

Ecophysiological responses of the halophyte *Lumnitzera racemosa* Willd. to heavy metal stress in an ultramafic mining environment

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ABSTRACT

Plants are essential components of life, providing food, habitat, and ecological balance. Their leaves harness solar energy, making them critical indicators of environmental health. Leaf size indices provide valuable insights into plant health, transpiration, growth, temperature regulation, and light capture. This study investigated the effects of six heavy metals—Molybdenum (Mo), Manganese (Mn), Lead (Pb), Chromium (Cr), Nickel (Ni), and Cadmium (Cd)—on the morphometric leaf traits of the halophyte *Lumnitzera racemosa* Willd. in an ultramafic mining site in Claver, Surigao del Norte, with a non-mining control site in Nasipit, Agusan del Norte. Sediment analyses revealed that Cr (4,434 ppm) and Ni (4,234 ppm) in the mining site exceeded WHO and FAO permissible limits (100 and 50 ppm, respectively), while Ni (552 ppm) and Cr (334 ppm) in the control site also surpassed thresholds, indicating diffuse contamination. Morphometric analysis showed significant differences ($p < 0.05$) in all measured leaf traits between sites. *L. racemosa* in Nasipit exhibited longer leaves (mean = 5.17 cm) and greater leaf area (mean = 7.53 cm²) compared with Claver (4.17 cm and 6.83 cm², respectively), representing increases of 22% in length and 9% in area. In contrast, leaves from Claver had broader widths (by 12%) and longer petioles (by 15%), possibly reflecting compensatory morphological plasticity under multi-metal stress. The widespread contamination of all six heavy metals likely induced synergistic or antagonistic toxic effects, altering cell division and photosynthetic activity. Leaf macronutrient analysis revealed nitrogen deficiency in both populations, optimum to high phosphorus, and excessive potassium levels, suggesting that leaf size variations were not driven by nutrient imbalance but by metal toxicity and environmental stress. The study concludes that *L. racemosa* exhibits adaptive morphometric responses to heavy metal exposure, indicating its potential as a bioindicator species for monitoring contamination in ultramafic and mining-affected mangrove ecosystems.

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1. INTRODUCTION

Leaves have a remarkable impact on ecosystems and human life through their cooling effect, aesthetics, and roles in food production and shade provision. They capture solar energy and sustain life on earth by facilitating photosynthesis and transpiration, and in some species, they even contribute to reproduction (Rahman et al., 2020). The condition of a plant can often be assessed by examining its leaf area, which plays a crucial role in regulating water loss, maintaining water balance, supporting material cycling, and influencing overall productivity. Moreover, leaf size significantly affects stress resistance, yield performance, and light energy efficiency (Zhu et al., 2025). It also governs physiological processes such as photosynthesis, transpiration, and respiration (Pedraza et al., 2024).

While leaf size and shape have long been studied as key indicators of plant functional ecology, these traits are highly sensitive to environmental stressors such as pollution

and soil toxicity. In recent years, attention has turned to the influence of heavy metals on plant morphology and physiology. Studies by Hu et al. (2023), El-Khatib et al. (2020), and Saravan and Sarkar (2022) demonstrated that excessive concentrations of metals such as Cd, Pb, and Cr reduce leaf area, limit cell division, and suppress photosynthetic activity in terrestrial plants. However, these investigations have primarily focused on agricultural or inland plant species, with relatively few studies addressing halophytes—plants naturally adapted to saline and intertidal environments.

Mangrove halophytes like *Lumnitzera racemosa* Willd. are often exposed to both salinity stress and contamination from nearby mining and industrial activities. Although halophytes possess adaptive traits such as succulence, salt glands, and thick cuticles that allow them to thrive in saline habitats, the combined effects of multiple heavy metals in ultramafic and mining-influenced soils on their leaf morphometric traits remain poorly understood. Most prior

works examined the effect of individual metals or general pollution stress, overlooking the synergistic or antagonistic interactions among multiple metals and their impact on leaf morphology and nutrient balance in mangrove ecosystems.

This research addresses that gap by investigating the variability of leaf morphometric and nutrient traits of *Lumnitzera racemosa* Willd. under multi-metal contamination in an ultramafic mining site compared to a control site. Understanding how heavy metals collectively influence the leaf traits of this halophyte provides new insight into the species' adaptive mechanisms and contributes to the broader understanding of mangrove resilience and bioindicator potential in contaminated coastal environments.

2. MATERIALS AND METHODS

2.1 The Study Site

Two distinct locations were carefully selected to facilitate a comparative analysis of the variables under investigation (Figure 1). The first study site was located in Sitio Kinalablaban, Barangay Cagdianao, Municipality of Claver, Province of Surigao del Norte. This area lies within the delta formed by the confluence of two small streams that drain into the coastal zone. The site was deliberately chosen because of its direct exposure to surface runoff and sediment deposition originating from active nickel mining operations in the upper catchment. As a result, the surrounding soils and sediments are potentially enriched with heavy metals. The broader upland region encompassing this watershed consists predominantly of lateritic soils derived from the prolonged weathering of exposed ultramafic rocks.

The first study site is strongly influenced by prevailing climatic systems, particularly the northeast monsoon (Amihan), the trade winds, and the easterlies, which collectively govern the seasonal hydrological and sedimentary dynamics in the area (Villarin et al. 2016). During periods of intense rainfall, the site experiences severe fluvial disturbances due to the high-energy flow of floodwaters. These hydrodynamic forces have led to observable morphological deformities in the mangrove vegetation, as indicated by the stunted growth and the distinct swirling or bending of boles among mature trees—an indication of mechanical stress caused by turbulent water movement and unstable substrates.

The second study site was established in Sitio Kabagtokan, Barangay Ata-atahon, Nasipit, Agusan del Norte, approximately 170 kilometers from the first location. This area represents a relatively undisturbed mangrove estuary that is not subjected to mining-derived effluents or sedimentation from upstream sources. In contrast to the mining-affected site, the Kabagtokan mangrove system remains hydrologically

stable and is minimally influenced by riverine flooding, even during episodes of heavy precipitation. The area provides an ideal control site for assessing baseline ecological and physicochemical conditions in a natural mangrove ecosystem.

Climatically, both study areas fall under the Type II classification of the Modified Coronas Climate System, characterized by a pronounced wet season with peak rainfall typically occurring in November and December, and an absence of a well-defined dry season. This climatic regime plays a critical role in determining the hydrological patterns, sediment input, and nutrient dynamics within the mangrove environments of both locations.

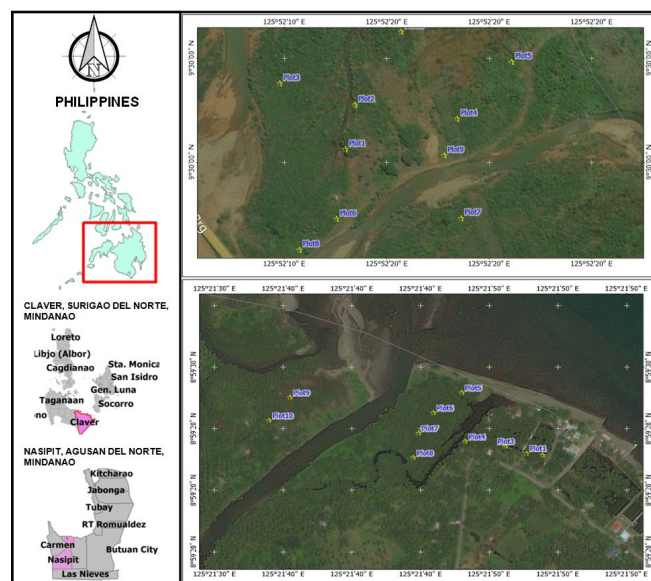


Figure 1: Location of the study

2.2 Sediment Analysis

Sediment samples from both locations were obtained for laboratory examination. The collection process followed the protocol set by the Regional Soils Laboratory under the Department of Agriculture – Caraga Region, which involves arranging sampling points in an 'S'-shaped pattern within the test plots. Once gathered, the samples were air-dried, ground, stored in clear polyethylene zip-lock bags, properly labeled, and analyzed for concentrations of molybdenum (Mo), manganese (Mn), lead (Pb), chromium (Cr), nickel (Ni), cadmium (Cd), and selected macronutrients.

2.3 Leaf Size Indices

Leaf samples of *Lumnitzera racemosa* from both study sites were collected. To eliminate additional sources of variation, only the leaves from the first branch of the assessed trees were collected (Aribal et al., 2016). A Vernier caliper was used to determine leaf-size indices such as length, breadth, and petiole length, while leaf area used Cain and De Oliveira Castro's (1959) formula of multiplying the leaf and width with a correction factor (CF) of 2/3. Variations in leaf

appearance and other characteristics such as color, chlorosis, browning, yellowing, the presence of spots, and/or alterations in the leaf's typical pigment, as well as the shape, either normal or distorted, were also observed.

3. RESULT AND DISCUSSION

3.1 Heavy Metals Concentrations in Sediments

Chromium (Cr) and nickel (Ni) concentrations in the sediments of Claver were recorded at 4,434 ppm and 4,234 ppm, respectively (Figure 2), both substantially exceeding the maximum permissible limits set by WHO and FAO at 100 for Cr and 50 for Ni (Table 1). These values are exceptionally high, reflecting the strong geochemical influence of the surrounding ultramafic terrain and the direct discharge of mine tailings and eroded materials from upstream nickel extraction activities. The unusually high concentration of these metals in Claver sediments confirms that mangrove ecosystems located near mining zones act as natural sinks for metal pollutants, accumulating contaminants through tidal influx, sediment deposition, and adsorption on fine particulate matter. This condition has been widely reported in mangrove environments adjacent to mining operations, where sediments serve as both storage sites and long-term sources of toxic metals to the biota (Lwin, 2022; Si et al., 2025).

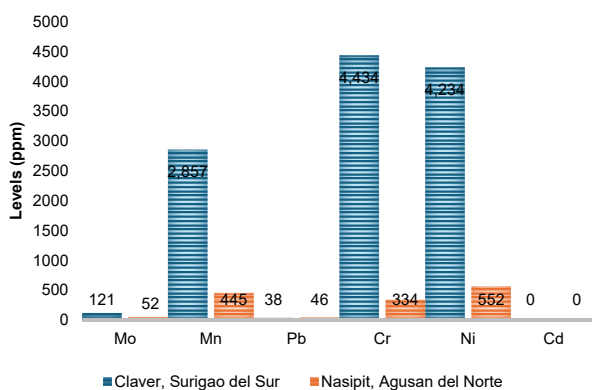


Figure 2: Heavy metal concentrations on the soil samples from Claver and Nasipit in parts per million (ppm)

While extremely elevated Ni levels are expected due to the proximity of the area to active nickel mining, the high chromium concentration is particularly noteworthy. Chromium often coexists with nickel in mineralized ultramafic deposits due to their geochemical association, as they both originate from the weathering of serpentinized peridotite rocks. Their co-occurrence suggests that natural lithological weathering is compounded by anthropogenic inputs. The simultaneous presence of these elements can further alter sediment chemistry by affecting pH and redox potential, which

consequently determines the bioavailability and toxicity of metals to mangrove flora and fauna.

Manganese (Mn) concentrations in Claver sediments (2,857 ppm) also exceeded the permissible threshold of 2,000 ppm. Although Mn is an essential micronutrient for plant metabolism, excess Mn can become toxic and interfere with the uptake of other nutrients such as iron, magnesium, and calcium. Its elevated level in Claver sediments may be due to the oxidative weathering of lateritic soils rich in Mn oxides and hydroxides, which act as scavengers of other metals. These oxides can absorb Cr and Ni, contributing to their high retention in the sediment layer.

Table 1: Permissible limits for heavy metals in soil

Heavy Metals	Maximum levels (ppm)	Heavy Metals	Maximum levels (ppm)
Arsenic (As)	20	Lead (Pb)	60
Cadmium (Cd)	3	Manganese (Mn)	2000
Cobalt (Co)	50	Molybdenum (Mo)	200
Chromium (Cr)	100	Nickel (Ni)	50
Copper (Cu)	100	Selenium (Se)	10
Iron (Fe)	50000	Zinc (Zn)	300

Source: WHO and FAO from Feyisa et al. (2025)

Sediment analysis from Nasipit revealed Ni and Cr levels of 552 ppm and 334 ppm, respectively, both surpassing permissible limits despite the absence of nearby mining activities. This implies that heavy metals can reach mangrove environments through non-point sources such as atmospheric deposition, riverine transport, or agricultural runoff. Wan et al. (2024) noted that diffuse sources of metal pollution—including the use of phosphate fertilizers, vehicular emissions, and domestic wastewater—can contribute significantly to background metal accumulation even in non-industrialized regions. Over time, these inputs are redistributed through tidal flushing and surface flow, allowing heavy metals to settle and accumulate within mangrove sediments.

The presence of heavy metals in Nasipit also highlights the high retention capacity of mangrove soils, which are rich in organic matter and fine clay particles. These components enhance the cation exchange capacity of sediments, facilitating metal binding and immobilization. However, environmental conditions such as salinity fluctuations, anaerobic decomposition, and microbial activity may cause the remobilization of these metals, increasing their potential bioavailability. Thus, even sites not directly exposed to mining, such as Nasipit, can still exhibit measurable contamination levels due to hydrological and geochemical connectivity with adjacent areas.

Interestingly, lead (Pb) concentrations in both Claver (38 ppm) and Nasipit (46 ppm) were within acceptable limits of 60 ppm. This may be attributed to Pb's strong tendency to form insoluble compounds that limit its mobility under reducing

conditions prevalent in mangrove soils. Similarly, manganese levels in Nasipit (445 ppm) were within permissible limits, reflecting the relatively lower degree of anthropogenic disturbance.

The observed difference in heavy metal concentrations between Claver and Nasipit underscores the influence of geological origin, land use practices, and hydrological connectivity on sediment chemistry. The Claver site, being directly affected by mining runoff, demonstrates the highest contamination intensity, while Nasipit provides a useful comparative baseline for understanding how secondary sources contribute to background metal accumulation.

The markedly high concentrations of Ni and Cr in Claver suggest a long-term legacy effect of mining, where continuous input from surface runoff and erosion sustains metal enrichment. This condition has important ecological implications because heavy metals, once deposited in sediments, can persist for decades and influence the physiology and growth of mangrove species such as *Lumnitzera racemosa*. Furthermore, the synergistic and antagonistic interactions between metals (Lwin, 2022) may modulate their toxicity; for instance, the presence of high Ni can suppress Cr uptake, while in some cases, their coexistence can intensify oxidative stress in plants.

3.2 Leaf Size Indices

The comparative analysis of leaf morphometric traits revealed marked variations between *Lumnitzera racemosa* populations in Claver and Nasipit (Figure 3). Trees growing in Nasipit exhibited greater average leaf length (5.17 cm) and leaf area index (7.53 cm²) than those in Claver (4.17 cm and 6.83 cm, respectively). Conversely, leaf width and petiole length were slightly higher in Claver at 2.46 cm and 1.34 cm respectively, compared to 2.18 cm leaf width and 1.15 cm petiole length in Nasipit.

Variations in leaf morphology often reflect adaptive responses to environmental stressors. Li and Wang (2021) emphasized that leaf traits, such as size, thickness, and surface area, are highly plastic and can change in response to changes in water availability, nutrient status, and pollutant levels. In the current study, the smaller leaves observed in Claver likely represent a morphological response to chronic metal stress. High concentrations of Ni and Cr can disrupt photosynthetic and metabolic processes, reducing the rate of cell division and elongation in leaf tissues (Hu et al., 2023). Consequently, the total assimilatory surface area of leaves decreases, which can reduce photosynthetic efficiency and carbon fixation capacity.

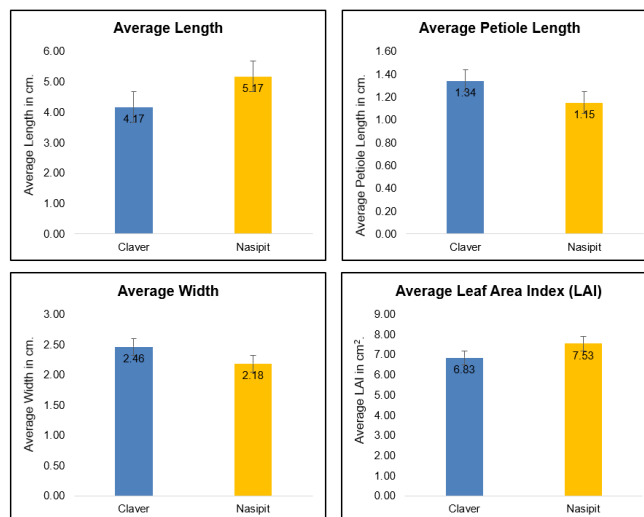


Figure 3: Comparisons of leaf morphometrics of *L. racemosa* in Claver and Nasipit

Similarly, Saravan and Sarkar (2022) observed that exposure of *Cyamopsis tetragonoloba* to Cd and Pb caused significant reductions in leaf area, shoot length, and root development. El-Khatib et al. (2020) reported comparable findings for *Eucalyptus globulus* and *Ficus nitida*, where heavy metal stress significantly decreased leaf area and specific leaf area. These physiological impairments are primarily associated with reduced chlorophyll content, impaired enzymatic activity, and oxidative stress. The findings in Claver are therefore consistent with established patterns observed in other plant species exposed to metal-contaminated environments.

The inhibitory effects of excess heavy metals can be attributed to several biochemical mechanisms. Heavy metals disrupt the normal functioning of chloroplasts and mitochondria, leading to decreased photosynthetic efficiency and ATP production. They also induce the formation of reactive oxygen species (ROS), which can damage cellular membranes and macromolecules (Zhu et al., 2020). Additionally, Cr and Ni interfere with the uptake and transport of essential nutrients such as Mg and Fe, further compounding the reduction in plant growth. The overall effect manifests as smaller, stunted leaves with limited biomass accumulation.

Interestingly, despite the reduced leaf length and area, *L. racemosa* in Claver exhibited slightly wider leaves and longer petioles compared to those in Nasipit. This counterintuitive finding may indicate a compensatory adaptation. Wider leaves could enhance light interception in sediment-laden environments where turbidity reduces light penetration, while longer petioles might help elevate the leaf lamina above the water surface, improving gas exchange and reducing submergence stress. Such plastic responses suggest that *L. racemosa* has a degree of morphological

flexibility that allows it to survive under multiple stress conditions, including heavy metal exposure and hydrodynamic disturbances.

Environmental salinity likely plays a complementary role in shaping leaf morphology. The mangrove forest in Nasipit, being closer to open estuarine waters, experiences higher salinity than the more freshwater-influenced Claver mangroves. Sanchez et al. (2021) reported that *Rhizophora mangle* developed narrower leaves under high salinity, an adaptation to reduce transpiration. Cao et al. (2023) further found that plants in low-salinity zones produce larger leaves to maximize gas exchange and carbon assimilation. Hence, the smaller leaf width in Nasipit and the broader leaves in Claver may reflect a trade-off between salinity stress and heavy metal toxicity.

Overall, the results suggest that *L. racemosa* expresses distinct morphological strategies in response to the combined influences of salinity, heavy metal contamination, and hydrological conditions. The significant morphological differences between populations indicate that this species possesses a high degree of phenotypic plasticity, a trait that may contribute to its ecological success in variable coastal environments.

The *t*-test analysis revealed statistically significant and consistent differences in the leaf morphometric traits of *Lumnitzera racemosa* between the two sampling sites, demonstrating how varying environmental conditions and heavy metal exposure influence leaf development (Table 2). On average, trees from the uncontaminated Nasipit mangrove estuary developed longer leaves, with a mean length of 5.17 cm, compared to 4.17 cm for those growing in the mining-influenced site of Claver. Likewise, the Leaf Area Index (LAI) was considerably higher in Nasipit (7.53) than in Claver (6.83), indicating a larger cumulative photosynthetic surface area and potentially greater canopy productivity in the less stressed site.

Table 2: T-test analysis for all variables

Trait	Claver (Mean)	Nasipit (Mean)	t(df)	p-value	Significance
Leaf length (cm)	4.17	5.17	499	2.91533E-54	hs
Leaf width (cm)	2.46	2.18	499	4.06665E-19	hs
Petiole length (cm)	1.34	1.15	499	3.64591E-18	hs
Leaf Area Index	6.83	7.53	499	5.85693E-05	hs

Note: hs = highly significant

In contrast, mean leaf width (2.46 cm) and petiole length (1.34 cm) were greater in Claver than in Nasipit (2.18 cm and 1.15 cm, respectively). This pattern suggests that while the mining-affected environment constrained overall leaf expansion and surface area, it may have triggered compensatory morphological adjustments such as broader

laminae and longer petioles. These traits can improve light interception efficiency and facilitate gas exchange under suboptimal conditions. Such plastic responses reflect the capacity of *L. racemosa* to tolerate physiological stress while maintaining basic photosynthetic functionality even under elevated metal concentrations.

All parameters analyzed yielded p-values well below the 0.05 significance threshold, confirming that the observed differences are highly significant and not attributable to random variation. The consistently high t-values and extremely low p-values provide strong statistical evidence that heavy metal contamination and associated soil stressors have a measurable influence on leaf morphology.

The suppression of leaf elongation and total leaf area in Claver may be attributed to physiological constraints such as inhibited cell division, altered water relations, and oxidative stress induced by elevated concentrations of nickel, chromium, and manganese in the sediments. Similar trends have been documented by Hu et al. (2023) and El-Khatib et al. (2020), who reported reduced leaf dimensions and photosynthetic efficiency in plants exposed to heavy metal stress. Conversely, the larger and more proportionate leaves in Nasipit reflect an environment conducive to normal metabolic processes, enabling optimal nutrient uptake, chlorophyll synthesis, and cell expansion.

Taken together, the results indicate that *L. racemosa* demonstrates both sensitivity and adaptive plasticity in leaf morphology in response to geochemical stress. The contrasting trends—narrower but longer leaves in Nasipit versus shorter but wider leaves in Claver—highlight how environmental stress can differentially shape growth strategies. This morphological variability provides valuable insight into how mangrove species physiologically adjust to metal-polluted environments and supports the use of leaf morphometric traits as reliable bioindicators of habitat quality and contamination levels in coastal ecosystems.

3.3 Leaf Nutrient Content (NPK Analysis)

To determine whether the observed morphological differences were due to nutrient imbalance rather than metal toxicity, leaf samples were analyzed for macronutrients (Table 3). The results showed that *L. racemosa* leaves in both Claver and Nasipit were deficient in nitrogen (1.57% and 1.07%, respectively), below the optimum requirement for healthy leaf growth (2.4–2.8%) as shown in Table 4. Nitrogen deficiency typically manifests in reduced leaf size, chlorosis, and lower photosynthetic rates. Aribal et al. (2016) emphasized that nitrogen is critical for chlorophyll and protein synthesis; thus, N limitation directly constrains leaf expansion and carbon assimilation.

The higher phosphorus concentration in Nasipit (0.25%) compared to Claver (0.12%) may partially explain the larger leaves observed in the former site. Phosphorus is essential for energy transfer and nucleic acid metabolism, and adequate P supply enhances root growth and photosynthetic efficiency. The relatively low P level in Claver suggests restricted energy metabolism, likely exacerbated by heavy metal stress, which can cause P immobilization in the soil through precipitation with Fe and Al oxides.

Potassium levels in both sites were above the optimum range, reaching 2.77% in Claver and 1.33% in Nasipit. Elevated K concentrations may be a compensatory response to osmotic imbalance caused by heavy metals and salinity. Potassium regulates stomatal conductance and osmotic potential, helping plants maintain turgor under stress conditions. Islam et al. (2024) reported that plants exposed to high metal concentrations often accumulate K to stabilize internal ionic equilibrium and mitigate oxidative damage.

Overall, the NPK results suggest that nutrient imbalance interacts with heavy metal stress to influence leaf morphology. The combined deficiency in N and excess of K, particularly in Claver, may further disrupt physiological processes such as photosynthesis, enzyme activation, and carbohydrate metabolism. Consequently, the smaller leaf length and area in Claver could result from both heavy metal toxicity and nutrient stress acting synergistically.

Table 3: NPK leaf nutrient contents of *L. racemosa* in Claver and Nasipit

Location	Nitrogen (%)	Phosphorus (%)	Potassium (%)
Claver	1.57	0.12	2.77
Nasipit	1.07	0.25	1.33

Table 4: Plant macronutrient requirements

Interpretation	Nitrogen (%)	Phosphorus (%)	Potassium (%)
Deficient	<2.2	<0.09	<0.40
Low	2.2 - 2.3	0.09 - 0.11	0.40 - 0.69
Optimum	2.4 - 2.8	0.12 - 0.16	0.70 - 1.09
High	2.7 - 2.8	0.17 - 0.29	1.10 - 2.00
Excess	> 2.8	> 0.30	> 2.30

Source: Agriculture and Food Development Authority, cited by Aribal et al. (2016)

4. CONCLUSION

This study demonstrated clear morphological and physiological distinctions between *Lumnitzera racemosa* populations growing in metal-contaminated and uncontaminated mangrove environments in northeastern Mindanao. The comparative analysis between the mining-affected site in Claver and the uncontaminated site in Nasipit revealed that exposure to heavy metals—particularly those

derived from ultramafic and lateritic substrates—exerts a substantial influence on leaf morphometric traits and overall plant performance. Significant differences were observed in all leaf size indices, with individuals from Nasipit developing longer leaves, wider laminae, and higher leaf area indices than those from Claver. The results of the t-test analysis confirmed that these differences were highly significant ($p < 0.05$), indicating that heavy metal stress can inhibit leaf expansion and alter morphological expression in *L. racemosa*.

The observed reduction in leaf size and petiole length among trees in Claver likely reflects physiological stress responses to excessive metal accumulation, nutrient imbalance, and the disturbance of normal water and ion regulation processes. In contrast, the relatively stable and unpolluted mangrove environment in Nasipit provides more favorable conditions for optimal growth and metabolic activity, resulting in more vigorous and structurally uniform foliage. These findings highlight the high degree of morphological plasticity exhibited by *L. racemosa*, suggesting that this species can survive and maintain growth even under suboptimal and metal-enriched conditions, albeit with measurable structural adjustments.

Overall, the results underscore the importance of *L. racemosa* as a potential bioindicator species for assessing the ecological impacts of mining-derived metal contamination in mangrove ecosystems. The significant contrasts in leaf traits between the two sites reflect both the adaptive strategies of the species and the degree of environmental stress imposed by anthropogenic activities. Future research integrating physiological and biochemical assessments, such as chlorophyll content, metal bioaccumulation, and antioxidant enzyme activity, would further elucidate the mechanisms underlying the species' tolerance and adaptive capacity. Such insights are crucial for formulating evidence-based strategies in mangrove conservation, rehabilitation, and the monitoring of metal pollution in coastal environments.

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