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Phytoremediation Potential of Landfill Vegetation in Aceh, Indonesia: Linking Pb Accumulation with Chlorophyll Content and Stomatal Traits

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ABSTRACT

Landfills are a major source of heavy metal contamination, particularly lead (Pb), which poses serious risks to the environment and health. This study aims to evaluate the phytoremediation potential of plant species in the Gampong Jawa landfill, Banda Aceh, by analyzing Pb accumulation, chlorophyll content, and stomatal characteristics. Leaf samples were collected from plant species in the Gampong Jawa landfill using a purposive sampling approach. Pb levels were measured using Atomic Absorption Spectrophotometry (AAS), chlorophyll content was determined spectrophotometrically at 645 and 663 nm, and stomatal traits were observed from paradermal sections of the abaxial epidermis. A total of 21 plant species from 13 families were identified, of which 13 species accumulated Pb with concentrations ranging from 0.004 to 0.208 ppm. *Mangifera indica* exhibited the highest Pb accumulation, along with the highest stomatal density (113–560 mm^{-2}) and stomatal index (10.4–33.2%). A consistent trend was observed in which species with higher stomatal density tended to show greater Pb accumulation, while increased Pb levels were associated with reduced chlorophyll content. These findings suggest that stomatal characteristics may influence Pb uptake in plants. The findings highlight that *M. indica* has potential as a phytoremediator and bioindicator of Pb pollution to support landfill rehabilitation and environmental management.

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INTRODUCTION

Rapid population growth and industrial expansion have led to a sharp rise in waste production, overwhelming current waste management systems and placing excessive stress on ecosystems (Asif et al., 2023). Pollution has become a pressing public health concern, as exposure to contaminants significantly increases the likelihood of various human diseases. Soil, air, and water pollution pose major obstacles for public health policies, driven by the high cost of environmental restoration and limited technological solutions (Cristaldi et al., 2020). Among the most harmful pollutants is heavy metal contamination, which persists in the environment without naturally breaking down, creating severe risks to plants, animals, aquatic organisms, and human health (Khan et al., 2023).

One of the primary sources of heavy metals is the increasing number of landfills, driven by rising consumption and population growth. Proper management and treatment of landfill sites are essential to minimize the presence of pollutants, including heavy metals (Stojanovska & Kostovska, 2022). These pollutants can seep into soil and water, causing severe contamination that threatens human health and surrounding biodiversity (Morales et al., 2021). Heavy metal contamination in landfill areas is often associated with leachate discharge, which contains various toxic elements that gradually infiltrate the soil and environment. Therefore, effective strategies are needed to reduce the environmental impact of landfill-derived pollutants (Teng et al., 2021).

Lead (Pb) is one of the most hazardous heavy metals commonly found in landfill environments, posing serious risks to human health and ecosystems due to its toxicity and persistence (Khan et al., 2023; Siraj et al., 2022; Yugatama et al., 2019). In landfill areas, Pb contamination primarily originates from leachate infiltration into the surrounding soil, which can subsequently affect plant growth and physiological processes. Phytoremediation has been widely recognised as a cost-effective and environmentally friendly approach for mitigating heavy metal contamination through the use of plants (Ruiz et al., 2020). However, the effectiveness of phytoremediation depends on the ability of plant species to tolerate and accumulate heavy metals under specific environmental conditions. Previous studies have shown that Pb exposure is associated with changes in plant anatomical traits, particularly stomatal characteristics, and may also affect physiological responses such as chlorophyll content under certain conditions (Sulistiana & Setijorini, 2016; Fida et al., 2021).

A landfill site located in Gampong Jawa Village, Banda Aceh, Sumatra, was selected as the study area due to its high potential for heavy metal contamination, as indicated by previous soil analyses showing the presence of Pb (Amalia et al., 2023). In addition, the site is characterised by diverse vegetation that has adapted to contaminated conditions, making it a suitable location for evaluating phytoremediation potential under real environmental settings. Therefore, this study aims to evaluate the phytoremediation potential of plant species in the Gampong Jawa landfill by analysing Pb accumulation, chlorophyll content, and stomatal characteristics. Insights into the Pb uptake by these plants are expected to provide a scientific basis for selecting suitable species for landfill rehabilitation and environmental management.

However, studies integrating Pb accumulation, chlorophyll content, and stomatal characteristics in plant species growing in landfill environments, particularly in tropical urban areas such as Banda Aceh, are still limited. Therefore, this study aims to evaluate the phytoremediation potential of plant species in the Gampong Jawa landfill by analyzing Pb accumulation, chlorophyll content, and stomatal properties. The novelty of this study lies in its integrative approach that combines chemical, physiological, and anatomical analyses to better understand plant responses to Pb contamination. This study is expected to contribute to the development of scientific knowledge on phytoremediation mechanisms and provide practical insights for selecting appropriate plant species for landfill rehabilitation and environmental management.

RESEARCH METHODS

This study used a descriptive research approach to analyse Pb accumulation, chlorophyll content, and stomatal characteristics of plant species in a landfill environment. Research samples were collected from the final Gampong Jawa, Banda Aceh, Sumatra disposal site (Figure 1). Lead (Pb) content was analyzed in the soil and plant leaves at the Environmental Quality Testing Laboratory, Department of Chemical Engineering, Faculty of Engineering, Syiah Kuala University. Plant species identification was carried out at the Herbarium Acehense. At the same time, anatomical observations and chlorophyll content analysis were performed at the Structure and Development Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Syiah Kuala University.

Tools and Materials

Tools used in this research include a hand microtome, polystyrene, light microscope, petri dish, brush, razor blade, object glass, cover glass, oven, multiware microwave, vessel tubes, atomic absorption spectrophotometer, porcelain mortar, test tubes, and UV-Vis spectrophotometer. The materials consist of leaf samples from the landfill area, representing 21 plant species, filter paper, gloves, and tissue. The chemicals used in this research include distilled water (aqua dest), clorox solution, alcohol solutions of varying concentrations (100%, 95%, 70%, and 50%), 37% formalin, safranin, fast green, canada balsam, glacial acetic acid, nitric acid (HNO_3 65% and 0.2%), hydrochloric acid (HCl), and 80% acetone.

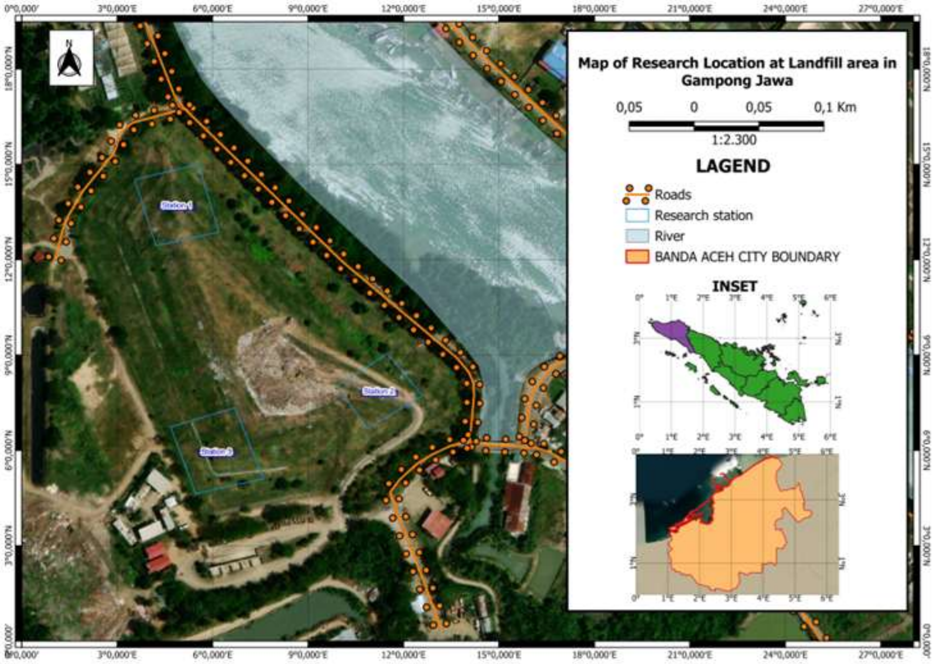

Figure 1. Map of research location at the landfill area in Gampong Jawa, Banda Aceh, Sumatra

Sample Collection Method

The samples collected in this study consist of leaves from plants distributed across the Gampong Jawa landfill site in Banda Aceh. Leaf sampling was conducted using an exploratory method with purposive sampling techniques, aimed at selecting representative plant species growing in the study area. The sampling area was determined at a distance of 30–50 m from the peak of the waste pile at the landfill site, based on field accessibility and the presence of vegetation.

The number of individuals per species varied depending on their availability in the study area. Plants selected as samples were those that appeared healthy and had sufficient leaves for analysis. From each selected plant, several leaves were collected from the same position (for example, leaves exposed to direct sunlight) to ensure consistency in the analysis.

Lead (Pb) Content

Leaf samples (0.05 g per sample) were weighed and placed in vessel tubes, followed by the addition of 9 ml of concentrated nitric acid (HNO_3 65%). The vessel tubes were sealed and heated in a multiwave microwave for 15 minutes at 160°C with a pressure of 600 atm, then cooled for 15 minutes. After cooling, the samples were filtered, centrifuged, and transferred into bottles, then diluted to a volume of 50 ml using 0.2% HNO_3 solution. The solution was measured using Atomic

Absorption Spectrophotometry (AAS). The Pb analysis was conducted in triplicate for 21 plant species.

Chlorophyll Content

Chlorophyll measurement was conducted at the Plant Anatomy Laboratory, FMIPA. Fresh leaf samples (1 g) were weighed and ground using a porcelain mortar. The ground leaf samples were placed in test tubes, and 10 ml of 80% acetone was gradually added, followed by filtration using filter paper. The absorbance of the filtrate was measured with a UV-Vis spectrophotometer at wavelengths of 645 nm and 663 nm.

Plant Anatomy

The plant species used were the 21 species found in the landfill area. The plant organs collected included leaves (midrib, lamina, and petiole) for anatomical observation. Leaf samples were taken from the fifth to tenth nodes from the shoot tip, assuming these leaves were neither too young nor too old. Leaf anatomical slides were prepared using the non-paraffin method. Leaf samples (2 x 2 cm) were cut from the field and fixed with FAA solution (Formalin-Alcohol-Acetic Acid) for tough leaves and spirit solution (Alcohol-Acetic Acid) for softer leaves. The samples were brought to the laboratory and sectioned using a hand microtome. Before sectioning, samples were placed in petri dishes containing distilled water. The samples were sandwiched between polystyrene and inserted into the hand microtome. The sections were placed in petri dishes with 10% clorox solution and left for 5 minutes to clean the plant tissues. The samples were rinsed three times with distilled water to remove Clorox residues, ensuring they could absorb other chemicals. The sections were stained using the following process: Safranin (3-5 minutes) – rinse 3x – fast green (3-5 minutes). Dehydration was performed using a series of alcohol solutions (50%-70%-95%-100%) for 5 minutes each. The slides were mounted with canada balsam and covered with cover glass. The slides were dried in an oven at 60°C for one week.

Observation of Stomata

Leaf epidermis slides were prepared by making paradermal sections of the leaf surface. Adaxial (upper surface) sections were made to observe the abaxial (lower surface) epidermis. This method allowed the observation of stomatal types, density, and index. Leaf samples (2 x 2 cm) were cut from the field and fixed with FAA solution for tough leaves and Spirit solution for softer leaves. The samples were rinsed with distilled water. The epidermis was carefully separated from the mesophyll by gently scraping with a scalpel. The epidermal layer was transferred to a clean petri dish and stained with safranin for 3-7 minutes. The samples were rinsed three times with distilled water. Dehydration was conducted using a series of alcohol solutions (50%-70%-95%-

100%) for 5 minutes each. The slides were mounted with canada balsam and covered with cover glass, then dried in an oven at 60°C for one week. Stomatal density and index were then calculated.

Data Analysis

Data analysis was conducted using qualitative and quantitative descriptive approaches. Qualitative analysis included descriptions of plant species and anatomical structures, while quantitative data, including Pb content and chlorophyll levels, were presented descriptively. The relationships between variables were interpreted as observed patterns or associations without applying inferential statistical tests.

RESEARCH RESULT

Plants Identified in the Waste Disposal Site Area of Gampong Jawa and Pb Content

The plants identified around the waste disposal area of Gampong Jawa consist of 21 species from 13 families (Table 1). Fabaceae dominates with four plant species, namely lamtoro (*Leucaena leucocephala*), trembesi (*Samanea saman*), tamarind (*Tamarindus indica*), and narra (*Pterocarpus indicus*). Several other families have two plant species: Anacardiaceae (mango and kedondong), Combretaceae (ketapang and golden ketapang), Meliaceae (neem and mahogany), Myrtaceae (Java plum and guava), and Poaceae (string bamboo and lemongrass). The remaining seven families are each represented by one plant species.

Of the 21 identified species, 13 species (61.9%) showed detectable Pb content in their leaves, whereas 8 species (38.1%) showed no detectable Pb (Table 1). Across all sampled species, Pb content ranged from 0 to 0.208 ppm, while among species with detectable Pb, the values ranged from 0.004 to 0.208 ppm. The highest Pb content was found in *Mangifera indica* (0.208 ppm), followed by *Ficus benjamina* (0.157 ppm), *Mimusops elengi* (0.151 ppm), and *Tamarindus indica* (0.135 ppm), whereas the lowest detectable Pb content was recorded in *Monoon longifolium* (0.004 ppm).

Based on the species recorded in Table 1, most plants with detectable Pb were woody species (trees or shrubs), whereas among the grass-like species only *Bambusa multiplex* showed detectable Pb (0.055 ppm), while *Cymbopogon citratus* showed no detectable Pb.

Table 1. Plants identified around the waste disposal area and Pb content in leaves

Number	Latin Name	Local Name	Family	Pb Content (ppm)
1	*Mangifera indica*	Mangga	Anacardiaceae	0.208
2	*Spondias pinnata*	Kedondong	Anacardiaceae	0.008
3	*Monoon longifolium*	Glodokan Tiang	Annonaceae	0.004
4	*Calotropis gigantea*	Biduri	Apocynaceae	0
5	*Terminalia catappa*	Ketapang	Combretaceae	0
6	*Terminalia mantaly*	Ketapang Kencana	Combretaceae	0
7	*Leucaena leucocephala*	Lamtoro	Fabaceae	0.064

Number	Latin Name	Local Name	Family	Pb Content (ppm)
8	*Samanea saman*	Trembesi	Fabaceae	0.061
9	*Tamarindus indica*	Asam Jawa	Fabaceae	0.135
10	*Pterocarpus indicus*	Angsana	Fabaceae	0
11	*Azadirachta indica*	Mimba	Meliaceae	0.044
12	*Swietenia mahagoni*	Mahoni	Meliaceae	0.034
13	*Ficus benjamina*	Beringin	Moraceae	0.157
14	*Syzygium cumini*	Jamblang	Myrtaceae	0
15	*Psidium guajava*	Jambu	Myrtaceae	0
16	*Bougainvillea* spp.	Bunga Kertas	Nyctaginaceae	0.053
17	*Phyllanthus acidus*	Cermai	Phyllanthaceae	0
18	*Bambusa multiplex*	Bambu Tali	Poaceae	0.055
19	*Cymbopogon citratus*	Serai	Poaceae	0
20	*Morinda citrifolia*	Mengkudu	Rubiaceae	0.015
21	*Mimusops elengi*	Tanjung	Sapotaceae	0.151

Chlorophyll and Stomata

Chlorophyll and stomatal analyses were conducted only on the 13 plant species with detectable Pb content in their leaves (Table 2). Chlorophyll content ranged from 15.55 mg/L to 53.58 mg/L, with the highest value recorded in *Ficus benjamina* (53.58 mg/L), followed by *Bambusa multiplex* (38.02 mg/L) and *Azadirachta indica* (37.00 mg/L), whereas the lowest value was found in *Spondias pinnata* (15.55 mg/L). Stomatal density ranged from 113 to 560 stomata/mm², with the highest value observed in *Mangifera indica* (560 stomata/mm²), followed by *Samanea saman* (541 stomata/mm²) and *Bambusa multiplex* (484 stomata/mm²), while the lowest value was found in *Mimusops elengi* (113 stomata/mm²). The stomatal index ranged from 10.4% to 33.2%, with the highest value found in *Mangifera indica* (33.2%), followed by *Bambusa multiplex* and *Spondias pinnata* (32.0%), whereas the lowest value was recorded in *Ficus benjamina* (10.4%). In general, species with relatively high Pb content did not always show the highest chlorophyll content. For example, *Mangifera indica* had the highest Pb content and the highest stomatal density and stomatal index, whereas *Ficus benjamina* had the second-highest Pb content but the highest chlorophyll content and the lowest stomatal index.

Observation of the stomata on the abaxial (lower) leaf surface revealed three stomatal types, namely anomocytic, paracytic, and anisocytic (Figure 2). The anomocytic type was found in *Samanea saman, Mangifera indica, Spondias pinnata, Azadirachta indica, Ficus benjamina, Bougainvillea spp., Swietenia mahagoni*, and *Bambusa multiplex*; the paracytic type was found *in Leucaena leucocephala, Tamarindus indica, Morinda citrifolia*, and *Monoon longifolium*; and the anisocytic type was found only in *Mimusops elengi*. Thus, anomocytic stomata were the most common type, occurring in 8 of 13 species (61.5%), followed by paracytic stomata in 4 species (30.8%), and anisocytic stomata in 1 species (7.7%).

Table 2. Chlorophyll content, stomata density, and stomata index in plant leaves containing Pb

Latin Name	Chlorophyll Content (mg/L)	Stomata Density (stomata/mm²)	Stomata Index (%)
Azadirachta indica	37.00	409	11.9
Bambusa multiplex	38.02	484	32.0
Bougainvillea spp.	24.99	176	12.3
Ficus benjamina	53.58	245	10.4
Mangifera indica	33.43	560	33.2
Mimusops elengi	26.00	113	11.7
Monoon longifolium	23.69	296	19.8
Morinda citrifolia	33.50	252	25.2
Leucaena leucocephala	32.15	333	25.1
Samanea saman	30.61	541	31.5
Spondias pinnata	15.55	453	32.0
Swietenia mahagoni	33.81	428	28.1
Tamarindus indica	26.35	352	30.3

Stomatal density ranges from 113 to 560/mm². The highest stomatal density is found in *M. indica* leaves, while the lowest stomatal density is found in *M. elengi* leaves. The stomatal index of leaves also varies, ranging from 10.4% to 33.2%. The highest stomatal index is found in *M. indica* leaves, while the lowest stomatal index is found in *F. benjamina* leaves.

Observation of stomata on the abaxial (lower) surface of leaves reveals three main types: anomocytic, paracytic, and anisocytic (Figure 2). The anomocytic type is found in *S. saman, M indica, S. pinnata, A. indica, F. benjamina, Bougainvillea spp., S. mahagoni,* and *B. multiplex*. The paracytic type is found in *L. leucocephala, T. indica, M. citrifolia*, and *M. longifolium*. The anisocytic type is found in *M. elengi*. Anomocytic type stomata are more commonly found than paracytic and anisocytic types. Stomata or epidermal pores generally bordered by two guard cells, play a crucial role in gas exchange in plants.

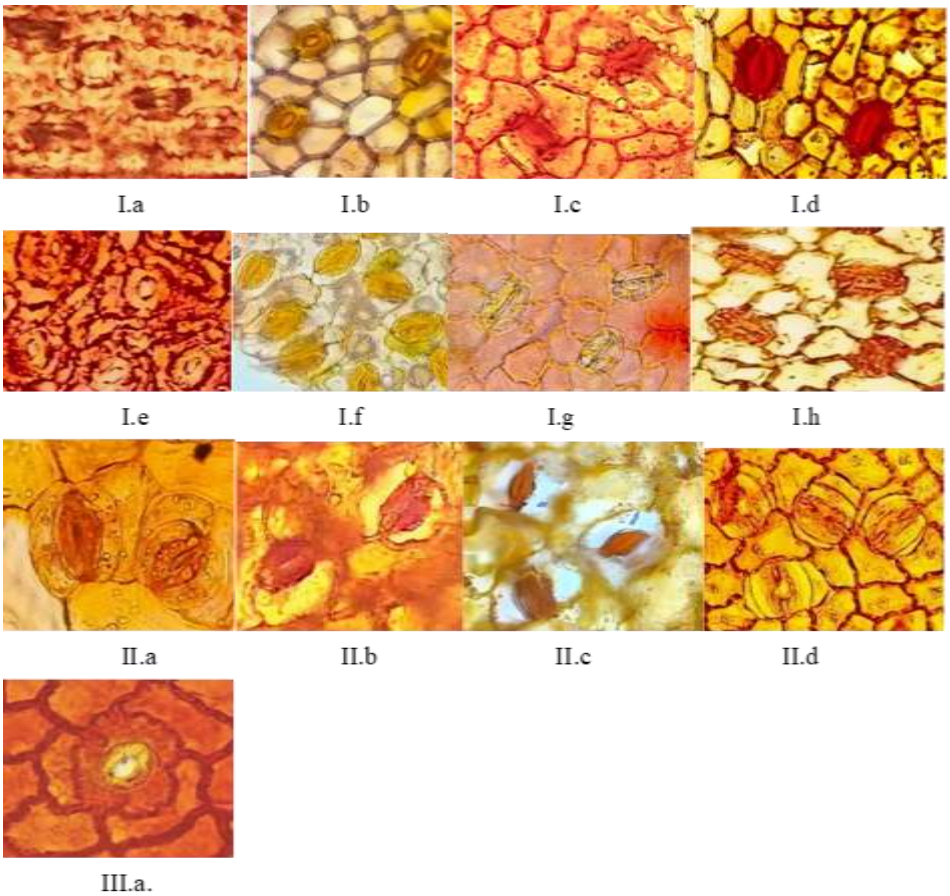

Figure 2. Stomata type in plant leaves containing Pb (40x magnification) Anomocytic (L.a. *Bambusa multiplex*, I.b. *Azadirachta indica*, L.c. *Bougainvillea spp*., I.d. *Ficus benjamina*. I.e. *Mangifera indica*, I.f. *Samanea saman*, I.g. *Spondias pinnata*. I.h. *Swietenia mahagoni*); Paracytic (II.a. *Morinda citrifolia*, II.b. *Leucaena leucocephala*, II.c. *Tamarindus indica*, II.d. *Monoon longifolium*); anisocytic (III.a. *Mimusops elengi*).

DISCUSSION

This study showed that 13 of the 21 identified plant species accumulated Pb in their leaves, with *Mangifera indica* showing the highest Pb content, while *Ficus benjamina* had the highest chlorophyll content and *M. indica* had the highest stomatal density and stomatal index. These findings suggest that Pb uptake capacity differed among species and may be associated with differences in their physiological and anatomical characteristics. Although Fabaceae was the most represented family in the study area, the species with the highest Pb accumulation did not belong to this family, indicating that species abundance did not necessarily correspond to Pb uptake capacity. The dominance of Fabaceae in the site may be related to their nitrogen fixation ability, which enables them to survive under stressed soil conditions through symbiosis with nitrogen-fixing bacteria such as Rhizobium (Foyer et al., 2016; Prasad & Freitas, 2003).

Although the Fabaceae family has the highest number of species, the plant that absorbed the most Pb did not belong to this family but to Anacardiaceae, namely *M. indica*. This species has been explored for its phytoremediation potential, as plants of this type may help eliminate, stabilize, or reduce environmental pollutants. Studies suggest that *M. indica* contributes to air pollution reduction through phylloremediation, in which leaves and their associated microbes may help minimize harmful contaminants in the environment (Mathew et al., 2022). In addition, each plant species may differ in Pb uptake due to variation in root architecture, transporter proteins, and interactions with soil microbes, which together influence the efficiency of metal absorption and translocation within the plant system (Plociniak et al., 2023). These species-specific differences may help explain why *M. indica* showed the highest Pb accumulation despite not being the most abundant family representative in the study area.

Among the 13 plant species evaluated at the study site, *M. indica* demonstrated the highest capacity for lead (Pb) absorption. This finding aligns with earlier studies indicating that mango leaves can bioaccumulate Pb (Franca et al., 2010; Adwiwartika, 2020). In addition to foliar tissues, other anatomical structures—including the stem (particularly the bark), fruit (notably the peel), and seeds—have also been shown to accumulate significant levels of Pb (Asuquo & Barde, 2020; Krishnani et al., 2021; Saiyidah et al., 2016). The physiological adaptation of mango to Pb exposure is marked by an upregulation of peroxidase enzyme activity, which increases in response to elevated Pb concentrations (Intan et al., 2023).

In the present study, *M. indica* did not show the highest chlorophyll content, whereas *F. benjamina* had the highest chlorophyll level despite showing lower Pb accumulation than *M. indica*. This comparison highlights contrasting physiological responses to Pb stress, where higher Pb uptake did not necessarily correspond to higher photosynthetic pigment levels. In *M. indica*, relatively high Pb accumulation combined with lower chlorophyll content may indicate a greater physiological burden under Pb stress, whereas *F. benjamina* may have experienced less disruption to chlorophyll biosynthesis. Previous studies have shown that Pb exposure may reduce chlorophyll concentration, particularly under prolonged or high-dose exposure (Supriatno et al., 2018). This occurs because Pb can disrupt nutrient homeostasis and interfere with chlorophyll biosynthesis, particularly by affecting magnesium and iron availability, which are essential for photosynthesis (Bouziani et al., 2023). Pb stress can also increase reactive oxygen species (ROS), such as hydrogen peroxide (H_2O_2) and superoxide anions (O_2^-), which may damage membranes, proteins, and chloroplast structures if not adequately neutralized (Wang et al., 2012). In this context, increased peroxidase activity previously reported in *M. indica* under Pb exposure may represent an important tolerance mechanism. At the molecular level, peroxidase helps detoxify ROS, particularly H_2O_2, thereby reducing oxidative damage, limiting lipid peroxidation, and maintaining cellular integrity under metal stress (Intan et al., 2023; Wang et al., 2012). Therefore, although *M. indica* showed lower chlorophyll content than *F. benjamina*, its higher Pb accumulation may reflect a stronger tolerance strategy supported by antioxidative defense rather than the maintenance of photosynthetic pigments alone.

Stomatal density data showed that *M. indica* had the highest stomatal density among the studied species. This finding suggests that stomatal density may be associated with Pb uptake capacity, as stomata function in gas exchange and may serve as entry points for airborne pollutants such as Pb. Similar patterns have been reported in previous studies, although the strength of this association may vary depending on species-specific traits and environmental conditions (Sulistiana & Setijorini, 2016; Haworth et al., 2021).

M. indica and *F. benjamina*, which showed relatively high Pb accumulation in this study, were both characterised by anomocytic stomata. Previous anatomical studies have reported the presence of anomocytic stomata in these species (Muti'ah, 2022; Mutaqin et al., 2017). Similar findings have shown that *M. indica* and *F. benjamina* have considerable potential for Pb uptake in polluted environments (Intan et al., 2023; Yasin et al., 2024). Although the direct role of stomatal type in Pb uptake efficiency remains unclear, stomatal characteristics may contribute to plant responses under Pb stress and may serve as supportive indicators of adaptation to contaminated environments (Gray et al., 2020; Wang & Chen, 2020).

This study has several limitations that should be considered when interpreting the findings. First, although Pb contamination in the study area was identified and Pb accumulation in leaves was measured, the study did not comprehensively examine Pb distribution along the soil–root–leaf pathway, particularly at the root level. Second, the relationships among Pb content, chlorophyll content, and stomatal characteristics were interpreted descriptively without inferential statistical tests. Therefore, the observed patterns should not be interpreted as statistically supported relationships. Third, other environmental factors that may influence Pb availability and plant uptake, such as soil pH and soil texture, were not evaluated in this study. Future research is needed to integrate soil, root, and leaf analyses with environmental measurements and appropriate statistical testing to better explain Pb uptake mechanisms and phytoremediation potential.

CONCLUSION

This study demonstrates that plant species growing in the Gampong Jawa landfill differ in their Pb accumulation, chlorophyll content, and stomatal characteristics, indicating that phytoremediation potential is influenced by species-specific physiological and anatomical responses to Pb stress. These findings highlight the importance of integrating Pb accumulation, chlorophyll content, and stomatal traits to better understand plant adaptation in contaminated environments. In this context, *Mangifera indica* and *Ficus benjamina* may be considered promising species for phytoremediation and supportive bioindicators of Pb pollution in landfill areas. Future research should further investigate the specific mechanisms underlying Pb uptake in these species, including antioxidative responses and anatomical adaptations, and evaluate the long-term sustainability of their application at a larger ecological scale.

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CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest regarding the publication of this paper.

KREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Amalia, A: software, methodology, writing – original draft, resources. **Amalia**: conceptualization, methodology, data curation, formal analysis. **Zumaidar**: funding acquisition, writing – review and editing, visualization, supervision, project administration, validation.

DECLARATION OF THE USE OF AI

The authors acknowledge the use of generative AI tools for language editing, grammar correction, and improving readability during manuscript preparation. No generative AI tool was used to generate research data and perform data analysis. The authors have reviewed and approved the final manuscript.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author.

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