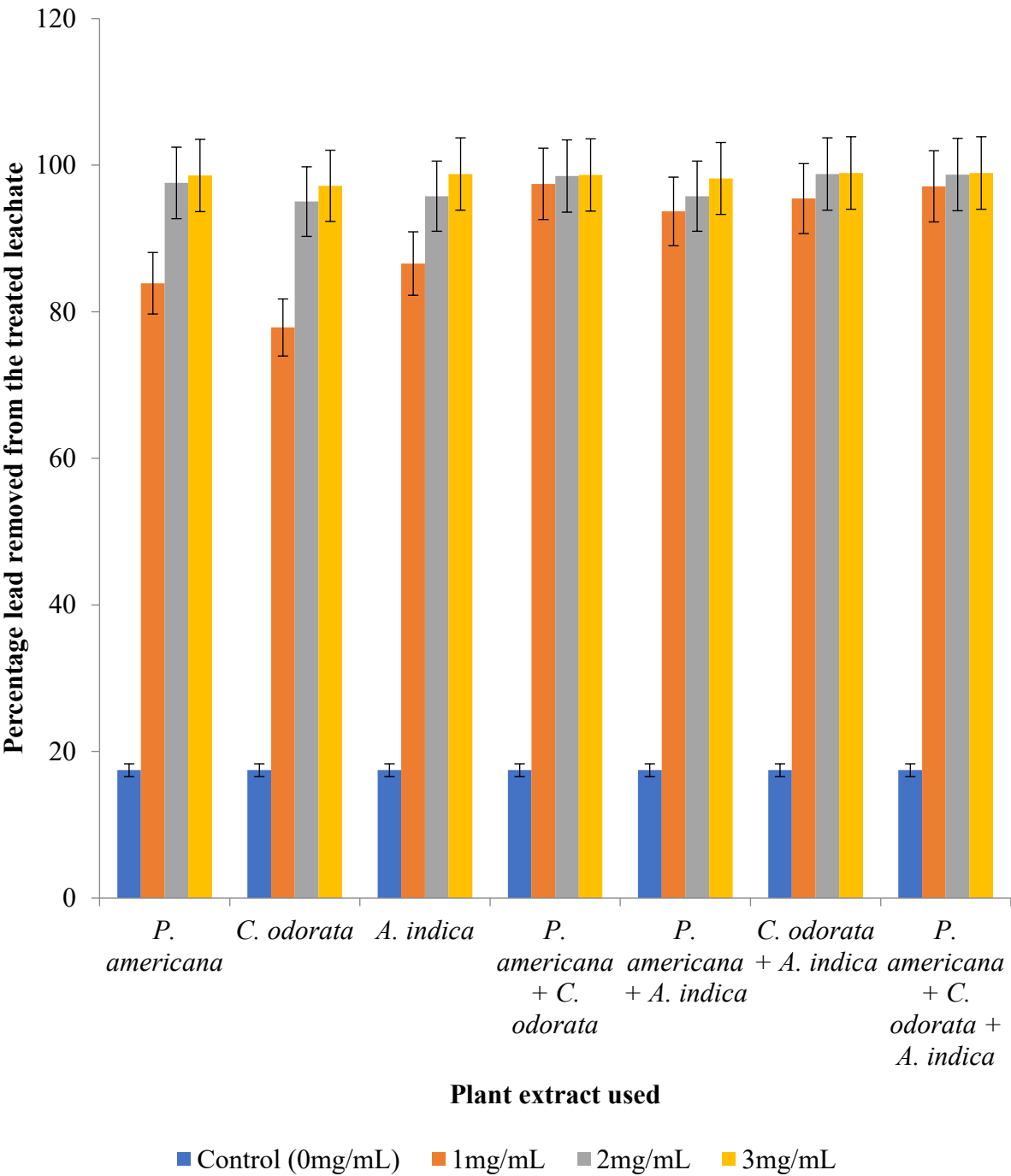


Figure 6: Percentage removal of Pb from landfill leachate following treatment with ethanol leaf extracts.

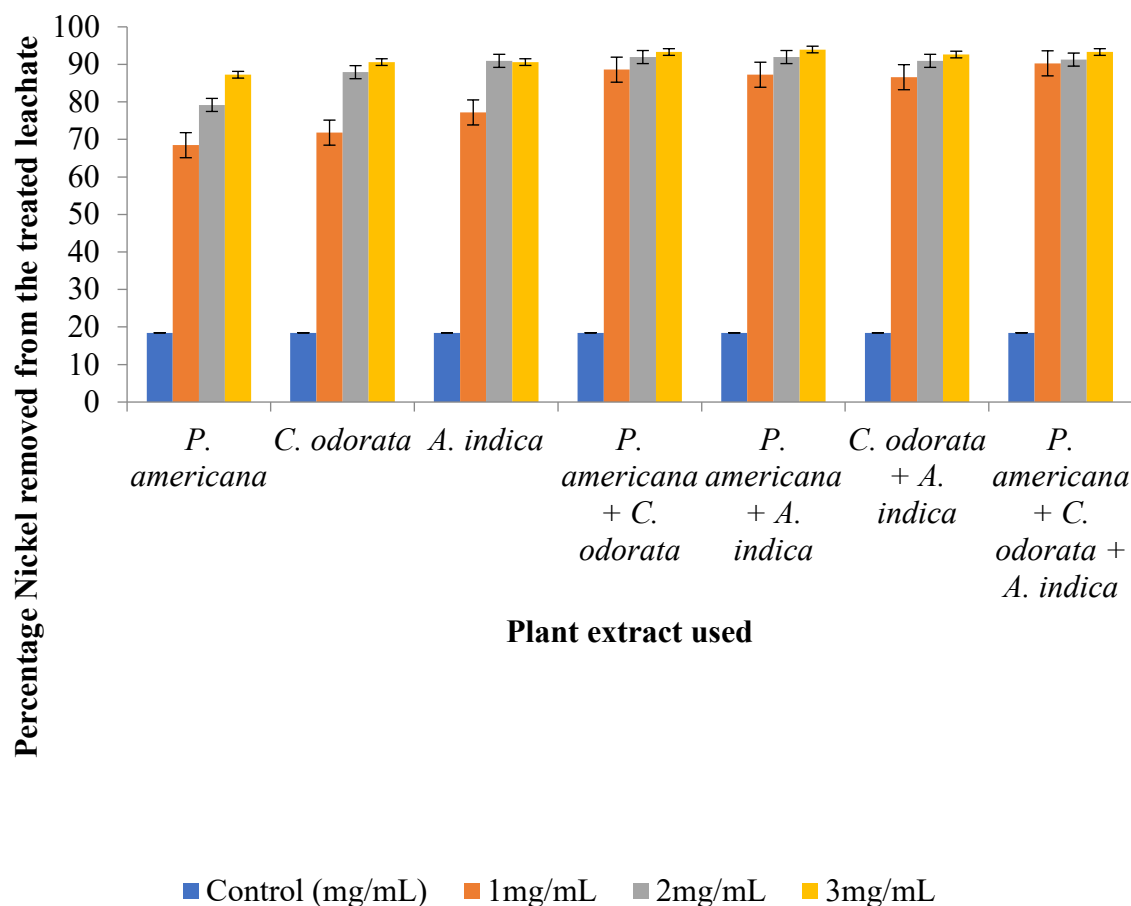


Figure 7: Percentage removal of Nickel from landfill leachate following treatment with ethanol leaf extracts

DISCUSSION

The present study demonstrates that plant-based extracts derived from *Persea americana*, *Chromolaena odorata*, and *Azadirachta indica*, applied individually and in combination, significantly influenced the concentration of heavy metals in landfill leachate. The observed reductions, particularly under combined extract treatments, are consistent with previous studies that have highlighted the effectiveness of plant-derived materials as eco-friendly and low-cost alternatives for heavy metal remediation (Ajmal *et al.*, 1998; Sud *et al.*, 2008). A major outcome of this study is the superior remediation efficiency observed in binary and ternary plant extract combinations compared to single-plant treatments. The ternary combination (*P. americana* + *C. odorata* + *A. indica*) consistently produced the lowest concentrations of most heavy metals. This finding aligns with earlier reports that combining plant materials enhances metal removal due to increased phytochemical diversity and the availability of multiple metal-binding functional groups ($-\text{OH}$, $-\text{COOH}$, $-\text{NH}_2$) (Wang & Chen, 2009; Bhattacharya *et al.*, 2018).

Previous studies have demonstrated that synergistic interactions among flavonoids, tannins, phenolics, and saponins significantly improve biosorption and chelation processes in contaminated aqueous environments (Bhatnagar & Sillanpää, 2010). The enhanced performance observed in the combined treatments in this study supports the hypothesis that multi-extract systems provide broader and more effective metal sequestration pathways than single-plant applications. Iron and manganese are common constituents of landfill leachate and are often difficult to remove using conventional treatment methods. The significant reduction in Fe and Mn concentrations observed under combined extract treatments is consistent with findings reported by Fu and Wang (2011), who noted that plant-derived organic compounds facilitate metal complexation and reduce metal solubility. Studies on *Azadirachta indica* and *Persea americana* extracts have reported strong metal-binding capacity due to their high phenolic and carboxylic acid content, which enhances chelation and adsorption processes (Ajao & Adepoju, 2017; Ojoawo *et al.*, 2020).

The progressive decrease in Fe and Mn concentrations with increasing extract complexity in the present study further supports these mechanisms. The reduction in Cu, Co, and Ni concentrations under combined treatments aligns with earlier studies indicating that these metals have a high affinity for plant-based ligands (Kurniawan *et al.*, 2006). The relatively higher concentrations observed in single-plant treatments suggest limited biosorption capacity due to fewer available binding sites. Nickel and cobalt, in particular, have been shown to respond positively to organic-rich remediation systems, where complexation and ion exchange processes dominate metal removal (Volesky, 2007). The improved removal efficiency observed in binary and ternary treatments in this study corroborates these findings and highlights the importance of phytochemical diversity in metal immobilization. Zinc and lead exhibited substantial reductions under combined extract treatments, especially those involving *A. indica*. Neem extracts have been extensively documented for their strong affinity for heavy metals, attributed to bioactive compounds such as azadirachtin and nimbin (Tripathi *et al.*, 2014). Soraganvi, & Desai (2024) reported a 97.2% and 96.34 % Pb removal from synthetic leachate using TiO₂ and Ag-doped TiO₂ nanomaterials by a heterogeneous photocatalytic process. The lack of significant differences among some combined treatments for Pb suggests that even dual-plant systems may be sufficient for effective lead removal. Similar observations have been reported in previous biosorption studies, where diminishing returns were observed beyond a certain level of phytochemical complexity (Sud *et al.*, 2008). Silver exhibited less consistent behavior compared to other metals, with certain binary combinations resulting in higher Ag concentrations. Such metal-specific interactions emphasize the need for careful evaluation of individual metal behavior when designing plant-based remediation systems. Overall, the findings of this study reinforce existing literature supporting the use of plant-derived materials for landfill leachate and wastewater treatment. Compared to conventional physicochemical methods, plant-based remediation approaches offer advantages such as biodegradability, reduced chemical input, and minimal secondary pollution (Fu & Wang, 2011). The enhanced performance of combined plant extracts suggests that optimized formulations of *P. americana*, *C. odorata*, and *A. indica* could be developed into practical remediation agents, particularly in developing regions where cost-effective and sustainable treatment options are required (Bhattacharya *et al.*, 2018).

CONCLUSION

In conclusion, the combined use of these three plant species represents an effective green remediation strategy capable of significantly reducing heavy metal contamination in landfill leachate while contributing to sustainable environmental management and protection of public health. However, further research focusing on adsorption kinetics, long-term stability of metal–phytochemical complexes, and field-scale validation is recommended to fully establish the applicability of these findings

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