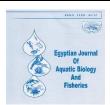
Egyptian Journal of Aquatic Biology & Fisheries Zoology Department, Faculty of Science, Ain Shams University, Cairo, Egypt. ISSN 1110 – 6131 Vol. 29(6): 449 – 461 (2025) www.ejabf.journals.ekb.eg



Bioaccumulation of Lead (Pb) and Microplastic Contamination in the Estuary of Kombal Bay, North Lombok, Indonesia Using *Padina* sp. as a Bioindicator

Reagan Saptory¹, Daniar Kusumawati^{1*}, Moh. Awaludin Adam¹, Apri I. Supii¹, Salnida Yuniarti Lumbessy², Hasriani Ayu Lestari¹, Raditya Andrean Saputra³, Sabilah Fi Ramadhani³, Asmanik¹, Lia Hadiawati⁴, Joko Sumarwan¹

¹Research Center for Marine Aquaculture, National Research and Innovation Agency (BRIN), Lombok, Indonesia

²Program Study of Aquaculture, Faculty of Agriculture, Mataram University, NTB, Indonesia

³Indonesia Biru Foundation (IBF), Indonesia

⁴Research Center for Food Crop, National Research and Innovation Agency (BRIN), Cibinong, Indonesia

Corresponding Author: dani019@brin.go.id

ARTICLE INFO

Article History:

Received: Aug. 25, 2025 Accepted: Oct. 27, 2025 Online: Nov. 14, 2025

Keywords:

Environmental pollutants, Heavy metal Pb, Microplastics, Lombok, *Padina* sp.

ABSTRACT

The Kombal Bay estuary in North Lombok, Indonesia, shows increasing pollution from anthropogenic sources such as shipping and domestic waste. This study assesses lead (Pb) bioaccumulation and microplastic contamination using Padina sp. as a bioindicator. Water, sediment, and tissue samples from Padina sp. were used in the sampling process. The content of Pb was analyzed using Atomic Absorption Spectrophotometry (AAS), while the distribution of Pb in tissues was analyzed using Scanning Electron Microscopy (SEM). Microplastics were analyzed by examining their morphological characteristics, type, and color using an optical microscope (Olympus Inverted). The research results show that *Padina* sp. significantly accumulates Pb, while the Pb content in the water is still below the quality standard threshold. SEM analysis revealed a consistent distribution of Pb throughout all part of *Padina* sp. tissue. On the other hand, microplastic contamination in Kombal Bay showed abundances ranging from 19.5±14.71 to 25±14.54 particles/L in water, 9.25±8.54 to 10.75±9.53 particles/kg in sediment, and 10.25±13.28 to 10.5±10.28 particles/individual on Padina sp. The types of microplastics found include fragments, films, pellets, and fibers, with dominant color variations of yellow, blue, red, black, and green. This finding indicates that coastal ecosystems are experiencing multi-pollutant pollution stress, which could potentially harm aquatic organisms and human health. Therefore, mitigation measures for pollution and further research are needed regarding the long-term toxicological impacts of heavy metals and microplastics on marine biota.

INTRODUCTION

Water pollution is a global problem that has become increasingly serious due to the rise in anthropogenic activities, such as domestic, industrial, agricultural, and maritime transportation activities (**Adam** *et al.*, **2024**; **Massahi** *et al.*, **2024**). This condition also occurs in the coastal area of North Lombok Regency, West Nusa Tenggara, particularly in Kombal Bay, which shows signs of multi-pollutant contamination, including heavy metals







and microplastics (Aprilia et al., 2024; Wu et al., 2024). Both types of contaminants are toxic and persistent and have the potential to disrupt marine ecosystem balance, food chains, and human health (Karlsson et al., 2017; Murphy et al., 2022).

Located near the Three Gili Islands, Kombal Bay has a small harbor and serves as a key docking point for ships and ferries catering to tourists visiting Gili Trawangan, Gili Meno, and Gili Air. However, the shipping activities in this bay have led to significant heavy metal pollution (Adam et al., 2023), particularly from lead residues in tetraethyl lead-based fuels (Krismonita et al., 2023; Sartika et al., 2024). Lead is a toxic, non-essential heavy metal that accumulates in living tissues and forms strong bonds with proteins (Wisha et al., 2019; Bakshi et al., 2023). Even minimal exposure to lead can be fatal to aquatic life due to chronic bioaccumulation (Buwono et al., 2022; Silva et al., 2023). Furthermore, the lead accumulation in the bay is influenced by various environmental factors, including water temperature, pH levels, oxygen content, and water hardness (Risjani et al., 2022; Widiastuti et al., 2025).

On the other hand, Kombal Bay is an estuarine area that receives waste runoff from various community activities on land, such as agriculture, fisheries, and households. This area has now become a hotspot for microplastic pollution (**Browne** et al., 2008; **Supriatna** et al., 2023). Microplastics (MPs) are plastic particles smaller than 5mm that originate from the degradation of larger plastics or primary plastic products. The types of MPs found include fragments, fibers, films, and pellets, with variations in size, color, and chemical composition (**Crawford & Quinn, 2017; Renner** et al., 2018). MPs can persist in the marine environment for a long time; they are ingested by organisms, and have the potential to disrupt physiological processes as well as serving as vectors for other contaminants (**Irnidayanti** et al., 2023; Wu et al., 2024).

One of the organisms vulnerable to exposure to both types of pollutants is macroalgae. As benthic organisms, macroalgae such as *Padina* sp. have a high tendency to absorb and accumulate heavy metals and microplastics from their surrounding environment (**Jaouani** *et al.*, **2022**; **Leman & Lubis**, **2023**). Therefore, macroalgae are often used as bioindicators of water quality (**Adam** *et al.*, **2024**; **Chairunnisa** *et al.*, **2025**). The presence of *Padina* sp. in the waters of Kombal Bay opens up opportunities to comprehensively assess the level of environmental pollution through a biological approach.

Based on this background, this study aims to analyze the accumulation of lead (Pb), heavy metals, and microplastics in the macroalgae *Padina* sp., as well as mapping its spatial distribution in the Kombal Bay region. The results of this study are expected to serve as a scientific basis for pollution mitigation efforts and the formulation of sustainable coastal ecosystem management strategies. We hypothesize that *Padina* sp. could serve as an effective bio monitor for Pb and microplastic pollution, with accumulation patterns reflecting the intensity of local anthropogenic activities.

MATERIALS AND METHODS

The research was conducted from August to September 2025 in the waters of Kombal Bay, Pemenang District, North Lombok Regency, West Nusa Tenggara. Field sampling was conducted at three stations—S1, S2, and S3—to collect water, sediment, and macroalgae (*Padina* sp.). The analysis of Pb metal content was conducted at the Analytical Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Brawijaya University, Malang.

The distribution of Pb in *Padina* sp. tissue was mapped utilizing a Hitachi SU3500 Scanning Electron Microscope (SEM) at the Advanced Characterization Laboratories, Serpong, National Research and Innovation Agency (BRIN).

Location and sampling technique

Sampling was conducted at three sampling points in Kombal Bay: S1 (estuary), S2 (near agricultural/fishing areas), and S3 (near settlements and waste disposal sites). Water samples were collected from the surface and bottom layers using plastic bottles, composited, and stored in labeled 1000mL polyethylene bottles placed in an ice box. Sediment samples were collected using a scoop during low tide, placed in labeled plastic bags, and stored in a cool box. *Padina* sp. samples were randomly collected by hand (10–20 colonies per site) during low tide.

Sample preparation

Water samples were composited, added with HNO₃ until pH < 2, then stored in a low temperature. Water quality parameters (temperature, pH, salinity, DO) were measured *in situ*.

The sediment was dried for 7 days, ground, sieved (mesh $63 \mu m$), and then stored in labeled bags.

Padina sp. were washed with seawater, then rinsed with freshwater, dried, and labeled. Sample replication number (n = 3 per station) was included for statistical reproducibility.

Analysis of Pb metal content

For the analysis of Pb content in the water, sediment, and *Padina* sp., the Atomic Absorption Spectrophotometry (AAS) method was used. Water samples were added with methyl isobutyl ketone, APDC, and NaDDC to remove major ions. Sediments were added with HF and heated to 130°C. *Padina* sp. tissue was degraded with concentrated HNO₃ and HClO₄ at a temperature of 60–70°C for 2–3 hours until being clear. After pre-process, all samples were measured using AAS with an air-acetylene flame.

Bioconcentration factor (BCF)

The bioconcentration factor was calculated to determine the accumulation capacity of Pb metal in *Padina* sp. in relation to water and sediment, using the formula:

BCF_o-w = C_org/C_water, BCF_o-s=C_org/C_sed

where:

C_org: Pb concentration in *Padina sp.* tissue

C_water: Pb concentration in water C_sed: Pb concentration in sediment

SEM preparation and observation

SEM preparation was carried out through the stages of fixation (using glutaraldehyde and osmium tetroxide), graded dehydration in 70% to 100% alcohol, drying using CPD or

hexamethyldisilazane, and conductive coating (C, Au, or Pt) using a sputter coater. The observation was conducted using a Hitachi SU3500 in variable pressure mode (VP-SEM).

Extraction and quantification of microplastics

Sediment and *Padina* sp. samples were dried at 70°C for 3 days, then treated with Fe (II), H₂O₂, and 30% NaOH solution (for *Padina* sp. only), and heated on a hotplate until bubbles appeared. After cooling, 6g of NaCl/ 20mL was added, heated, and allowed to settle for 24 hours. The solution was filtered using a 0.3µm plankton net and Whatman paper, then observed under a microscope. Microplastics were identified based on shape (fragments, fibers, films, pellets), color, and quantity.

Water quality analysis

Water quality testing included chemical parameters (NH₄⁺, NO₃⁻, NO₂⁻, PO₄³⁻) and physical parameters (temperature, DO, TDS), using Salifert test kits and a TDS Meter.

Data analysis

The measurement data were statistically evaluated with IBM SPSS v25 software. ANOVA tests were employed to detect differences across locations, and T-tests were performed to compare the number and properties of microplastics. Data were tabulated with Microsoft Excel 2013.

RESULTS

Heavy metal content of pb in water, sediment, and *Padina* sp.

The measurement of lead (Pb) concentration showed that sediment contained the highest Pb level compared to water and *Padina* sp. tissue (Table 1).

Table 1. Bioaccumulation of Pb heavy metals in Kombal Bay

Parameter / Station	Lead (Pb)	Lead (Pb)	Lead (Pb)	Factor B	Bio-	Factor	Bio-
	Water	Sediment	Padina sp.	accumulation		accumulation	
	(ppm)	(ppm)	(ppm)	Water		Sediment	
S1	0.0037	0.0216	0.0196	5.2972		0.9074	
S2	0.0049	0.0218	0.0195	3.9795		0.8944	
S3	0.0058	0.0277	0.0212	3.6551		0.7653	

Bio Concentration Factor (BCF)

The Bio Concentration Factor (BCF) was calculated to determine *Padina* sp.'s ability to acquire lead from water and sediment. *Padina* sp. had a higher BCF value for water (BCF o-w) compared to sediment (BCF o-s), indicating better Pb absorption from water. This BCF result demonstrates *Padina* sp.'s role as a bioindicator of heavy metal Pb in coastal settings.

SEM (Scanning Electron Microscopy) observation

SEM analysis of *Padina* sp. tissue (Fig. 1.) reveals uneven surface morphology, including scratches, gaps, and foreign particles (Fig. 2) that could represent heavy metals or clinging chemicals.

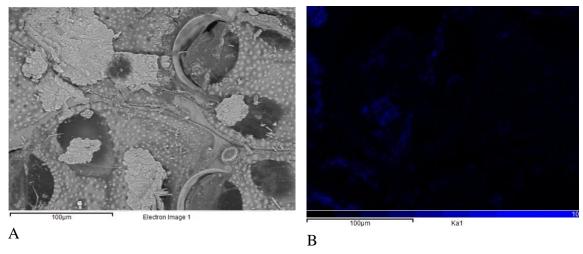
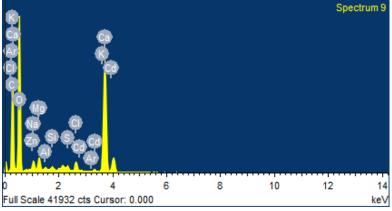


Fig. 1. (A) Observation of *Padina* sp structure with SEM observation of 100μm magnification (B) Mapping of Pb distribution in *Padina* sp. structure with SEM observation of 100 μm magnification



Element	Weight%	Atomic%
СК	33.99	44.65
ОК	44.20	43.58
Mg K	1.82	1.18
Al K	4.25	2.49
Si K	7.85	4.41
Cl K	2.36	1.05
Ca K	1.84	0.72
Pb M	0.03	0.00

Fig. 2. Distribution of Pb in the *Padina* sp. structure with SEM observations at 100μm magnification and SEM observation data spectrum on Pb uptake in *Padina* sp.

Content and type of microplastics in Padina sp. and sediment

Table (2) displays the abundance of MPs in the three sampling areas in Kombal Bay, representing the initial step of this study.

Table 2. The abundance of the microplastics Kombal Bay

Type of Comple	The abundance of MPs at three sampling points			
Type of Sample	Site 1	Site 2	Site 3	
Water (particle/L)				
Mean±SD	19.5 ± 14.71	25 ± 14.54	21.5±14.62	
Sediment (particle/kg dw)				
Mean±SD	10.75 ± 9.53	9.25 ± 8.53	9.75±6.31	
Padina sp. (particle/individual)				
Mean±SD	15.25±9.53	10.25 ± 13.3	10.5±10.27	

Following the determination of MPs abundance, their forms were identified, as depicted in Fig. (3).

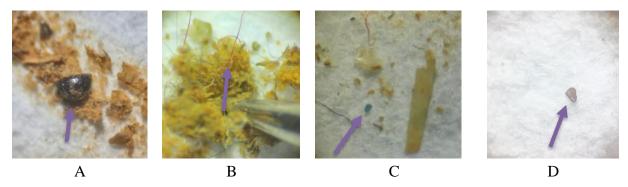


Fig. 3. Forms of microplastics: (a) Fragments, (b) Film, (c) Fibers, and (d) Pellets. Using an inverted microscope, four forms of microplastics (MPs) were identified in water, sediment, and *Padina* sp. samples—fragments, films, fibers, and pellets.

The distribution of these microplastic forms across the sampling stations is shown in Fig. (4).

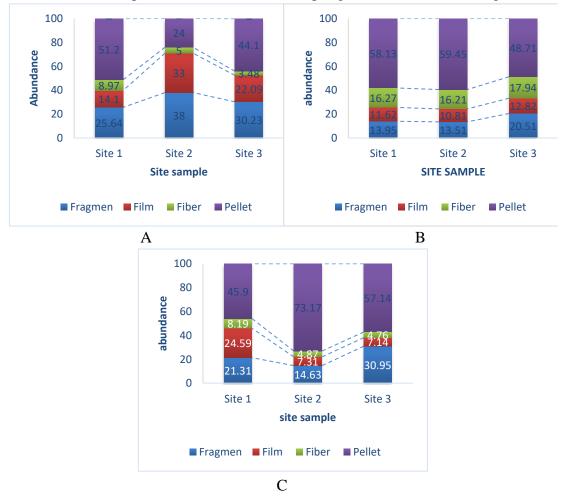


Fig. 4. The percentage (%) of MPs abundance collected from (a) water, (b) sediment, (c) and *Padina* sp.

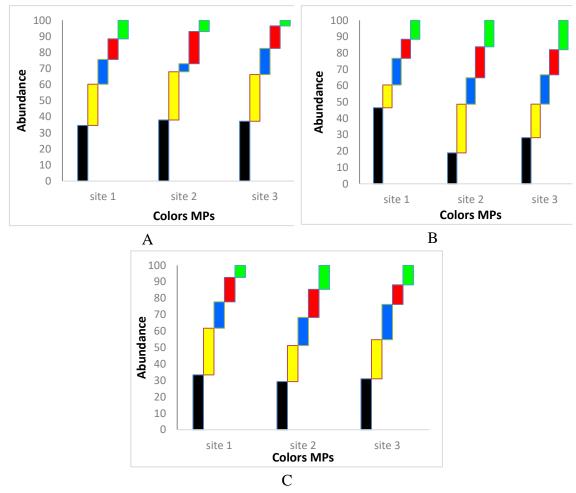


Fig. 5. The dominant colors of MPs collected from the a) water, b) sediment, and c) *Padina* sp. samples

Water quality as a supporting factor

Water quality parameters such as temperature, pH, salinity, DO, ammonia, phosphate, and nitrate are still within the tolerance range for marine organisms. However, nutrient concentration values such as ammonia and phosphate tend to be higher in areas close to settlements and agriculture, which could potentially worsen the condition of coastal ecosystems in the long term.

Table 3. Comparison of Kombal Bay river water quality based on chemical and physical parameters

Parameter	Water Quality	Sampling Point			
	Standards	S 1	S2	S3	
TDS (ppm)	500	259	274	337	
DO (mg/L)	3.00	6.00	5.00	4.00	
Ammonia(mg/L)	< 0.5	0.25	0.25	< 0.15	
Nitrate(mg/L)	1	0	0	0	
Nitrite(mg/L)	1	0	0	0	
Phosphate(mg/L)	0.5	0.1	0.25	0.25	

DISCUSSION

The concentrations of Pb in sediment, water, and *Padina* sp. tissues ranged from 1.84–3.96mg/ kg, 0.008– 0.019mg/ L, and 0.21– 0.66mg/ kg, respectively. The highest Pb concentrations were observed in locations near anthropogenic activities such as residential areas and garbage disposal sites. This pattern indicates that human activities significantly influence the accumulation of heavy metals in the aquatic environment. Sediments act as the final repository for heavy metal contaminants due to their strong adsorption capacity, as supported (**Adam** *et al.*, **2025**; **Widiastuti** *et al.*, **2025**), who noted that heavy metals, being largely insoluble in water, tend to associate with particulate matter and settle into the sediment column. Consequently, elevated Pb in sediments may serve as a long-term indicator of environmental stress and pollution.

Morphological observation of *Padina* sp. revealed that specimens from Pb-rich sites exhibited more severe surface damage and cracks on their thallus compared to those from relatively unpolluted sites. This suggests that *Padina* sp. responds structurally to heavy metal stress, consistent with the findings of **Risjani** *et al.* (2022) and **Chairunnisa** *et al.* (2025), who reported that heavy metals can disrupt algal cell wall integrity and alter metabolic and physiological processes. Thus, the observed physical degradation in *Padina* sp. likely reflects the synergistic effects of Pb accumulation and other contaminants in Kombal Bay. Moreover, *Padina* sp. tends to absorb more Pb from the water column than from sediment because its thallus surface is in direct and continuous contact with surrounding seawater, allowing efficient adsorption through ion exchange and binding to polysaccharides such as alginate and fucoidan in the cell wall. In contrast, sediment-derived Pb is often bound to particulate matter or organic complexes, making it less bioavailable for uptake. Therefore, the higher Pb concentration in *Padina* sp. primarily reflects the dissolved and suspended Pb fraction in the water rather than the sediment-bound fraction (Saldarriaga-Hernandez *et al.*, 2020).

In addition to heavy metal contamination, microplastic (MP) pollution was also prominent in the study area. The mean abundance of MPs in water, sediment, and Padina sp. were 19.5 ± 14.71 particles/L, 10.75 ± 9.53 particles/kg (dry weight), and 15.25 ± 9.53 particles/individual, respectively. Interestingly, the estuary site exhibited the lowest MP concentration in water but the highest in sediments and algal tissues. This pattern highlights the estuary's function as a sink, where microplastics transported from upstream accumulate and eventually settle (Barboza et al., 2018; Chakraborty et al., 2022; Gilroy et al., 2023). The bioaccumulation of MPs in Padina sp. further suggests that this species may serve as a potential bioindicator of plastic pollution in coastal ecosystems.

Microscopic identification revealed four major MP morphotypes—fibers, films, fragments, and pellets—with varying colors and material origins. Fibers, mostly black, yellow, red, white, and blue, likely originated from fishing gear or textile fibers (Hadi et al., 2022; Massahi et al., 2024). Films appeared transparent to brownish, suggesting household plastic sources (Gopal et al., 2022; Irnidayanti et al., 2023). Fragments, typically opaque and thicker, originated from degraded plastic containers or packaging materials (Junaidi et al., 2020; Jaouani et al., 2022), while pellets were mostly black and possibly derived from cosmetic microbeads or industrial resins (Supriatna et al., 2023; Wu et al., 2024). The diversity in

microplastic shapes and colors underscores the multiple anthropogenic sources contributing to contamination (Murphy et al., 2022; Leman & Lubis, 2023).

Spatially, pellet-type MPs were dominant at sampling points S1 and S3, while fragments prevailed at S2. The high MP abundance at S1 (downstream of Kombal Bay) may be linked to the inflow of riverine plastic waste and the frequent use of fishing nets, which release synthetic fibers. This finding aligns with **Ghosh** *et al.* (2021), who observed similar pellet dominance in the East Flood Canal estuary, largely due to urban runoff and domestic waste discharge. Furthermore, film-shaped MPs (mainly polyethylene and polypropylene) found across all stations are associated with household plastic bags and are more prone to degradation (Sobhani *et al.*, 2019; Wu *et al.*, 2024). The presence of black-colored MPs also suggests a mix of organic matter or oxidized plastic (Adam *et al.*, 2024; Chairunnisa *et al.*, 2025).

The coexistence of heavy metals and microplastics in Kombal Bay indicates potential synergistic pollution effects, as microplastics can adsorb heavy metals onto their surfaces, enhancing metal bioavailability and toxicity to marine biota. This relationship has been widely reported in recent studies (Hara et al., 2020; Koongolla et al., 2020; Massahi et al., 2024), reinforcing the ecological risk posed by combined contamination in estuarine environments. Furthermore, the accumulation of Pb and microplastics in Padina sp. suggests the possibility of trophic transfer through the food web, as herbivorous invertebrates and fish feeding on benthic algae could assimilate these contaminants. The sediment—water flux of Pb also plays a critical role in maintaining contamination levels, where resuspension or diffusion processes can reintroduce sediment-bound Pb into the water column, sustaining chronic exposure to aquatic organisms. These mechanisms collectively imply potential risks to local fisheries, as bioaccumulation and biomagnification may result in elevated Pb levels in edible species, posing health concerns for coastal communities relying on these resources for food and livelihood (Boshoff et al., 2023; Ritter & Bourne, 2024).

Regarding water quality, ammonium levels (<0.15– 0.25mg/ L), phosphate concentrations (0.1– 0.25mg/ L), and dissolved oxygen (4– 6mg/ L) remained within the acceptable limits of the Indonesian water quality standards (**Peraturan Pemerintah No 22 Tahun, 2021, 2021**). The absence of detectable nitrate and nitrite further indicates limited eutrophication pressure (**Tong** *et al.*, **2023**; **Sun** *et al.*, **2024**). However, the detectable presence of Pb and microplastics suggests that despite good physicochemical water quality, non-nutrient pollutants remain a concern. This highlights the need for integrated management approaches that address both chemical (heavy metals) and physical (microplastics) pollutants in coastal ecosystems like Kombal Bay.

CONCLUSION

This study demonstrates that *Padina* sp. can accumulate the heavy metal Pb and microplastics in the coastal waters of Kombal Bay, North Lombok. The highest Pb content was detected in the sediment, followed by the *Padina* sp. tissue and seawater. The relatively high BCF value indicates that *Padina* sp. is effective in absorbing Pb from the aquatic environment, especially from water. Observation of tissue morphology with SEM revealed structural damage

on the thallus surface due to heavy metal exposure. Additionally, *Padina* sp. has also been proven to capture various types of microplastics, especially fiber types, with a dominance of blue and black colors. Overall, *Padina* sp. can be used as a bioindicator of environmental pollution, both for heavy metals like Pb and microplastics, and has the potential as a monitoring tool for water quality in coastal areas affected by human activities.

AUTHOR CONTRIBUTIONS

Conceptualization, M. A. A. and D.K.; methodology, M.A.A., R.S., H.A.L., and D.K.; software, M.A.A, S.Y.L, D.K., A., S.R. and R.S.; validation, A, M.A.A., D.K. and J.S.; formal analysis, R.S. and D.K.; investigation, M.A.A., A.I.S. and S.Y.L.; resources, M.A.A.; data curation, M.A.A and R.S.; writing—original draft preparation, M.A.A, R.S. and D.K.; writing—review and editing, M.A.A, H.A.L., A., S.R. and S.Y.L.; visualization, R.S., A.I.S., D.K., S.R. and A..; supervision, M.A.A.; project administration, M.A.A. .; funding acquisition, M.A.A.

FUNDING

The present study was financially supported funding from the National Research and Innovation Agency (BRIN) contract number B-26728/III.11/TK.01.01/9/2025 about the Research Batch II at Rumah Program OR Pertanian dan Pangan (ORPP) BRIN TA 2025.

ACKNOWLEDGEMENT

The authors acknowledge the facilities, scientific and technical support from Lombok Marine Bioindustry Laboratory and Advanced Characterization Laboratories, Serpong, National Research and Innovation Agency.

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