

Type of the Paper (Original Research)

# Lead (Pb) Bioaccumulation and Physiological Responses of *Acanthus ilicifolius* in the Mangrove Forest, Wonorejo, Surabaya, East Java, Indonesia

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Key Words	<i>Acanthus ilicifolius</i> , Bioindicator, Translocation factor, Heavy metals, Lead bioaccumulation, Mangrove forest, Chlorophyll
DOI	<a href="https://doi.org/10.46488/NEPT.2026.v25i04.D1908">https://doi.org/10.46488/NEPT.2026.v25i04.D1908</a> (DOI will be active only after the final publication of the paper)
Citation for the Paper	Rachmadiarti, F., Winarsih, Fitrihidajati, H., Purnomo, T., Kunt-joro, S., Sari, R. N., Ritiau, Y. A. P., Rizqita, E. A. and Nafidiastri, F. A., 2026. Lead (Pb) bioaccumulation and physiological responses of <i>Acanthus ilicifolius</i> in the mangrove forest, Wonorejo, Surabaya, East Java, Indonesia. <i>Nature Environment and Pollution Technology</i> , 25(4), D1908. <a href="https://doi.org/10.46488/NEPT.2026.v25i04">https://doi.org/10.46488/NEPT.2026.v25i04</a> . D1908

## ABSTRACT

Mangrove ecosystems in coastal areas are susceptible to heavy metal inputs from various anthropogenic activities. One of the heavy metals frequently found in coastal environments is lead (Pb), which can accumulate in sediments and mangrove vegetation. The Jeruju plant (*Acanthus ilicifolius*) is a type of mangrove vegetation capable of absorbing heavy metals from the environment, making it a potential bioindicator of pollution. This study aims to evaluate Pb levels in seawater and sediments, as well as Pb levels in the roots, stems, and leaves of *A. ilicifolius*; to evaluate its functions as a bioaccumulator and translocator; to determine the chlorophyll content and protein content of *A. ilicifolius*; and to analyze the relationship between Pb levels in *A. ilicifolius*, chlorophyll content, and protein content of *A. ilicifolius*. Sampling was conducted at three stations, with three replicates at each station. Lead (Pb) concentration analysis was performed using atomic absorption spectrophotometry (AAS), while protein content analysis was conducted using the Kjeldahl method and chlorophyll content using spectrophotometry. Data analysis was conducted using descriptive qualitative methods and Pearson's correlation analysis via SPSS. The results showed that the highest Pb concentrations were found in sediments at 1.135–1.440 mg/kg. In plant tissues, the highest Pb concentrations were detected in roots, ranging from 0.294 to 0.380 mg/kg, and were lower in stems and leaves. Leaf chlorophyll content ranged from 1.133–1.487 mg/g, while protein content ranged from 21.866–26.466 mg/kg. Correlation analysis results indicate a positive relationship between Pb levels and plant physiological parameters. This suggests that *A. ilicifolius* is still capable of tolerating the presence of Pb without experiencing

significant physiological disturbances, making it a potential bioindicator for heavy metal contamination in mangrove ecosystems.

## 1. INTRODUCTION

The Wonorejo Mangrove Forest Area in Surabaya is one of the mangrove ecosystems in an urban coastal zone with high levels of anthropogenic activity. Domestic and industrial activities, as well as river flows that discharge into the area, can introduce various pollutants into the mangrove environment, including heavy metals. Heavy metals such as lead (Pb) are among the contaminants frequently found in coastal ecosystems due to their persistence and tendency to accumulate in sediments and living organisms (Rahman et al. 2021, Chowdhury, Naz, & Maiti, 2021). Heavy metal pollution in coastal areas generally originates from industrial activities, domestic waste, and river flows that carry pollutants from the mainland to estuarine and mangrove areas (Mariwy, Dulanlebit & Ode, 2024).

In mangrove ecosystems, sediments serve as the primary medium for heavy metal accumulation because metal particles tend to bind to organic matter and fine particles that settle to the bottom of the water. Heavy metals accumulated in sediments can be absorbed by mangrove roots and subsequently distributed throughout various plant tissues. This process has led to mangrove plants being frequently used as bioindicators and bioaccumulators to assess the presence of heavy metals in coastal environments. Research by Permatasari, Aditya, and Mindari (2024) reports that several mangrove species can absorb and accumulate heavy metals, such as Pb, Cd, and Zn, from sediments and surrounding waters. This ability makes mangrove plants potentially useful as bioindicators and bioaccumulators in studies of heavy metal pollution in coastal ecosystems.

One of the plant species commonly found in mangrove ecosystems is the Jeruju (*Acanthus ilicifolius*). This plant has a well developed root system that interacts directly with sediment, potentially allowing it to absorb various heavy metals from its surrounding environment. Several studies indicate that *A. ilicifolius* can accumulate various heavy metals, including Pb, Cu, and Zn, in plant tissues, with differing distribution patterns among plant organs. Generally, metal concentrations are higher in roots than in stems and leaves because roots are the first parts to interact directly with contaminated sediments (Siregar et al. 2025). Additionally, mangrove plants' ability to accumulate metals can affect their physiological condition, including changes in chlorophyll

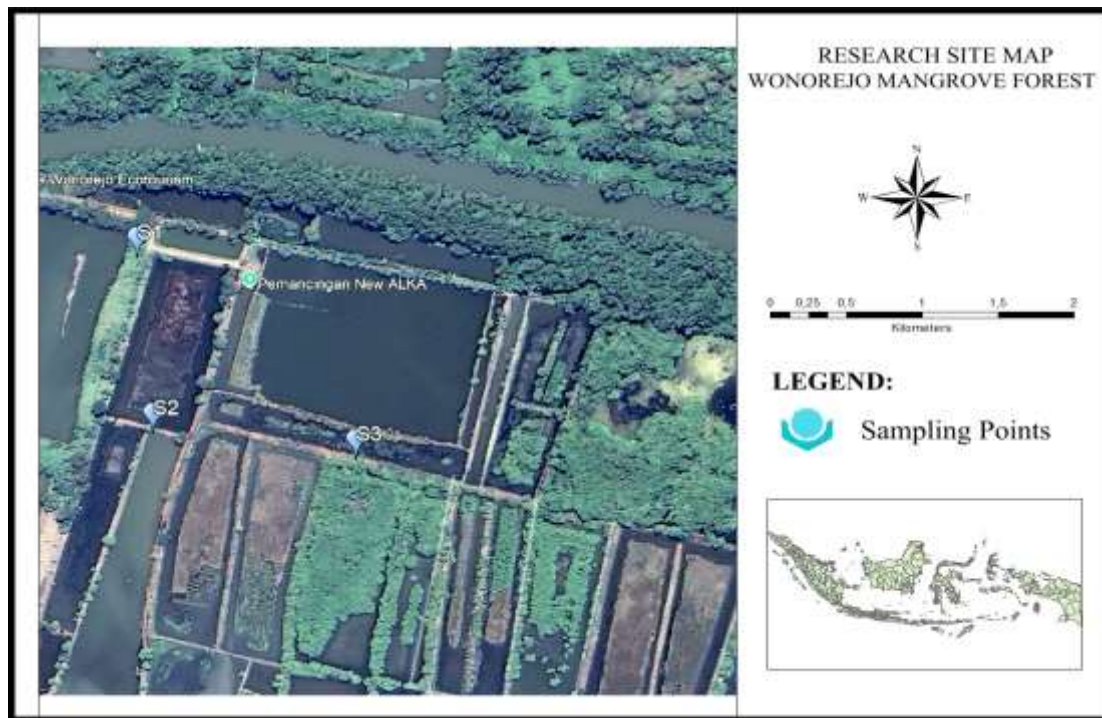
and protein content in response to heavy metal stress (Rahman et al. 2021). Furthermore, the ability of mangroves to accumulate heavy metals demonstrates their potential as bioindicators and phytoremediation agents in polluted coastal ecosystems (Rahman et al., 2021; Siregar et al., 2025).

Although numerous studies have documented mangrove plants' ability to absorb heavy metals, information on the bioaccumulation of lead (Pb) in *A. ilicifolius* and its impact on plant physiological responses in urban mangrove ecosystems remains relatively limited. Therefore, a study on Pb bioaccumulation in *A. ilicifolius* and the physiological responses of its leaves, such as chlorophyll and protein content, is needed to understand the effects of heavy metal exposure on the physiological condition of mangrove plants. Against this background, the objective of this study is to evaluate Pb concentrations in water and sediment, as well as Pb concentrations in the roots, stems, and leaves of Jeruju (*Acanthus ilicifolius*); to evaluate its function as a bioaccumulator and translocator; to determine the chlorophyll and protein content in the leaves of Jeruju (*Acanthus ilicifolius*); and to analyze the relationship between Pb levels in Jeruju (*Acanthus ilicifolius*), chlorophyll content, and protein content.

## 2. MATERIALS AND METHODS

### a. Research Location

This study was conducted in the Wonorejo Mangrove Forest, Surabaya. Observations were made at three stations with three replicates (Fig. 1). Research samples included water, sediment, and Jeruju (*Acanthus ilicifolius*), as well as measurements of physicochemical parameters in the water. Analysis of heavy metal (Pb) levels and protein content was conducted at the Nutrition Laboratory of Airlangga University.



**Fig 1.** Sampling site

### **b. Sample Collection**

Water samples for measuring physicochemical parameters were collected in the Wonorejo Mangrove Forest habitat in Surabaya. The study included three observation stations with three replicates. The instruments used in this study included a Gea S-006 stem thermometer for temperature, a Milwaukee pH meter for water pH, an Atago S-28 refractometer for salinity, and a Milwaukee MW-600 DO meter for dissolved oxygen. Materials required for this study included roots, stems, and leaves of the Jeruju tree, seawater, marine sediment, clean water, distilled water,  $\text{Na}_2\text{SO}_4\text{-HgO}$  (catalyst), 96% ethanol, 65%  $\text{HNO}_3$ ,  $\text{H}_2\text{O}_2$ , 0.2 N HCl, 1% NaOH, 4%  $\text{H}_2\text{BO}_4$ , 98%  $\text{H}_2\text{SO}_4$ , and a 1000 ppm Pb solution.

Water and sediment samples were collected for lead (Pb) heavy-metal testing at three observation stations, with three replicates per station. Water samples were collected in volumes of  $\pm 1$  L using polyethylene (PE) sample bottles that had been rinsed with distilled water and the sample water. Each water sample was preserved by adding  $\text{HNO}_3$  until the pH was  $< 2$  to prevent changes in metal content. Sediment samples were placed in PE plastic bags.

Sampling of *A. ilicifolius* plants for testing lead (Pb) levels, protein content, and chlorophyll levels was conducted at three observation stations, with three replicates per station. *A. ilicifolius* samples were collected

from plants of relatively similar height and size. A total of 150 g of *A. ilicifolius* from each of the root, stem, and leaf sections in healthy condition was then cleaned and placed into PE plastic bags. All water, sediment, and Jeruju plant samples were labeled and named, then stored in a cool box at  $\pm 4^{\circ}\text{C}$ . The samples were then transported to the laboratory for analysis.

### c. Analysis of Pb Metal Content

Heavy metal analysis of plant samples (roots, stems, and leaves) and sediments was performed using the dry ashing method. Solid samples weighing 3.5 grams were placed in porcelain crucibles and heated in a furnace at  $800^{\circ}\text{C}$  for 3 hours, until a grayish-white ash formed. After cooling to room temperature, the ash was dissolved in 1 mL of concentrated nitric acid ( $\text{HNO}_3$ , 65%) and 10 mL of deionized water, then homogenized and filtered through a Whatman No. 42 filter paper. The filtrate was transferred to a volumetric flask and diluted with deionized water to the final volume. Heavy metal analysis of water samples was performed non destructively. Water samples were preserved by adding concentrated  $\text{HNO}_3$  until the pH was  $< 2$  immediately after field sampling to prevent changes in metal concentrations, then filtered using Whatman No. 42 filter paper prior to analysis. All samples were analyzed using Shimadzu AA-7000 Atomic Absorption Spectrophotometry (AAS) at a wavelength of 283.3 nm for Pb. Calibration curves were prepared using Pb standard solutions prepared from 1000 ppm stock solutions. Each series of analyses was performed in duplicate with a Relative Percent Difference (RPD)  $\leq 10\%$  and included reagent blank testing to correct for potential contamination from the chemicals used.

### d. Analysis of Bioconcentration Factor and Translocation Factor

The bioaccumulation of the heavy metal Pb in *A. ilicifolius* plants was analyzed using the Bioconcentration Factor (BCF) and the Translocation Factor (TF) (Baker 1981, Takarina and Pin 2017). The BCF value was calculated using the formula:

$$\text{BCF} = \frac{C_{\text{Leaf}}}{C_{\text{Sediment}}} \quad \dots(1)$$

Where (C leaf) is the Pb concentration in the leaf tissue (mg/kg) and (C sediment) is the Pb concentration in the sediment (mg/kg). An interpretation of a value  $< 1$  indicates that the plant is an accumulator, a BCF of 1 indicates = accumulation, and a BCF  $> 1$  indicates that the plant is an excluder. The TF value is calculated using the formula:

$$TF = \frac{C_{Leaf}}{C_{Root}} \quad \dots(2)$$

Where (C leaf) is the Pb concentration in the leaf tissue (mg/kg) and (C root) is the Pb concentration in the root tissue (mg/kg). An interpretation of a TF value < 1 indicates that more heavy metals are retained in the roots, while a TF value > 1 indicates that heavy metals are translocated to the upper parts of the plant.

#### e. Analysis of Chlorophyll Content

The analysis was conducted using spectrophotometry. A 1 gram sample of healthy, thoroughly washed *A. ilicifolius* leaves was ground with 100 ml of 96% ethanol until fine, then filtered to obtain a liquid extract. The absorbance of the extract was measured using a V1100D UV-Vis spectrophotometer at 665 nm and 649 nm. Chlorophyll content was calculated using the formula (Wintermans and De Mots 1965; Li et al. 2022):

$$\text{Chlorophyll a} = 25.8 \times A_{665} - 7.6 \times A_{649}$$

$$\text{Chlorophyll b} = 25.8 \times A_{649} - 7.7 \times A_{665}$$

$$\text{Chlorophyll total} = 20 \times A_{649} + 6.1 \times A_{665}$$

#### f. Analysis of Protein Content

The analysis was performed using the Kjeldahl method by grinding 0.2 g of the dried sample, placing it in a Kjeldahl flask, and adding a Na<sub>2</sub>SO<sub>4</sub>-HgO catalyst mixture in a 2:1 ratio and 2 mL of H<sub>2</sub>SO<sub>4</sub>. The sample was digested for approximately 2.5 hours until the solution became clear. After cooling, 15 mL of distilled water was added, followed by distillation with the addition of 10 mL of NaOH. The distillate was collected in an Erlenmeyer flask. The distillate collected in the H<sub>3</sub>BO<sub>4</sub> solution, which contains an indicator, turns green. Next, the solution was titrated with 0.2 N HCl until the color changed from green to pink, indicating the endpoint. During the analysis, a blank digestion was also performed using the same procedure without adding a sample to correct for possible nitrogen contamination from the reagents. The total nitrogen content obtained was then converted to protein content using a conversion factor of 6.25, yielding the sample's protein content value. The nitrogen content was calculated using the following formula:

$$\% N = \frac{mL \text{ HCl} \times N \text{ HCl} \times 14.008}{\text{Biomass (mg)}} \times 100\%$$

The protein content was then determined by multiplying the nitrogen content by a conversion factor:

$$\% \text{ Protein} = \% \text{ N} \times 6.25$$

### 3. RESULTS AND DISCUSSIONS

Research conducted in the Wonorejo Mangrove Forest in Surabaya yielded the following data:

#### 1. Lead (Pb) Heavy Metal Content in Jeruju (*Acanthus ilicifolius*)

Based on the analysis of lead (Pb) concentrations in water at all research stations, the levels remain below the quality standard set by Government Regulation of the Republic of Indonesia No. 22 of 2021, which is 0.03 mg/L. Pb concentrations in the water range from 0.022 to 0.029 mg/L. In sediments, Pb concentrations range from 1.135 to 1.440 mg/kg, which is significantly below the Threshold Effect Level (TEL) of 35 mg/kg set by the Canadian Council of Ministers of the Environment (CCME) in 2002. Lead concentrations in *A. ilicifolius* plants showed variation across plant parts, with the highest values in the roots (0.294–0.380 mg/kg), followed by the stems (0.271–0.307 mg/kg), and the lowest in the leaves (0.109–0.198 mg/kg). These values remain below the heavy metal reference range of < 30 mg/kg, as defined by Alina Kabata-Pendias (Table 1), indicating that the *A. ilicifolius* plants at the study site are still in normal condition and have the potential to serve as bioindicators of heavy metal presence in mangrove ecosystems.

**Table 1.** Lead (Pb) Heavy Metal Content in Water, Sediments, and Jeruju Plants (*Acanthus ilicifolius*).

Parameters	Station I	Station II	Station III	Quality Standards
Water (mg/L)	0.029 ± 0.001	0.026 ± 0.001	0.022 ± 0.001	0.03 mg/L*
Sediment (mg/kg)	1.440 ± 0.005	1.381 ± 0.007	1.135 ± 0.006	35 mg/kg**
Roots (mg/kg)	0.364 ± 0.004	0.380 ± 0.005	0.294 ± 0.008	
Stem (mg/kg)	0.307 ± 0.008	0.299 ± 0.009	0.271 ± 0.007	< 30 mg/kg***
Leaves (mg/kg)	0.198 ± 0.005	0.133 ± 0.007	0.109 ± 0.005	

**Note:** The data shown is the average of the replication ± standard deviation.

\*Government Regulation (PP) Number 22 of 2021.

\*\*Sediment quality guideline from Canadian Council of Ministers of the Environment (CCME) of 2002.

\*\*\*Reference value for heavy metals in plants from Kabata Pendias of 2011.

Heavy metal lead (Pb) concentrations in water that remain below quality standards are influenced by anthropogenic activities such as domestic wastewater discharge, industrial activities, and transportation, which

carry various pollutants into estuarine areas (Zhang et al. 2020). Additionally, tidal dynamics and sedimentation processes also play a role in distributing heavy metals in coastal waters, causing metal concentrations in the water column to fluctuate between study sites (Wang et al. 2021). Lead (Pb) concentrations in sediments remain well below the Threshold Effect Level (TEL). Sediments serve as the primary medium for heavy metal accumulation in coastal ecosystems because metal particles tend to bind with organic matter and fine minerals that settle on the seabed (Zhang et al. 2020, Kumar et al. 2022). Therefore, heavy metal concentrations in sediments are generally higher than in water because sediments act as long-term storage sites for various metal contaminants (Liu et al. 2021).

The distribution of Pb in *A. ilicifolius* shows variations in concentration across different plant organs. The highest concentration was found in the roots, followed by the stems, with the lowest concentration in the leaves. This distribution pattern indicates that the roots are the primary part of the plant involved in the uptake of heavy metals from sediments and surrounding water bodies (Zhou et al. 2021). Mangrove plants possess a complex root system, enabling them to absorb and accumulate various heavy metals from the sedimentary environment through adsorption and ion transport processes (Rahman et al. 2022). Higher metal accumulation in roots than in other plant organs is also frequently observed in various mangrove species as an adaptive mechanism against environmental stress caused by metal contamination (Chen et al. 2023). Higher Pb accumulation in roots compared to stems and leaves indicates the presence of physiological mechanisms in plants that limit the movement of heavy metals toward photosynthetic tissues. Mangrove plants are known to retain heavy metals in root tissues through binding to cell walls, precipitation within vacuoles, and the formation of complexes with organic compounds such as phytochelatins and metallothioneins (Sarker et al. 2021, Huang et al. 2023). These mechanisms help reduce potential damage to leaf tissues, thereby ensuring that photosynthesis and plant metabolism can continue to function optimally (Rahman et al. 2022). Therefore, the lower Pb concentration in the leaves of the mangrove in this study indicates a process of restricting heavy metal translocation from the roots to the upper parts of the plant.

## **2. Bioaccumulation of Lead (Pb) in Jeruju Plants (*Acanthus ilicifolius*)**

Based on the analysis results, different Bioconcentration Factors (BCF) and Translocation Factors (TF) were obtained at each station (Table 2). The BCF values ranged from 0.096 to 0.137, indicating values  $< 1$ , while the TF values ranged from 0.351 to 0.543, indicating values  $< 1$ .

**Table 2.** Bioconcentration Factor (BCF) and Translocation Factor (TF) Values in the Jeruju Plant (*Acanthus ilicifolius*).

Station	BCF	Interpretation	TF	Interpretation
1	$0.137 \pm 0.003$		$0.543 \pm 0.006$	
2	$0.097 \pm 0.004$	$< 1$	$0.351 \pm 0.015$	$< 1$
3	$0.096 \pm 0.004$		$0.370 \pm 0.025$	

**Note:** The data shown is the average of the replication  $\pm$  standard deviation.

The bioconcentration factor (BCF) and translocation factor (TF) values for the Jeruju plant (*Acanthus ilicifolius*) indicate the plant's ability to absorb and distribute lead (Pb) from aquatic environments and sediments. The BCF value patterns obtained indicate that *A. ilicifolius* can accumulate Pb from its surrounding environment, consistent with the results in (Table 1), where Pb concentrations in plant tissues are relatively higher in roots than in stems and leaves. These conditions indicate that the root system plays a crucial role in heavy metal uptake from the sediment substrate, which serves as the primary medium for metal accumulation in coastal ecosystems (Rahman et al. 2022). Sediments are known to bind heavy metals through interactions with organic matter and fine mineral particles, making them the primary source of metals that can be absorbed by mangrove plants (Kumar et al. 2022, Liu et al. 2021).

The TF values obtained indicate that some of the Pb absorbed by the roots can be translocated to the stem and leaves, although the concentration is lower than that in the roots. This pattern is consistent with the distribution of Pb in plant tissues shown in (Table 1), where metal accumulation is more dominant in the root organs. This condition indicates that plants possess a physiological mechanism that limits the movement of heavy metals toward photosynthetic tissues, as a form of adaptation to environments containing metals. Mangrove plants are known to retain heavy metals in root tissues through binding to cell walls, storage in vacuoles, and the formation of complexes with certain organic compounds, thereby controlling the translocation of metals to upper plant organs (Sarker et al. 2021, Huang et al. 2023). These mechanisms allow plants to maintain their physiological functions even when in environments containing heavy metals.

### 3. Chlorophyll Content in Jeruju Leaves (*Acanthus ilicifolius*)

Based on the analysis results from the three research stations, chlorophyll values ranging from 1.133 to 1.487 mg/g were obtained. These values fall within the range of chlorophyll concentrations reported in the study by Y. Li et al. (2018), which ranged from 0.87 to 15.92 mg/g.

**Table 3.** Chlorophyll Content in Jeruju Leaves (*Acanthus ilicifolius*).

Station	Chlorophyll (mg/g)	Quality Standards (mg/g)*
1	1.487 ± 0.128	
2	1.483 ± 0.126	0.87-15.92
3	1.133 ± 0.127	

**Note:** The data shown is the average of the replication ± standard deviation.

\*Reference value for chlorophyll content from Li et al. 2018.

Chlorophyll levels in the leaves of the *A. ilicifolius* at the three research stations showed a relatively uniform range of values. This condition indicates that the plants' photosynthetic activity is still proceeding normally despite the presence of Pb in the aquatic environment and sediments, as shown in (Table 1) Chlorophyll is the primary pigment involved in photosynthesis; therefore, its presence is significantly influenced by environmental conditions and the availability of nutrients that support plant metabolism (Li et al. 2022). Under conditions of high heavy metal exposure, chlorophyll content in leaves generally decreases due to damage to the chloroplast structure and disruption of photosynthetic pigment biosynthesis (Sharma et al. 2021). However, the relatively stable chlorophyll values in this study indicate that Pb concentrations at the study site remain at levels tolerable to the mangrove plants.

This relates to mangrove plants ability to regulate the distribution of heavy metals within their tissues. As shown in (Table 1) and (Table 2), the majority of Pb tends to accumulate in the roots, so only a small portion is translocated to the leaves. This mechanism helps protect photosynthetic tissues from the toxic effects of heavy metals, ensuring the photosynthetic process continues optimally (Chen et al. 2023). Stable photosynthetic activity supports plant metabolic processes, including the formation of organic compounds such as proteins, which play a crucial role in plant growth and adaptation to environmental stress (Huang et al. 2023). Thus, the chlorophyll levels in the leaves of the mangrove indicate that the plants are still capable of maintaining their physiological functions despite exposure to lead (Pb).

#### 4. Protein Content of Jeruju Leaves (*Achantus ilicifolius*)

Based on the analysis results from the three observation stations, protein values ranged from 21.866 to 26.466 mg/kg, with the highest at Station 1 and the lowest at Station 3 (Table 4). These values are higher than the reference values reported by Murdinah (2009), which range from 2.8 mg/kg.

**Table 4.** Protein Content in Jeruju Leaves (*Acanthus ilicifolius*).

Station	Protein Content (mg/kg <sup>-1</sup> )	Quality Standards (mg/kg <sup>-1</sup> )*
1	26.466 ± 404.15	
2	24.766 ± 305.51	2.8
3	21.866 ± 208.17	

**Note:** The data shown is the average of the replication ± standard deviation

\* Reference value for protein content from Murdinah 2009.

Stations with higher leaf Pb concentrations also had relatively higher protein levels. This indicates that although the detected Pb concentrations were still in the low range, the presence of this heavy metal could still trigger physiological responses in plants, including changes in protein metabolism as part of an adaptive mechanism to environmental stress. At relatively low to moderate levels of heavy metal exposure, plants can maintain their metabolic functions by increasing the synthesis of specific proteins involved in heavy metal detoxification. These proteins can function as heavy metal chelators, thereby reducing the potential for damage to plant cells. This mechanism is part of the plant's physiological defense system, enabling plants to survive in environments containing heavy metals. Several studies report that plants can produce metal-binding proteins, such as metallothionein and phytochelatin, which bind and neutralize heavy metals entering plant tissues (Al Safran et al. 2022, Rahman et al. 2021).

In addition to serving as a response to heavy metal exposure, leaf protein content is also influenced by the plant's physiological condition and the availability of nutrients in its growing environment. Plants growing in environments with relatively good water quality generally maintain photosynthetic activity and nitrogen metabolism, which support protein synthesis. This aligns with research findings indicating that environmental quality at the study site remains within acceptable standards, allowing the mangrove plants to maintain normal metabolic functions. Thus, the relatively high protein content in *A. ilicifolius* leaves indicates that this plant not

only has good nutritional value but also can adapt to the presence of heavy metals in the mangrove ecosystem (Chowdhury et al. 2021, Zhao et al. 2022).

### 5. Analysis of Heavy Metal (Pb) and Protein Content in Jeruju (*Acanthus ilicifolius*)

Based on the analysis using Statistical Product and Service Solutions (SPSS) 22.0 for Windows, correlation coefficients ranged from 0.574 to 0.919. The highest correlation value was found in the relationship between protein and stem Pb ( $r = 0.919$ ), while the lowest correlation value was found in the relationship between leaf Pb and root Pb ( $r = 0.574$ ) (Table 5). These values indicate that most parameters show a positive correlation, with varying degrees of strength, suggesting a relationship between the distribution of Pb in plant tissues and the physiological condition of *A. ilicifolius*.

**Table 5.** Correlation Analysis of Heavy Metal (Pb) Levels in Jeruju (*Acanthus ilicifolius*) with Protein and Chlorophyll Content.

	Pb Roots	Pb Stems	Pb Leaf	Leaf Protein	Chlorophyll
Pb Roots	1	0.822	0.574	0.842	0.896
Pb Stems	0.822	1	0.772	0.919	0.870
Pb Leaf	0.574	0.772	1	0.912	0.646
Leaf Protein	0.842	0.919	0.912	1	0.820
Chlorophyll	0.896	0.870	0.646	0.820	1

Correlation analysis revealed a relationship between Pb content in the tissues of Jeruju (*Acanthus ilicifolius*) plants and physiological parameters, namely chlorophyll and protein. A fairly strong positive correlation was also observed between Pb in the roots, stems, and leaves, indicating that Pb absorbed by the root system can be distributed to other plant tissues through translocation. This pattern aligns with research showing that mangrove plants can absorb and accumulate heavy metals from sediments via their roots, after

which some of the metals can be translocated to the plant's stems and leaves (Rahman et al. 2021). This ability indicates that the mangrove plays a crucial role in the bioaccumulation of metals within the mangrove ecosystem and holds potential as a phytoremediator in water bodies contaminated with heavy metals.

The positive correlation between Pb content and chlorophyll and protein levels indicates that the presence of Pb at the measured concentrations at the study site has not yet caused significant toxic effects on the plants' physiological activities. Several studies suggest that mangrove plants possess tolerance mechanisms against heavy metals, including metal binding in specific tissues and metabolic activities that aid in metal detoxification within plant cells. This allows plants to maintain physiological functions, such as photosynthesis and protein synthesis, even in environments containing heavy metals (Siregar et al. 2025). This tolerance makes *A. ilicifolius* a potential bioindicator and bioaccumulator of heavy metals in coastal and mangrove ecosystems.

## 6. Physical and Chemical Quality of Mangrove Forests

Based on the physical and chemical water quality analysis (Table 6), the measurement results from the three stations still meet the marine water quality standards set forth in Government Regulation No. 22 of 2021 on the Implementation of Environmental Protection and Management, falling under Class III. pH values ranged from 6.7 to 7, which aligns with the pH quality standard of 6 to 9 for all water classes. Water temperature ranged from 29.3 to 33°C, remaining within the normal range for tropical coastal ecosystems. Salinity ranges from 1.67 to 5.67 ppt, reflecting the characteristics of a mangrove transitional zone influenced by tides and freshwater inflow from rivers. Dissolved oxygen (DO) levels range from 6.4 to 8.1 mg/L, well above the minimum quality standard of 3 mg/L.

**Table 6.** Physical and Chemical Water Quality Data.

Parameters	Station I	Station II	Station III	Quality Standards
pH	6.7 ± 0.58	7 ± 1.00	7 ± 0.05	6-9
Temperature (°C)	29.3 ± 0.31	29.2 ± 0.10	33 ± 2.24	deviation 3 (with air temperature)
Salinity (ppt)	4.33 ± 0.57	5.67 ± 0.57	1.67 ± 0.57	-
DO (mg/L)	7.5 ± 0.02	6.4 ± 0.03	8.1 ± 0.03	≥ 3 mg/L

**Note:** The data shown is the average of the replication ± standard deviation

Water quality parameters indicate that the water conditions still support the life of coastal organisms, including mangrove vegetation such as Jeruju (*Acanthus ilicifolius*). Relatively stable pH values indicate that the water is neither too acidic nor too alkaline, thus remaining within a range suitable for biological processes in the mangrove ecosystem. Temperature and salinity parameters also play a crucial role in determining water conditions, as both influence organismal metabolic activity and various chemical processes occurring in the water (Ali et al. 2021, Kumar et al. 2022).

The dissolved oxygen (DO) levels measured at the study site indicate that DO remains sufficient to support the respiratory activity of aquatic organisms. Dissolved oxygen is a key indicator of water quality as it is directly linked to ecosystem balance and microbial activity in the sediment. Good DO conditions indicate that the decomposition of organic matter and water circulation in the area are still proceeding effectively (Wang et al. 2021). These relatively stable water quality conditions are also consistent with the results in (Table 1), which show that Pb concentrations in seawater and sediments remain within a relatively low range.

These favorable water-quality conditions also help maintain the physiological functions of the mangrove plants observed in this study. As shown in previous results, Pb tends to accumulate at higher levels in roots than in stems and leaves, suggesting a mechanism controlling metal distribution within plant tissues. This mechanism allows the photosynthetic tissues to continue functioning properly, as reflected in the relatively stable chlorophyll and leaf protein levels. Mangrove plants are known to possess various physiological adaptation mechanisms to maintain metabolic activity even in environments containing certain levels of heavy metals (Chen et al. 2023, Huang et al. 2023).

## 5. CONCLUSIONS

This study shows that Pb concentrations in water, sediments, and the tissues of the Jeruju plant (*Acanthus ilicifolius*), including roots, stems, and leaves, at the three study sites exhibit distribution patterns across both environmental media and plant tissues. The highest Pb concentrations were found in sediments at 1.135–1.440 mg/kg, followed by root tissues at 0.294–0.380 mg/kg, while concentrations in stems and leaves were relatively lower. This pattern indicates that sediment serves as the primary accumulation site for Pb in the aquatic environment, while the roots of the *A. ilicifolius* plant play a crucial role in the uptake of metals from sediment, which are then translocated to other parts of the plant. Nevertheless, Pb levels in water, sediment, and plant

tissues remain within tolerable ranges and do not indicate contamination exceeding safe limits. This is supported by the chlorophyll content of *A. ilicifolius* leaves, ranging from 1.133–1.487 mg/g, and protein levels of 21.866–26.466 mg/kg, which still indicate a good physiological condition of the plants. Correlation analysis results also indicate a positive relationship between Pb content in plant tissues and plant physiological parameters, with strong correlations found between protein and stem Pb ( $r = 0.919$ ) and protein and leaf Pb ( $r = 0.912$ ), as well as between chlorophyll and root Pb ( $r = 0.896$ ) and chlorophyll and stem Pb ( $r = 0.870$ ). This indicates that Pb accumulation in *A. ilicifolius* plants remains tolerable, without causing significant negative impacts on their physiological condition. Therefore, the Jeruju plant (*Acanthus ilicifolius*) shows potential as a bioindicator of lead (Pb) in coastal aquatic ecosystems.

## 6. ACKNOWLEDGMENTS

The authors are grateful to the people around the river for their assistance during sampling session. Didik for his great help at the molecular laboratory. This research has received funding from LPPM with grant number 309/UN38/HK/PP/2024.

## REFERENCES

- Ali, H., Khan, E. and Ilahi, I., 2021. Environmental Chemistry and Ecotoxicology of Hazardous Heavy Metals. *Journal of Chemistry*, pp.1–14. [\[DOI\]](#)
- Alsafran, M., Usman, K., Ahmed, B., Rizwan, M., Saleem, M. H., and Al Jabri, H., 2022. Understanding the Phytoremediation Mechanisms of Potentially Toxic Elements: A Proteomic Overview of Recent Advances. *Frontiers in Plant Science*, 13, 881242. [\[DOI\]](#)
- Baker, A.J.M., 1981. Accumulators and Excluders Strategies in the Response of Plants to Heavy Metals. *Journal of Plant Nutrition*, 3(1–4), pp. 643–654. [\[DOI\]](#)
- Canadian Council of Ministers of the Environment (CCME), 2002. *Canadian Sediment Quality Guidelines for the Protection of Aquatic Life: Summary Tables*. Winnipeg: Canadian Council of Ministers of the Environment. Available at:

[https://support.esdat.net/Environmental%20Standards/canada/ccme/sedqg\\_summary\\_table.pdf](https://support.esdat.net/Environmental%20Standards/canada/ccme/sedqg_summary_table.pdf) (Accessed: 10 August 2025).

- Chen, Y., Wang, C., Liu, H. and Zhang, J., 2023. Heavy Metal Accumulation and Detoxification Strategies in Mangrove Plants: a Review. *Marine Pollution Bulletin*, 188, 114672. [\[DOI\]](#)
- Chowdhury, A., Naz, A., & Maiti, S. K., 2021. Bioaccumulation of Potentially Toxic Elements in Three Mangrove Species and Human Health Risk Due to Their Ethnobotanical Uses. *Environmental Science and Pollution Research*, 28, 1–18. [\[DOI\]](#)
- Huang, L., Li, J., Wang, Y. and Chen, G., 2023. Physiological Responses and Detoxification Mechanisms of Mangrove Plants Exposed to Heavy Metal Stress. *Environmental Research*, 216, 114708.
- Wintermans, J.G.F.M. and De Mots, A., 1965. Spectrophotometric Characteristics of Chlorophylls A and B and Their Pheophytins in Ethanol. *Biochimica et Biophysica Acta*, 109(2), pp. 448–453. [\[DOI\]](#)
- Kabata Pendias, A., 2011. *Trace Elements in Soils and Plants*. 4th edn. Boca Raton: CRC Press. [\[DOI\]](#)
- Kjeldahl, J., 1883. Neue Methode zur Bestimmung des Stickstoffs in Organischen Körpern, *Zeitschrift für Analytische Chemie*, 22, pp. 366–382.
- Kumar, V., Sharma, A. and Pandita, S., 2022. Heavy Metals in Sediments of Coastal Ecosystems: Sources, Distribution and Ecological Risks. *Environmental Monitoring and Assessment*, 194, 354. [\[DOI\]](#)
- Li, J., Hu, W., Lu, Z., Meng, F., Cong, R., Li, X., Ren, T. and Lu, J., 2022. Imbalance Between Nitrogen And Potassium Fertilization Influences Potassium Deficiency Symptoms In Winter Oilseed Rape (*Brassica napus* L.) Leaves. *The Crop Journal*, 10(2), pp. 565–576. [\[DOI\]](#)
- Liu, X., Song, Q., Tang, Y. and Li, W., 2021. Distribution Characteristics and Ecological Risk Assessment of Heavy Metals In Sediments. *Science of the Total Environment*, 755, 142649. [\[DOI\]](#)
- Li, Y., He, N., Hou, J., Xu, L., Liu, C., Zhang, J., Wang, Q. and Wu, X., 2018. Factors Influencing Leaf Chlorophyll Content In Natural Forests at the Biome Scale. *Frontiers in Ecology and Evolution*, 6, 64. [\[DOI\]](#)
- Mariwy, A., Dulanlebit, Y.H. & Ode, A., 2024. Bioaccumulation of Lead Metal (Pb) by Mangrove Plants In Ambon Coastal Waters. *Indonesian Journal of Chemical Research*, 12(1), pp.71–78.
- Murdinah., 2009. Effect of Extracting and Aging Agents on the Quality of Carrageenan from Seaweed *Eucheuma cottonii*. Indonesia: UGM Fisheries and Marine Affairs with the Research Center for Marine and Fisheries Product Processing and Biotechnology.

- Peraturan Pemerintah Republik Indonesia (PP). 2021. Peraturan Pemerintah Nomor 22 Tahun 2021 tentang Penyelenggaraan Perlindungan dan Pengelolaan Lingkungan Hidup (Baku Mutu Air Laut – Lampiran VIII). Available online: <https://peraturan.bpk.go.id/Details/161852/pp-no-22-tahun-2021> (accessed on 10 August 2025).
- Permatasari, F.D., Aditya, H.F. & Mindari, W., 2024. Phytoremediation Potential of Mangrove Species In Lead Contaminated Coastal Areas. *Jurnal Ilmiah Pertanian*, 22(1).
- Rachmadiarti, F., Fitrihidajati, H., Purnomo, T., Kuntjoro, S., Nafidiasri, F.A., Yolanda, R., Ambarwati, R., Anggorowati, D., Budijastuti, W., Faizah, U. and Putriarti, D., 2024. Bioaccumulation of Lead (Pb) and Cadmium (Cd) in *Padina australis* Hauck at Palang Beach, Tuban, East Java, Indonesia. *Nature Environment & Pollution Technology*, 23(3), pp. 1507–1516. [\[DOI\]](#)
- Rahman, M., Hossain, M., Islam, M. and Rahman, M., 2022. Heavy Metal Accumulation and Translocation in Mangrove Plants Growing in Coastal Ecosystems. *Environmental Science and Pollution Research*, 29, pp.39245–39257. [\[DOI\]](#)
- Rahman, M. S., Saha, N., Ahmed, A. S. S., Babu, S. M. O. F., Islam, A. R. M. T., Begum, B. A., Jolly, Y. N., Akhter, S., & Choudhury, T. R., 2021. Depth Related Dynamics of Physicochemical Characteristics and Heavy Metal Accumulation in Mangrove Sediment and Plant: *Acanthus ilicifolius* as a Potential Phytoextractor. *Marine Pollution Bulletin*, 173, 113160. [\[DOI\]](#)
- Sarker, U., Oba, S. and Daramy, M., 2021. Mechanisms of Heavy Metal Tolerance in Plants: Physiological and Molecular Perspectives. *Plants*, 10(12), 2590.
- Sharma, A., Nagpal, A. and Singh, J., 2021. Impact of Heavy Metals on Photosynthesis and Chlorophyll Metabolism in Plants. *Environmental Science and Pollution Research*, 28, pp.239–252. [\[DOI\]](#)
- Siregar, S.R., Irwan, S.N.R. and Putra, E.T.S., 2020. Kandungan Logam Berat Timbal (Pb) dan Pengaruhnya pada Angsana (*Pterocarpus indicus*), Tanjung (*Mimusops elengi*), dan Asam Jawa (*Tamarindus indica*) di Jalan Lingkar Alun–Alun Yogyakarta. *Vegetalika*, 9(1), pp. 316–329. [\[DOI\]](#)
- Siregar, E. S., Jumilawaty, E., Tanjung, M., Syafitri, A., Kusmana, C., Basyuni, M., Rahmania, R., 2025. Bioaccumulation of Heavy Metals by *Acanthus ilicifolius* in Polluted Mangrove Ecosystems. *Emerging Science Journal*, 9(2), 557–568. [\[DOI\]](#)
- Takarina, N.D. and Pin, T.G., 2017. Bioconcentration Factor (BCF) and Translocation Factor (TF) of Heavy Metals in Mangrove Trees of Blanakan Fish Farm. *Makara Journal of Science*, 21(2), pp. 79–83. [\[DOI\]](#)

- Wang, J., Liu, G., Liu, H. and Lam, P., 2021. Multivariate Statistical Evaluation of Heavy Metal Contamination in Coastal Environments. *Marine Pollution Bulletin*, 164, 112036. [\[DOI\]](#)
- Zhang, C., Nie, S., Liang, J., Zeng, G., Wu, H., Hua, S., Liu, J., and Yuan, Y., 2020. Levels, Sources and Ecological Risks of Heavy Metals in Sediments From A Typical Urban River in China Using PCA and PMF. *Ecological Indicators*, 117, 106630. [\[DOI\]](#)
- Zhao, P., Sanganyado, E., Wang, T., Sun, Z., Jiang, Z., Zeng, M., Huang, Z., Li, Y., Li, P., Bi, R., & Liu, W., 2022. Accumulation of Nutrients and Potentially Toxic Elements in Plants and Fishes in Restored Mangrove Ecosystems in South China. *Science of the Total Environment*, 838, 156471. [\[DOI\]](#)