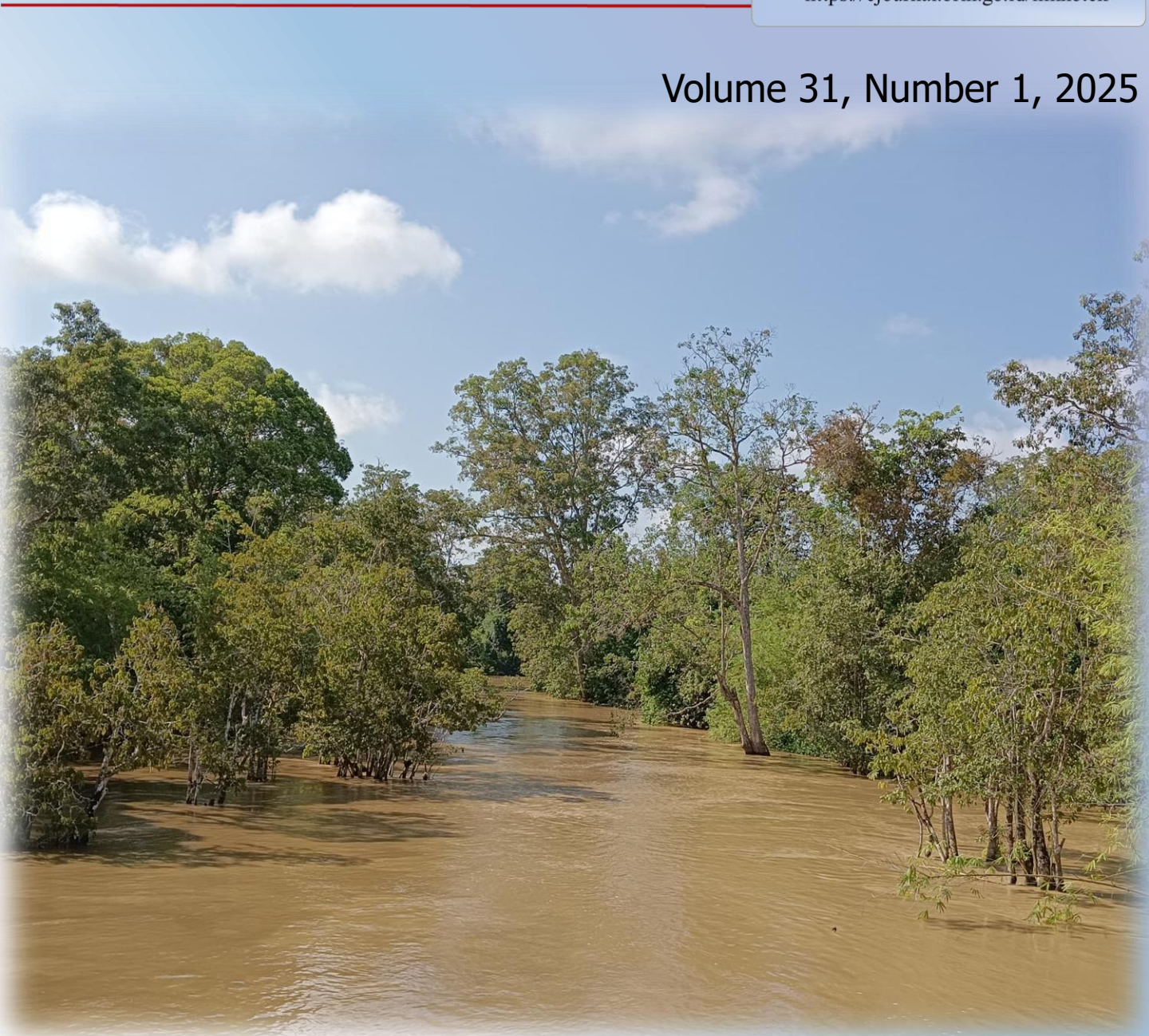


Volume 31, Number 1, 2025



National Research and Innovation Agency (BRIN)

Indonesian Limnology Society (MLI)



BRIN



p-ISSN: 0854-8390

e-ISSN: 2549-8029

**LIMNOTEK**

**Perairan Darat Tropis di Indonesia,**

***transforming into the Journal of Limnology and Water Resources***

**Volume 31, Number 1, 20 June 2025**

**DOI:** <https://doi.org/10.55981/limnotek.v31i1>

**Published by:** National Research and Innovation Agency (BRIN) and Indonesian Limnology Society (MLI)

**Advancing Sustainable Water Management Through Research and Innovation:  
Bridging Knowledge to Impact**

Limnotek transforming into Journal of Limnology and Water Resources (JLWR) continuously focusing and scoping in establishing an integrated understanding of the interface between natural water processes, inland aquatic ecosystems, and human interactions.

In this 2025 volume 1 issue, we are proud to present a collection of nine research articles and reviews that explore critical and interconnected aspects of limnology and water resources. Topics covered spatial analyses of water quality drivers in reservoirs, groundwater potential mapping using geospatial techniques, the impact of land cover changes on river water quality, and the ecological risks of mercury contamination near mining areas. Two studies highlight the growing concern of microplastic pollution in aquatic ecosystems from fishponds in Banten to the digestive tracts of invasive catfish in the Citarum River. Other contributions explore innovative bioremediation approaches using fungi and food waste, as well as the ecological functions of vetiver root extracts. Together, these studies reinforce our commitment to advancing sustainable and science-based water resource management across diverse inland water systems.

Aligned with our vision of continuous learning, innovation, and collaboration, we hope this volume provides valuable insights for researchers, scholars, practitioners, the publics and policymakers to join us in advancing the sustainable use and management of lakes and water resources.

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**Accreditation:** **SINTA-2** period 2020-2025, *Surat Keputusan Direktur Jenderal Pendidikan Tinggi, Riset dan Teknologi - Kementerian Pendidikan, Kebudayaan, Riset dan Teknologi Nomor 105/E/KPT/2022*, dated 7 April 2022



**Cover Image:** Lake Lais a natural lake in Tanjung Sangalang Village, Central Kalimantan. Images courtesy of Dr. Rosana Elvince, Study Program of Aquatic Resource Management, Department of Fisheries, Faculty of Agriculture, University of Palangka Raya, Central Kalimantan, Indonesia.



p-ISSN: 0854-8390

e-ISSN: 2549-8029

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## **Spatial Analysis of Physical Parameters Influencing Water Quality: A Case Study in Seloromo Reservoir, Central Java, Indonesia**

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Received: 08 August 2024; Accepted: 27 April 2025; Published: 20 June 2025

**Abstract:** Reservoirs are vital water resources that have an essential role from the perspective of living things. Utilization of reservoirs by humans frequently causes problems in the reservoir ecosystem, such as water pollution. Seloromo Reservoir was the second reservoir built to support various activities such as agriculture, fisheries, and tourism. Complex activities can influence the quality of reservoir water because sources of pollution are absorbed and flow into the water body. For this reason, monitoring is essential to determine factors that affect water quality of the reservoir. The aim of this research is to analyze the water quality of Seloromo Reservoir by testing physical parameters such as water temperature, degree of acidity (pH), Electrical Conductivity (EC), Total Dissolved Solid (TDS). In order to show the spatial distribution of each physical characteristic, grab samples were collected and measured in real time using a water checker. The results will be shown on a map. Spatial distribution maps were created by using the Inverse Distance Weighted (IDW) method which provides the simplest formulation and accurate results. Results show that human activities such as agriculture, livestock, and tourism around the reservoir and inputs from the river inlet indicate the condition of water quality in Seloromo Reservoir. Spatial distribution maps of physical parameters show that the northern area is more affected than the southern area of the reservoir. Local communities have an important role in conserving the water resources in this reservoir. Consequently, the research can contribute new knowledge about the condition of the reservoir and provide additional reference in establishing policies.

**Keywords:** reservoir, water quality, physical parameter, spatial distribution

DOI: <https://doi.org/10.55981/limnotek.2025.6546>

### **1. Introduction**

Reservoirs are freshwater resources to supply the water needs of life. According to the Regulation of the Minister of Public Works and Public Housing No. 27 of 2015, reservoirs are artificial water bodies formed as a result of the construction of dams. According to Nurruhwati et al. (2017), reservoirs are built by river damming. Reservoir ecosystems are categorized as inland public waters. Technically, inland public waters are useful for irrigation sources, electricity generation, tourism, industry, water transportation, and

aquaculture development. Ecologically, inland public waters are useful for the habitat of aquatic biota and suppliers of nutrients to the surrounding marine waters (Jummiati et al., 2021).

Sources of water pollution originate from natural and human contaminants (Rezagama & Tamlikha, 2016). The World Health Organization (WHO) predicts that approximately 80% of water pollution in developing countries is caused by domestic waste (Bouslah et al., 2017). Anthropogenic activities in the industrial, domestic and

agricultural fields produce waste containing physical, chemical and microbiological pollutants (Khouni et al., 2021). Reduction in freshwater quality caused by these contaminants is a national problem because it frequently occurs in Indonesian territorial waters (Soeprbowati et al., 2016). Cases of decreased freshwater quality examples such as domestic activities and capture fisheries cages in Lake Rawapening (Piranti et al., 2018), waste disposal in Situ Gunung Putri Bogor (Aristawidya et al., 2020), settlements and farmlands in Jatibarang Reservoir (Putrisia et al., 2022), also industrialization in Ameenpur Lake, India (Srinidhi et al., 2022).

Seloromo Reservoir is multipurpose and open so that its utilization can be carried out by various parties as long as they meet the stipulated conditions. The construction of this reservoir originally came from the Netherlands' desire to make a dam with the main function as irrigation. The Seloromo Reservoir, constructed in 1930 to dam the flow of the Sani River, is currently used for irrigating agricultural land. Since then, the reservoir has been developed as a fish farming and nature tourism destination. Unfortunately, development of population activities around the river can produce waste that has an impact on water quality. This is because the water flow from upstream to downstream passes through densely populated areas with various activities. The degradation of reservoir water quality can come from many human activities, both human activities directly on the reservoir body and river water that carries waste into reservoir waters.

Water quality is influenced by natural processes and human activities (Bouslah et al., 2017). This research refers to analyzing the physical parameters of water quality, such as temperature, pH, TDS, and EC. These parameters present critical environmental information and provide useful data for water quality control and can be used to monitor changes in water quality due to human activities (Marisi et al., 2016; Romdania et al., 2018). This research aims to analyze the factors that affect water quality in Seloromo Reservoir and determine the spatial distribution of water quality based on physical parameters of water quality. Spatial distribution approaches

are used in the water quality monitoring process to represent the distribution of sample points (Prasetyo et al., 2022). The research location is a location that has never been used for academic research so the journal is still limited. Results can support water quality management, identify and control pollution sources, and increase knowledge to protect water resources in Seloromo Reservoir.

## 2. Materials and Method

### 2.1. Study Area

Seloromo Reservoir, locally known as Gembong Reservoir, is located on the southeastern slopes of Mount Muria. It serves as an important water body within the region. The geographic setting of the study area is shown in Figure 1. Administratively, the reservoir falls within the jurisdiction of Gembong Sub-district, Pati Regency, Central Java Province, Indonesia. The study area exhibits varied topography, ranging from lowlands to elevations of up to +159.5 meters above sea level (BBWS Pemali Juana, 2019). Seloromo Reservoir covers approximately 5 km<sup>2</sup>, with a catchment area of 15 km<sup>2</sup>. Land use is dominated by residential areas, agricultural land, forests, and minor shrubland patches. The reservoir receives inflow from the Sentul, Jering, Lampeyan, Wuni, and Tempur sub-watersheds (BBWS Pemali Juana, 2019). It is utilized for agricultural irrigation, freshwater aquaculture, and recreational activities (Tourism Information Center Kabupaten Pati, 2020).

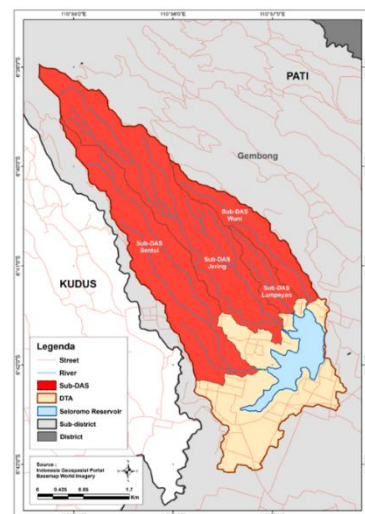


Figure 1. Map of the study area showing Seloromo Reservoir, its catchment boundary, and the major contributing sub-watersheds.



Reservoir water sources come from rainwater catchment and river flow. River flows are Inlet 1 (Wuni River), Inlet 2 (Lampeyan River), Inlet 3 (Jering River), Inlet 4 (Sentul River). Growth in population activities around the watershed can generate waste and reduce reservoir water quality. It happens because the upstream to downstream water flow moves through populated areas with various activities. Water quality degradation of reservoirs can occur due to many human activities, both human activities directly on the water body and river water that carries effluents into reservoir waters.

## 2.2. Data Collection and Sampling

Survey conducted on 11 February 2023, in the morning (09.00 a.m.) until noon (12.00 p.m). Conditions at that time had begun the dry season based on BMKG predictions indicating that the peak of the rainy season in the Eastern Pati Regency area occurred around December 2022 until January 2023 (BMKG, 2022). Primary data were collected from direct measurements (in situ) at the reservoir. These physical parameters included temperature, acidity (pH), electrical conductivity (EC), and total dissolved solid (TDS) because these parameters can be measured with a water checker directly in the field (Faisyal et al., 2017). Water pH measurements were also taken using litmus paper (Krisno et al., 2021), specifically to measure pH of water in the Ngablak Dengklek DAM to support the analysis (Figure 2).



Figure 2. Measuring water pH at Ngablak Dengklek Dam using litmus paper for pH assessment.

Sampling locations were identified using a systematic random sampling method by creating a grid with a size of 150 m × 150 m using ArcGIS software, then point locations were randomly selected within each grid (Sejati, 2017). It is recommended that one

observation point represents a grid of 2.25 ha with a mapping scale of 1:15,000 (Triadi et al., 2016). Total sampling was determined by grid results of 50 samples. Figure 3 shows the sampling locations based on the systematic random sampling method. Water samples are composite, a combination of samples collected at the water surface to a depth of 5 meters. Water samples were collected using a water sampler, then measured with a water checker.

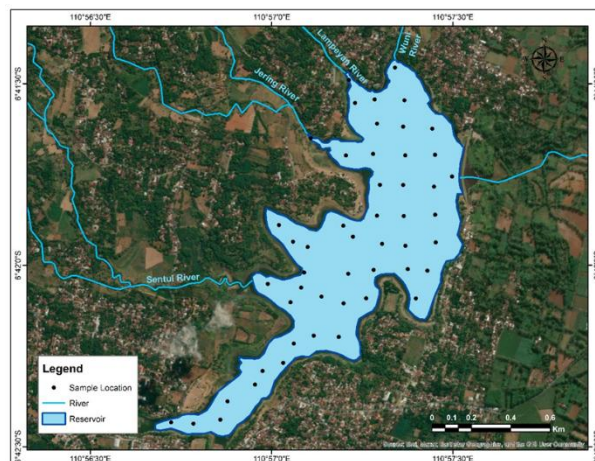


Figure 3. Map of sampling locations showing 50 sample points distributed across Seloromo Reservoir for water quality assessment.

## 2.3 Analytical Method

Reservoir water quality data were analyzed using descriptive - quantitative methods to explain the results of field measurements in order to identify factors that affect water quality in Seloromo Reservoir. Spatial analysis was also performed to present reservoir water quality data in a map created using the Inverse Distance Weighted (IDW) interpolation technique. This method is commonly used in spatial analysis (Yang et al., 2020). Maps shown are spatial distribution maps of temperature, pH, EC, and TDS parameters for a simplification of water quality analysis.

## 3. Result and Discussion

Water quality analysis was conducted on 50 samples that were evenly distributed in the water body, including the inlet and outlet points. Results of in situ water quality samples for the physical parameters of temperature, pH, DHL, and TDS at the inlet and outlet locations can be seen in Table 1.



### 3.1. Temperature

Temperature is influenced by sun light intensity which is related to the level of water depth (Alfionita et al., 2019; Koniyo, 2020). In addition, water temperature is affected by observation time, seasonal conditions, air circulation, weather cloud cover, latitude, and vegetation. Based on the inlet and outlet water temperatures (Table 1), the values were varied. According to Alfionita et al. (2019), high temperature can be due to sampling during the daytime. However, the results of this study actually disagree with this opinion because at inlet 3 with observation time at 10:45 WIB had lower temperature values than other locations with observation time about 30 minutes earlier. These results were due to the location of the temperature measurement at inlet 3 close to the trees. In contrast to inlet 1, 2, 4, and outlet

locations which have higher temperature values because sampling is taken during sunny weather and without vegetation cover.

Figure 4 shows the distribution of water temperature conditions in Seloromo Reservoir varying between 27,9°C – 29,3°C and almost dominated by the range of 28,6°C – 29,0°. The water temperature of this reservoir is included in the optimum temperature for the life of aquatic organisms with values that are close to each other. The small difference in temperature values resulted from the weather during sampling, which was rather cloudy so that the intensity of sunlight hitting the water body was lower. Moreover, measurements were taken compositely from the surface to a depth of 5 meters so the depth factor as a reason for differences in water temperature was unable to be proven.

Table 1. Physical water parameters of Seloromo Reservoir, including pH, electrical conductivity (EC), and total dissolved solids (TDS), as measured across various locations

Physical parameter	Location				
	Inlet 1	Inlet 2	Inlet 3	Inlet 4	Outlet
Local time (AM)	10.15	10.14	10.45	09.46	09.24
Temperature (°C)	29.00	29.10	28.10	29.00	28.60
Acidity/pH	8.34	9.06	8.57	8.80	8.89
TDS (mg/L)	100.00	99.80	102.00	97.00	100.00
EC (µs/cm)	139.70	140.20	143.80	137.00	140.90

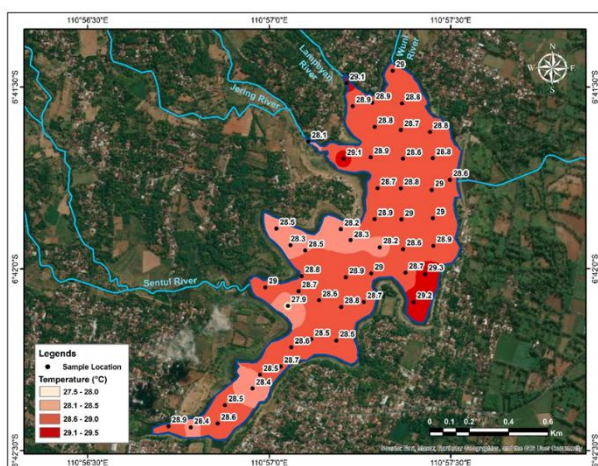


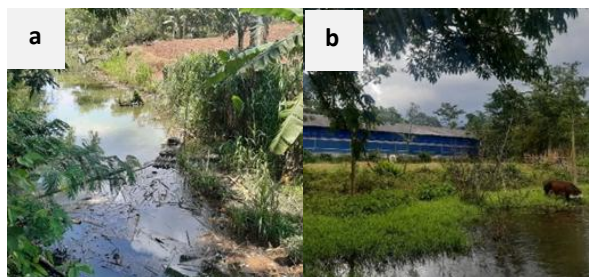
Figure 4. Spatial Distribution Map of Temperature in Seloromo Reservoir

### 3.2 Degree of Acidity/pH

Various environmental factors contribute to changes in the pH of water, both high and low. Several factors that affect pH changes include the concentration of CO<sub>2</sub> in water, chemicals,

temperature, soil and rock composition, and the decomposition process of organic materials (The Healthy Journal, 2023). According to inlet and outlet (Table 1), the pH condition of the water at inlet 1 is the lowest and lower than the outlet because of organic waste such as leaves and tree branches that fall into the water, and the possibility of animal corpses (Figure 5a).

High levels of organic matter can reduce the pH value due to the process of respiration and decomposition process of organic substances in sewage (Supriatna et al., 2020). Highest pH value is in inlet 2 due to the activities of livestock farms by citizens near inlet 2 (Figure 5b). Livestock activities produce liquid waste that causes water pollution to reservoir water quality. This condition agrees with the research of Tatangindatu et al. (2013) and Putri (2019) which proves that the sampling location near the livestock activity causes the pH value of the water to be alkaline.

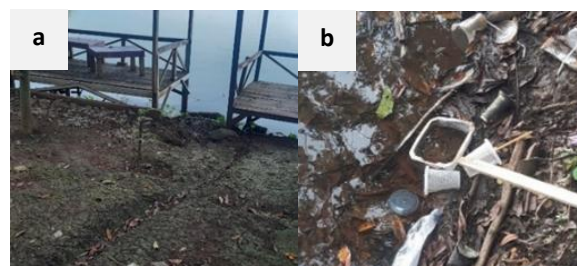


**Figure 5.** Inlet 2 conditions showing (a) organic waste accumulation and (b) effects of agricultural activities on the surrounding environment

According to the distribution of water pH conditions in Seloromo Reservoir, the values varied between 8,00 – 9,34 (Figure 7). Water pH values in the north area are more alkaline than the south area. Northern areas of the reservoir receive water supply from three inlets (Wuni, Jering, Lampeyan Rivers) that carries pollutants from the sub-watershed area. Meanwhile, southern areas of the reservoir receive water supply from one inlet, the Sentul River. Related to this, water supply from the Wuni River inlet is the largest supply that drains water from the Ngablak Dengklek dam (north of the reservoir). The simplification observation at Ngablak Dengklek Dam shows that the pH of the water is alkaline with a value of 8 (Figure 2). Land use surrounding this dam is dominated by agricultural activities. Agricultural land use in the sub-watershed area can have an impact on changes in pH of reservoir water (Saputra et al., 2020; Soeprobowati et al., 2019). Fertilizers and pesticides carried by runoff water and river flow will enter the reservoir waters. Besides, agricultural activities in the Jolong area probably supply the Wuni River.

In addition to agricultural activities, many commercial activities are also carried out in the northern area of the reservoir, specifically the periphery of the reservoir. This activity can increase the pH to alkaline through soap waste that is washed by rainwater and then enters the water body. This condition is consistent with the research of Wijayanti (2019) which states that domestic waste in the form of detergents and soaps is alkaline, it causes water to have an alkaline pH. Evidently, there are several small drains near food stalls as water drainage to the water body (Figure 6a). In addition, human waste from commercial and tourism activities is abundant around the periphery of

the reservoir (Figure 6b). Housing waste is suspected to increase the pH to alkaline (Dewi et al., 2022).



**Figure 6.** Conditions affecting the reservoir: (a) Drainage runoff entering the reservoir and (b) Contribution of housing waste to water pollution

Finally, third factor that can affect pH water is human activity around the reservoir area. Land use as a housing area around the reservoir sub-watershed is relatively large with a total area of 131,74 hectares. Based on the housing area in the Wuni, Jering and Lampeyan sub-watersheds have a total area of 67,95 hectares, which is relatively larger than the Sentul sub-watershed of 64,90 hectares. This indicates that human activities in the northern area of the reservoir are higher, so that the impact of water discharges from land use is more significant than in the southern area of the reservoir. For example, household waste such as soapy water from car/motorcycle washing and laundry businesses. Reservoir water flow conditions also support this analysis because the dominant direction flows from west to east and a small portion to the northeast. Elevations in the western side of the reservoir that are higher than the eastern side cause water to flow to lower areas. Therefore, the distribution of pH from the inlet location to the center, the pH value is increasing caused by the direction of water flow towards the outlet. It shows that sediment or waste material is spread in accordance with the distribution of water flows (Indrayana et al., 2014).

### 3.3 Total Dissolved Solid (TDS)

The TDS limit according to WHO is <1000 mg/L (Fatima et al., 2022). TDS originates from natural sources (soil erosion, leaves and silt), feces, surface rainwater, fertilizers and pesticides, as well as domestic and industrial waste (Ustaoglu & Tepe, 2019). According to Table 1, the highest TDS value was 102,0 mg/l



at inlet 3 (flow from Jering River). The presence of agricultural activities beside the river that leads to inlet 3 caused the fertilizer/pesticide solution material to be carried into the reservoir. Agricultural land in the Jering sub-watershed is also the largest at 308,65 hectares. In addition, animal feces are suspected to have entered the reservoir because of livestock activities near inlet 3. Animal feces waste carried in irrigation waterways causes an increase in nutrient content (Saputra et al., 2020).

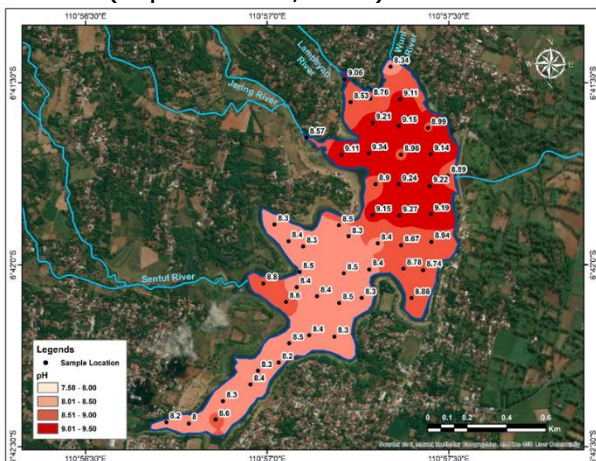


Figure 7. Spatial Distribution Map of pH in Seloromo Reservoir

TDS spatial distribution maps show higher values in the north between 98,6 – 100,0 mg/l than the south between 97,0 – 98,5 mg/l (Figure 8). The northern side has more inputs from three inlets, while the southern side has only one inlet. Northern areas get the largest supply from the Wuni River, where the surrounding land use is dominated by agricultural and forest activities. In addition, total area of agricultural and forest land use from all three sub-watersheds (Jering, Wuni, Lampeyan) is approximately 623,15 hectares, while the Sentul sub-watershed is approximately only 331 hectares. Water flow from irrigated land in the sub-watershed area can increase the concentration of dissolved substances in the river flow, so that the condition of river water quality is getting worse (Helmi et al., 2017).

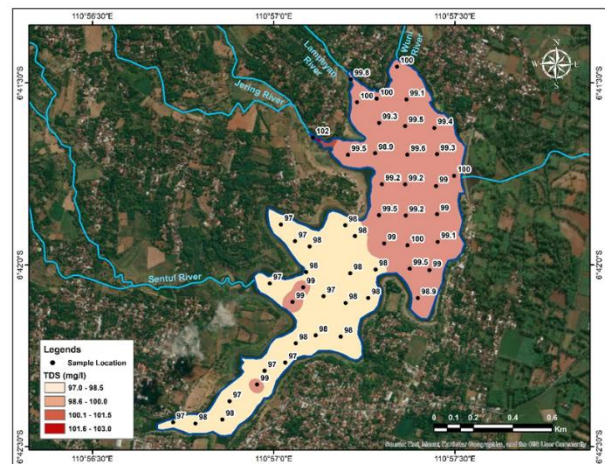


Figure 8. Spatial Distribution Map of TDS in Seloromo Reservoir

### 3.4 Electrical Conductivity (EC)

EC describe as an ability of water to conduct electrical energy, higher value means more salt content in the water. Table 1 show that the lowest EC value was found at inlet 4 (input from Wuni River). This is caused by fertilizers from agricultural activities near the inlet not being absorbed by the plants and then carried by the flow of water towards the outlet (Saputra et al., 2020). Inlet 3 (input from Jering River) are higher than outlet because of agricultural activities with fertilizers or pesticides carried in the water. Agricultural land in the Jering sub-watershed is the largest at 308,65 hectares. In addition, the housing area in the Jering sub-watershed is also relatively large at 39,08 hectares which assumes activities such as washing and bathing. Higher EC in the inlet can be caused by human activities that use soap and detergents (Saputra et al., 2020). EC ranged from 136,0 – 143,8  $\mu\text{S}/\text{cm}$  and was categorized as fresh water because it was below 1,500  $\mu\text{S}/\text{cm}$ . EC distribution pattern is similar to the TDS distribution pattern, which is higher in the northern side than the southern side of the reservoir (Figure 9). This means that the salt ion content is more in the northern side of the reservoir. TDS conditions in the northern side are higher because of the influence of agricultural activities, so it is assumed that these activities increase EC values.



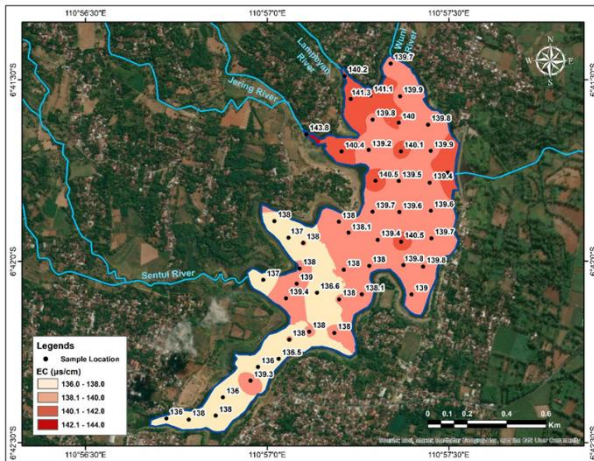


Figure 9. Spatial Distribution Map of EC in Seloromo Reservoir

Water quality conditions in Seloromo Reservoir based on physical parameters can be indicated by human activities around the reservoir area. For example, TDS conditions in the southern part of Seloromo Reservoir are lower than the northern part, in accordance with research (Tyas et al., 2021) that areas that are less contaminated by pollutants from natural activities such as erosion and activities of the surrounding community have lower TDS values.

Analysis of factors affected water quality in Seloromo Reservoir only limited to human activity factors, and not natural factors. Additionally, the parameters used are also physical parameters that can be tested directly in the field. This study neither used bathymetry maps to show the water depth and bottom conditions of the reservoir waters, nor measurements of reservoir water discharge at the inlet and outlet locations. Consequently, the analysis of water quality conditions was only based on the conditions during sampling and also observations of activities in the catchment area.

#### 4. Conclusion

This study assessed the impact of human activities around the Seloromo Reservoir catchment on water quality, focusing on physical parameters such as pH, electrical conductivity (EC), and total dissolved solids (TDS). The findings reveal that agricultural practices, human settlements, tourism, and livestock contribute to water quality variations, particularly in the northern part of the

reservoir, which is more vulnerable due to the higher concentration of these activities. This research underscores the importance of understanding the relationship between land use and water quality in freshwater ecosystems.

While the study provides valuable insights, the reliance on physical parameters limits a comprehensive analysis of water quality. Future research should expand to include more complex indicators, such as nutrient levels and microbial contamination, to better understand the ecological health of the reservoir. The findings highlight the need for effective catchment management strategies to address the impacts of human activities, ensuring the sustainability of Seloromo Reservoir for its various uses, including irrigation, aquaculture, and recreation.

#### Funding Agencies

The research was supported by personal funds.

#### Data availability statement

All data used in the research is public knowledge and there is not any confidential information

#### Acknowledgment

The author expresses his gratitude towards everyone who contributed during data collection. As well, special appreciation to BPBD Pati Regency for giving support and transportation services while collecting the data.

#### Author Contributions

The author was responsible for the conceptualization, design, and execution of the study, including data collection, analysis, and interpretation. The author also drafted and revised the manuscript for publication

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## Groundwater Potential Assessment in the Semarang-Demak Basin Using Geospatial and Multi-Criteria Analysis

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Received: 1 October 2024; Accepted: 26 December 2024; Published: 20 June 2025

**Abstract:** Groundwater is a critical resource for meeting the increasing demands of urban, agricultural, and industrial sectors. However, overexploitation and contamination pose significant threats to its sustainability. This study aims to assess the groundwater potential zones (GWPZ) within the Semarang-Demak Groundwater Basin, a rapidly urbanizing region in Central Java, Indonesia, where clean water availability faces mounting challenges. A geospatial approach integrating Geographic Information Systems (GIS), Remote Sensing (RS), and the Analytical Hierarchy Process (AHP) was employed to identify and classify GWPZ. Key factors analyzed include geology, lithology, slope, soil type, drainage density, rainfall, geomorphology, and land use/land cover. The results delineate the basin into five classes of groundwater potential zones, ranging from very poor to very good, offering actionable insights into areas optimal for groundwater recharge and extraction. Validation using groundwater table measurements from 71 infiltration wells confirmed a significant correlation between predicted zones and observed water levels. The results showed that zones with very high groundwater potential are concentrated in the central part of the basin, primarily due to favorable conditions such as high rainfall, alluvial formations, and low drainage density. These findings provide actionable insights for stakeholders to implement targeted groundwater management strategies, ensuring sustainable water resource utilization in the face of growing urbanization and environmental pressures.

**Keywords:** Analytical Hierarchy Process (AHP), Geospatial Approach, Groundwater, Groundwater Potential Zone, Multi-Criteria Approach

DOI: <https://doi.org/10.55981/limnotek.2025.8066>

### 1. Introduction

Groundwater is vital for industrial, agricultural, and residential uses, but

contamination and increasing demand threaten its sustainability (Alabdulkreem *et al.*, 2023). Groundwater extraction has significantly

increased due to population growth, urbanization, and industrialization, leading to declines in quality and quantity in many regions (Gleeson *et al.*, 2020). Overexploitation has resulted in severe depletion and quality degradation globally, affecting human populations and the environment (Jia *et al.*, 2020).

Mapping GWPZ is crucial for sustainable water resource management, enabling informed decision-making regarding groundwater development and conservation. Recent studies have used Multi-Criteria Decision Analysis (MCDA), remote sensing, and GIS methodologies to delineate GWPZ, integrating key factors such as geology, slope, rainfall, land use/land cover, soil type, linear density, and drainage density shown in Table 1. (Ahirwar *et al.*, 2021; Pande *et al.*, 2021; Razi *et al.*, 2024). The Analytical Hierarchy Process (AHP) is often used to assign weights to these parameters, aiding in classifying groundwater potential from very poor to very good (Razi *et al.*, 2024). Validation techniques, including Receiver Operating Characteristic (ROC) analysis and well data, have shown high accuracy in mapping groundwater recharge zones (Pande *et al.*, 2021). Remote sensing and GIS techniques, combined with satellite imagery such as Landsat and ASTER, enable the development of thematic maps that classify regions into different recharge potential zones (Ahirwar *et al.*, 2021). Studies of groundwater potential in urban areas, utilizing advanced geospatial techniques and AHP, have proven crucial in ensuring environmental sustainability and water security.

As one of Indonesia's major urban areas, Semarang City faces significant challenges regarding clean water availability. Rapid population growth and urbanization have increased the demand for clean water, while surface water resources are limited and sometimes unreliable in quality (Pertwi, 2017). As a result, groundwater has become a vital water source for Semarang's residents, but overuse without proper management could lead to groundwater table depletion, seawater intrusion, and contamination (Suprayogi *et al.*, 2024). The trend of urban and rural populations in Central Java from 2010 to 2023 shows a significant shift toward urbanization. The

proportion of the population living in urban areas steadily increased from 47.2% in 2010 to approximately 51.5% in 2023. This growth reflects the migration of people from rural to urban areas, likely driven by better economic opportunities, improved infrastructure, and easier access to public services in cities. Conversely, the percentage of the rural population declined from 52.8% in 2010 to around 48.5% in 2023 (BPS Central Java Province, 2019; Karra *et al.*, 2021). This decrease is due to migration and the reclassification of some rural areas into urban zones due to infrastructure development and urban sprawl.

This study aims to assess the GWPZ within the Semarang-Demak Groundwater Basin, a rapidly urbanizing region in Central Java, Indonesia, where clean water availability faces mounting challenges. By integrating thematic spatial data, the study aims to inform sustainable groundwater management strategies in Semarang City, a rapidly urbanizing area facing water resource challenges due to population growth and urbanization.

## 2. Materials and Methods

### 2.1 Study Area

This study focuses on the Semarang-Demak Groundwater Basin (Figure 1) (Rifai, 2022), which is one of the essential groundwater basins in Central Java. The basin extends from Demak Regency in the north to Semarang City in the south, covering an area of approximately 1,000 km<sup>2</sup>. The Semarang-Demak Basin is formed by alluvial deposits and folded sedimentary rocks, with varying topography, ranging from coastal lowlands to hills. This geomorphological diversity significantly influences groundwater potential in the region (Kurnianto, 2019).

Distinctive hydrogeological conditions, with an unconfined upper aquifer and a confined lower aquifer, characterize the Semarang-Demak Basin. The unconfined aquifer is near the ground surface, while the confined aquifer consists of lenses of sand and gravel covered by a layer of clay or sandy loam (Lo *et al.*, 2022). Rainfall in this region is relatively high, with annual precipitation varying from 1500 to 3000 mm (BPS Central Java Province, 2019). The

area experiences two distinct seasons: a dry season from April to September and a wet season from October to March. Heavy rainfall during the wet months contributes to groundwater recharge; however, uneven rainfall distribution can lead to significant fluctuations in groundwater levels, especially during the dry season (Lo *et al.*, 2021). Recent data from the Semarang Climatology Station for the year 2020 indicates monthly variations in precipitation, reflecting the seasonal patterns typical of the region. These hydrogeological characteristics, combined with the variability in rainfall distribution, underscore the importance of effective water resource management in the

Semarang-Demak Basin to mitigate the impacts of seasonal fluctuations on groundwater levels.

## 2.2. Geological Factors Affecting Groundwater Potential Zones

Groundwater aquifers are classified by depth: very shallow (<5 m) and shallow (5–15 m) aquifers are vulnerable to contamination but suitable for small-scale uses. Medium-depth (15–30 m) and deep (30–60 m) aquifers are more stable and suited for larger-scale supplies, while very deep aquifers (>60 m) provide clean, reliable water for municipal and industrial needs but require significant investment (Olago, 2019).

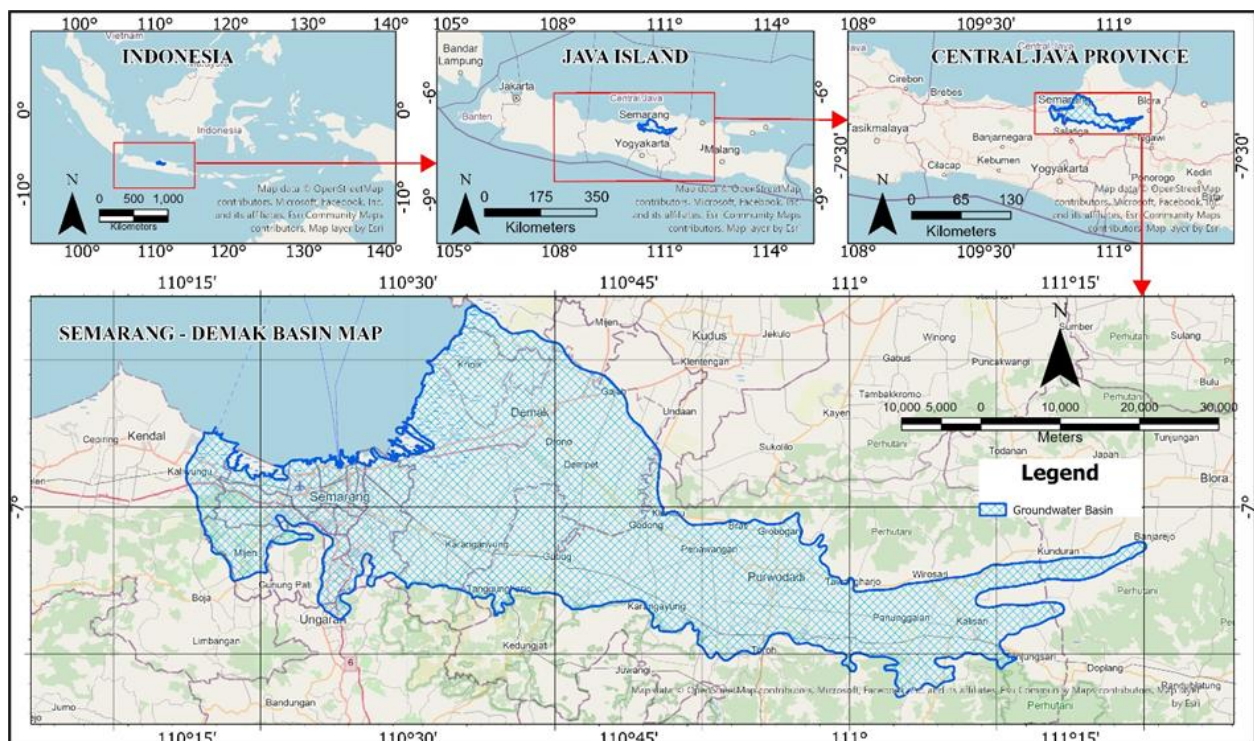


Figure 1. Map of the Study area, the Semarang-Demak Groundwater Basin, Indonesia

## 2.3. Groundwater Potential Parameters

Various geological, geomorphological, and environmental factors shape groundwater potential in the Semarang-Demak Basin. Alluvial formations exhibit the highest potential due to their porous and permeable nature, allowing significant groundwater storage and movement (Ardaneswari *et al.*, 2016; Kurnianto, 2019; Ridha & Darminto, 2016). Volcanic rocks, such as dense andesite, have lower potential because of limited permeability. The Kalibeng and Tambakromo formations demonstrate moderate potential, depending on

sediment composition and stratigraphy. Other formations like Tuban and Mundu rely on porous layers to enhance groundwater potential (Figure 2a).

Geomorphology significantly affects groundwater potential (Figure 2b). Alluvial plain's extensive aquifer has the highest potential, while eroded mountainous regions exhibit poor potential due to topsoil loss and reduced infiltration capacity (Ardaneswari *et al.*, 2016). Regarding lithology (Figure 2c), unconsolidated sediments such as gravel, sand, and clay contain significant groundwater



potential thanks to their high porosity. Consolidated sediments offer varying potential (Trihatmoko *et al.*, 2020), especially if fractures or more permeable materials are present. The base data for the soil classification in Fig 2d of present study has been obtained from Center for Research and Development of Agricultural Land Resources, Indonesia. The result of soil classification found that, the study area has three types of major soils such as gleisol, kambisol, mediteran, podsolik, regosol and

other. Soil type is another critical factor (Pratama *et al.*, 2018), with highly permeable regosol soils offering excellent water recharge, while saturated or acidic soils, like gleisol and podsolic, hinder infiltration.

Rainfall plays a vital role in recharging groundwater (Figure 2e). High-rainfall areas generally have excellent potential, primarily when supported by suitable geological and soil conditions (Kurniawan *et al.*, 2023).

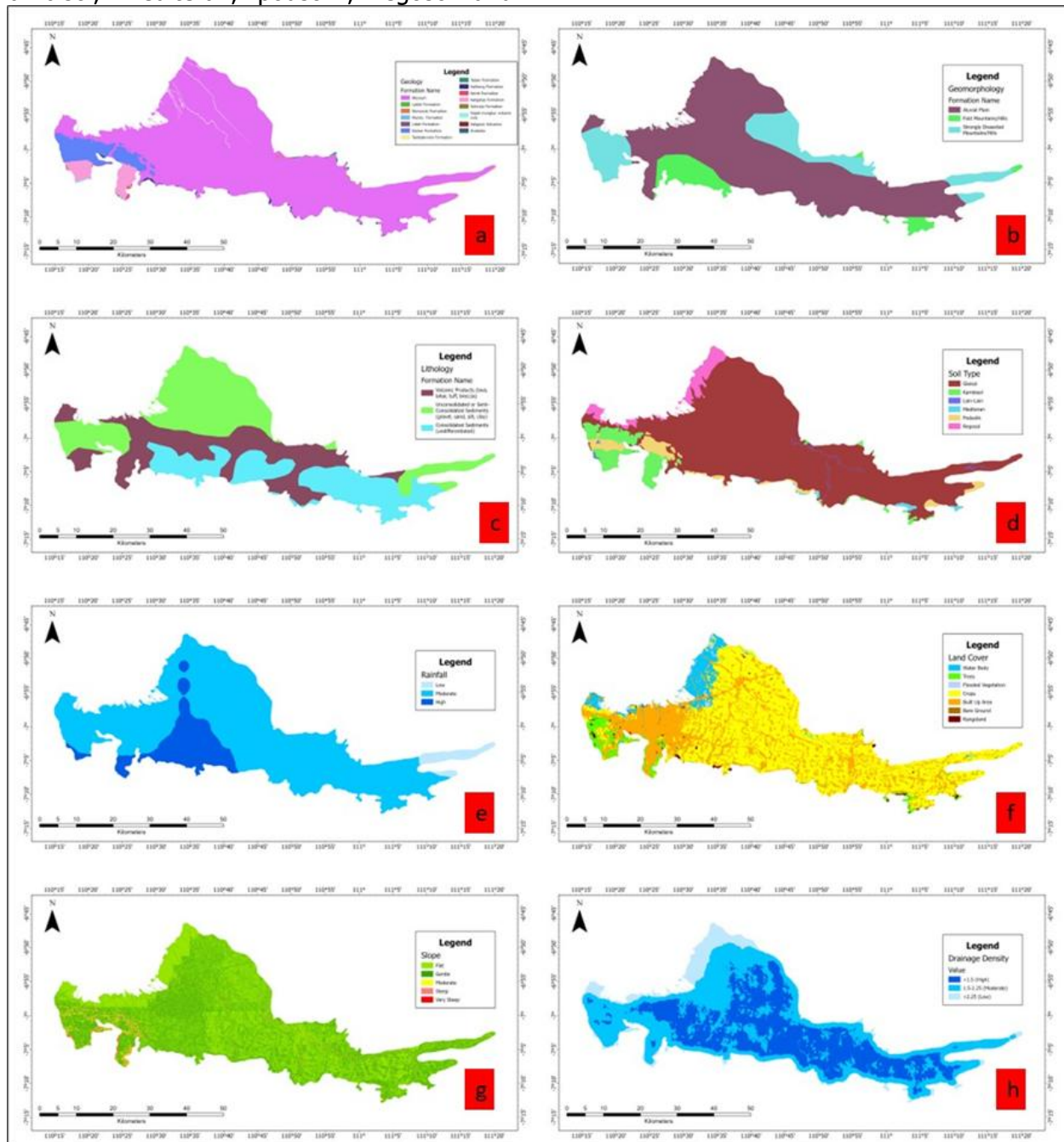


Figure 2. Potential Groundwater Parameter Map of the Semarang-Demak Groundwater Basin (GWB): (a) Geology; (b) Geomorphology; (c) Lithology; (d) Soil Type; (e) Rainfall; (f) Land Cover; (g) Slope Gradient; (h) Drainage Density.

The Sentinel 2 satellite image has been used for the study to find out the land cover of study area. The supervised classification method has been used with level – 2A classification. The result of the study found the study area covered by seven different classes such as water body, trees, flooded area, crops, built-up area, bareground and rangeland (Figure 2f) (Karra *et al.*, 2021). Forested and vegetated areas offer moderate to high potential due to better infiltration, while urbanized or impervious surfaces limit groundwater recharge (Fakhrudin & Daruati, 2017). The slope gradient further determines recharge potential (Figure 2g), with flat to gentle slopes enabling more water percolation into the ground, whereas steep slopes reduce infiltration due to rapid runoff (Darwis & Sc, 2018).

The drainage system of the study area is shown in Fig. 2h. The drainage pattern was extracted directly from DEMNAS (8x8m). Then, the coordinates of the center of each grid were used to prepare the surface drainage density map by IDW interpolation technique (Rahmati *et al.*, 2015). Regions with low drainage density provide better infiltration opportunities, as slower water flow allows for more excellent percolation.

Eight factors were analyzed using GIS to map GWPZ, including slope, drainage density, land cover, rainfall, soil type, lithology, geomorphology, and geological formations (Table 1). These spatial data layers were integrated to identify zones with varying groundwater recharge and storage capacities, aiding in sustainable water resource management.

Table 1. Geospatial Data in this Research

No.	Data	Format	Scale/cell size	Source
1.	DEMNAS	Raster	Cell Size 8 m	Geospatial Information Agency
2.	Geology	Tabular/GIS	1: 50.000	Ministry of Energy and Mineral Resources (ESDM)
3.	Geomorphology	Vector (Polygon)	1: 50.000	Ministry of Energy and Mineral Resources
4.	Litology	Vector (Polygon)	1: 50.000	Ministry of Energy and Mineral Resources
5.	Soil Type	Vector (Polygon)	1: 50.000	Center for Research and Development of Agricultural Land Resources
6.	Rainfall	Tabular/GIS	1: 50.000	Central Java Statistics Agency
7.	Land Cover	Raster	Cell Size 10 m	Sentinel-2
8.	Drainage Density	Raster	Cell Size 8 m	Geospatial Information Agency

## 2.4. Methods

This study employs a geospatial approach that relies on compiling Remote Sensing data (Widyaningrum *et al.*, 2021) and GIS to prepare GWPZ maps (Danso & Ma, 2023). In predicting GWPZ, AHP can be used to integrate multiple parameters that influence groundwater availability and potential (Arulbalaji *et al.*, 2019). Eight controlling parameters were used

to delineate the GWPZ: geology, slope gradient, drainage density, rainfall, lithology, soil type, geomorphology, and land cover. The spatial analysis results of these parameters were tested using the AHP method with normalized weights (Riyandi *et al.*, 2019) to evaluate GWPZ in the Semarang-Demak Groundwater Basin (GWB) area (Figure 3).

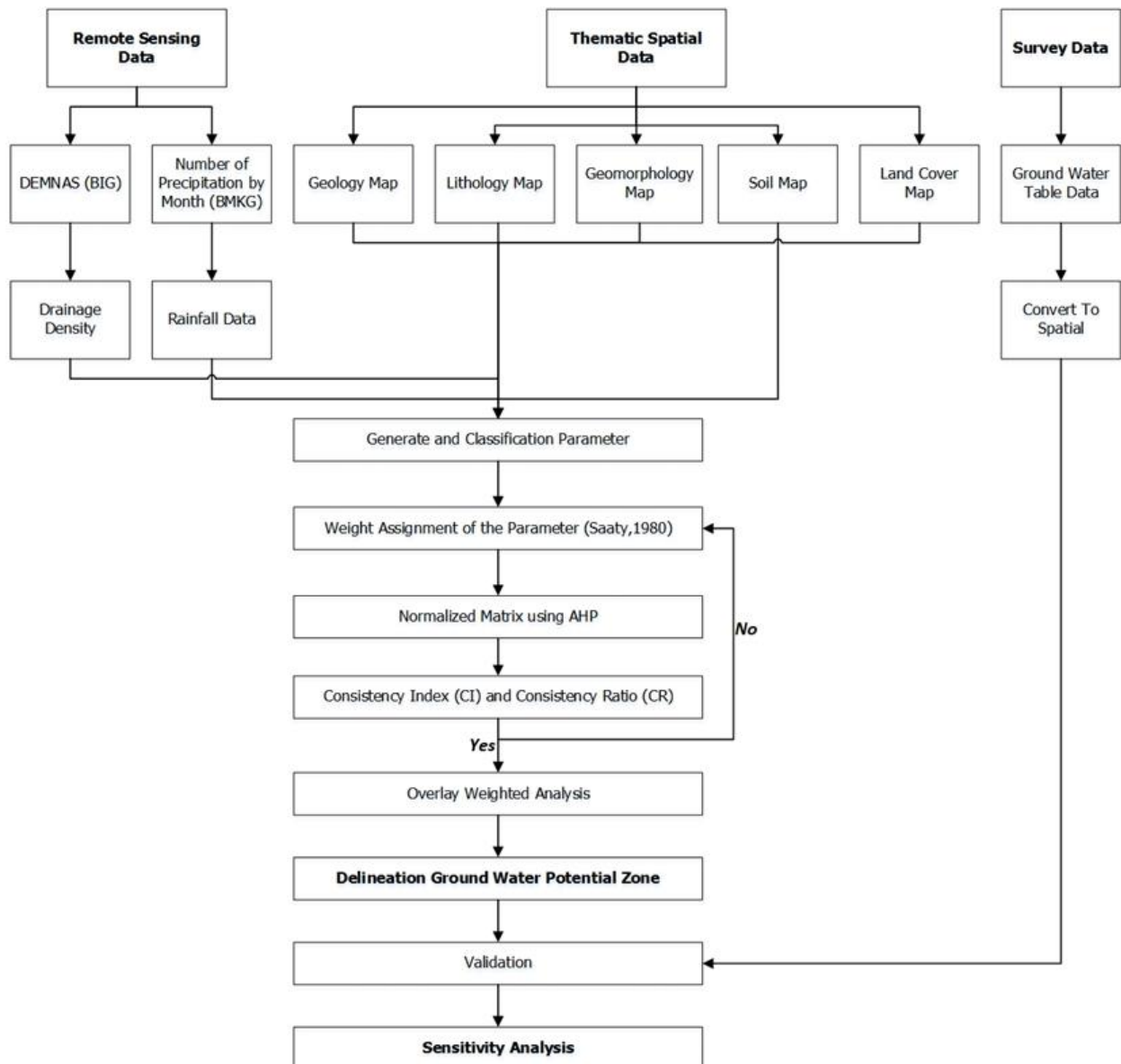


Figure 2. Flowchart diagram of the Groundwater Potential Zone Identification in the Semarang-Demak Groundwater Basin, Central Jawa, Indonesia

The groundwater potential index (GWPI) is a dimensionless index value that expresses the potential of groundwater (equal 1) in a specific area. It can be calculated using the equation below:

$$GWPI = \sum_{w=1}^m \sum_{j=1}^n (W_j \times X_i) \quad \dots (1)$$

where  $W_j$  is the normalized weight of the  $j$ -th parameter,  $X_i$  refers to the class weight of the  $i$ -th parameter,  $m$  represents the number of parameters, and  $n$  represents the number of classes for a specific parameter. For each grid,

the GWPI (equation 2) is calculated using the following equation:

$$GWPI = GeaGeb + LiaLib + GeoaGeob + SlaSlb + RaaRab + LCaLCb + StaStb + DDaDDb \quad \dots (2)$$

where  $Ge$ =Geology;  $Li$ =Lithology;  $Geo$ =Geomorphology;  $Sl$ =Slope;  $Ra$ =Rainfall;  $LC$ = Landcover;  $St$ =Soil Type;  $DD$ =Drainage Density. The parameter weight is denoted by "a," while the subclass weight is denoted by "b."



In this study, the AHP was employed to determine the relative importance of various factors influencing groundwater potential, assigning weights to each parameter (Saaty, 2004). Scoring was then applied to classify these parameters into categories based on their characteristics, with higher scores indicating greater groundwater potential. For instance, flat slopes, low drainage density, permeable soils, and high rainfall were scored higher due to their positive contributions to groundwater recharge (Ridha & Darminto, 2016); these scores range from 1 (low potential) to 5 (high potential), ensuring consistent evaluation across parameters. These weights and scores were integrated into a spatial analysis using the Weighted Overlay tool in ArcGIS Pro. This tool combines reclassified spatial data layers according to their scores and weights, derived from AHP, to produce a comprehensive groundwater potential map (Tešić *et al.*, 2020). Parameters such as geomorphology, lithology, soil type, rainfall, slope, drainage density, and land cover were analyzed in detail to capture

their individual and combined impacts (Suja Rose & Krishnan, 2009; Ramu, 2014). The result is a visually intuitive and data-driven representation of groundwater potential zones, offering valuable insights for resource management and environmental planning (Ardiyanto *et al.*, 2022).

### 3. Result

#### 3.1. Variable Weighting

##### 3.1.1. Pairwise Comparison Matrix

Priorities were assigned on a scale of one to eight points for each pair of layers. For example, lithology is considered more important than rainfall for groundwater potential and was thus assigned a value of 2 (Table 2).

The eigenvector expresses the order of influence of the layers on groundwater potential. The normalized primary eigenvector (NPE) in this study was calculated using an Excel sheet created by the researchers (Table 2).

Table 2. Pairwise Comparison Matrix for AHP-based GWPZ

	Ge	Li	Geo	Sl	Ra	LC	St	DD	NPE
Ge	1	1	1 1/4	3	2	4	5	5	22.80%
Li	1	1	1	3	2	4	5	5	22.24%
Geo	4/5	1	1	2	1 1/4	3 2/3	4 1/2	4 1/2	18.50%
Sl	1/3	1/3	1/2	1	1	2 2/5	3 2/3	3 2/3	10.96%
Ra	1/2	1/2	4/5	1	1	1 5/9	3 2/3	4	12.18%
LC	1/4	1/4	1/4	2/5	2/3	1	1 1/4	2 2/5	5.84%
St	1/5	1/5	2/9	1/4	1/4	4/5	1	1 5/9	4.10%
DD	1/5	1/5	2/9	1/4	1/4	2/5	2/3	1	3.38%
<b>Total</b>	4.28	4.48	5.24	10.90	8.42	17.82	24.75	27.12	100%

Ge = Geology; Li = Lithology; Geo = Geomorphology; Sl = Slope; Ra = Rainfall; LC = Landcover; St = Soil Type; DD = Drainage Density; NPE = Normalized Principal Eigenvector

##### 3.1.2. Matrix Consistency Value

This study calculated the consistency index (CI) value of 0.02 based on the eigenvector value of 8.14 (Table 3). The random index (RI) value for eight parameters in this study is 1.41 (Table 4). Therefore, the consistency ratio (CR) is calculated as follows:

$$CR = \frac{0.02}{1.41} = 0.0138 = 1.38\%$$

Since the resulting CR value is 1.38% (less than 10%), the process can proceed to the weighted overlay, integrating all parameters to produce the groundwater potential map for the Semarang-Demak Groundwater Basin.

Table 3. Results of Eigenvector Value Calculation ( $\lambda_{\max}$ )

	<b>Column Sums (Row 9 of Table 2)</b>	<b>Eigenvector (Row 10 of Table 2)</b>	<b>Parameter Rank (1) x (2)</b>
	<b>(1)</b>	<b>(2)</b>	<b>(3)</b>
Ge	4.28	0.23	0.98
Li	4.48	0.22	1.00
Geo	5.24	0.19	0.97
Sl	10.90	0.11	1.19
Ra	8.42	0.12	1.03
LC	17.82	0.06	1.04
St	24.75	0.04	1.01
DD	27.12	0.03	0.92
Sum ( $\lambda_{\max}$ )			8.14

Ge = Geology; Li = Lithology; Geo = Geomorphology; Sl = Slope;  
Ra = Rainfall; LC = Landcover; St = Soil Type; DD = Drainage Density

Table 4. Index Ratio (RI) for Scores of n Parameters

<b>N</b>	<b>3</b>	<b>4</b>	<b>5</b>	<b>6</b>	<b>7</b>	<b>8</b>	<b>9</b>	<b>10</b>
RI	0.58	0.89	1.12	1.24	1.32	1.41	1.45	1.49

### 3.1.3. Parameter Weighting Values

The variable weighting analysis using AHP yielded the following weights for each variable: geology scored 22.80, lithology scored 22.20, and geomorphology scored 18.50. The slope was weighted at 11, rainfall at 12.20, soil type at 4.10, and drainage density at 3.40. These results indicate that each variable significantly contributes to understanding groundwater potential in the study area. Geology provides critical insights into the subsurface structure that influences the distribution and types of aquifers.

Lithology is critical in determining porosity and permeability, which affect groundwater storage capacity. Geomorphology helps determine landforms and surface features that influence water flow and infiltration. Combining these parameters allows for identifying areas with the highest groundwater potential, which is the primary goal of this research (Table 5).

Table 5. Parameter Classification for Identifying GWP

<b>Factor (Unit)</b>	<b>Class</b>	<b>Parameter Weight</b>	<b>Class Rank</b>
Geology	Andesite	22.80%	1
	Kaligesic Volcanics		1
	Gajah mungkur volcanic rock		1
	Tuban Formation		2
	Wonocolo Formation		2
	Mundu Formation		2
	Lidah Formation		2
	Selorejo Formation		2
	Kaligetas Formation		2
	Damar Formation		2
	Tambakromo Formation		3
	Kerek Formation		3
	Ledok Formation		3
	Kalibeng Formation		4
	Aluvial		5

Factor (Unit)	Class	Parameter Weight	Class Rank
Lithology	Unconsolidated or Semi-consolidated Sediments (Gravel, Sand, Silt, Clay)	22.20%	1
	Consolidated Sediments (undifferentiated)		3
	volcanic eruption (lava, lahars, tuff, breccia)		5
Geomorphology	Strongly Eroded Mountains/Hills	18.50%	1
	Folded Mountains/Hills		3
	Alluvial Plain (Dataran Aluvial)		5
Slope	Flat (0-3.98)	11.00%	1
	Gentle (2.98-8.37)		2
	Moderate (8.37-14.95)		3
	Steep (14.95-24.32)		4
	Very Steep (>24.32)		5
Rainfall	Low (< 1900 mm /years)	12.20%	1
	Moderate (1900-2000 mm /years)		3
	High (>2000 mm /years)		5
Landcover	Built Up Area	5.80%	1
	Bare Ground		2
	Rangeland		3
	Crops		4
	Trees		5
	Flooded Vegetation		5
	Water Body		5
Soil Type	Podsolik	4.10%	1
	Mediteran		2
	Gleisol		3
	Kambisol		4
	Regosol		5
Drainage Density	Low (0-250m)	3.40%	1
	Moderate (250-500m)		3
	High (more than 500m)		5

### 3.2. Groundwater Potential Zones

This study shows the distribution of area based on the Groundwater Potential Zone (GWPZ) classification in a specific region, with a total area of 185,479.3 hectares. Of the total area, the "Very Poor" zone encompasses 1,084.665 hectares, equivalent to approximately 0.6% of the total area, indicating that only a small portion of this area has very low groundwater potential. Furthermore, the "Poor" zone, with an area of 11,074.16 hectares, accounts for about 6.0%, suggesting that although it is larger, water resources still have limited potential. The "Moderate" classification dominates with an area of 34,899.25 hectares, which makes up

approximately 18.8%, reflecting better conditions for groundwater management.

The "Good" zone has the largest area at 107,214.8 hectares, contributing approximately 57.8% of the total, indicating a significant potential for water resource availability. Lastly, the "Very Good" zone, covering 31,206.4 hectares, accounts for around 16.8%, showing the presence of areas with very good quality for groundwater. Thus, the GWPZ area distribution provides a clear picture of groundwater potential spread in the region, highlighting the need for resource management focus on areas with better potential.



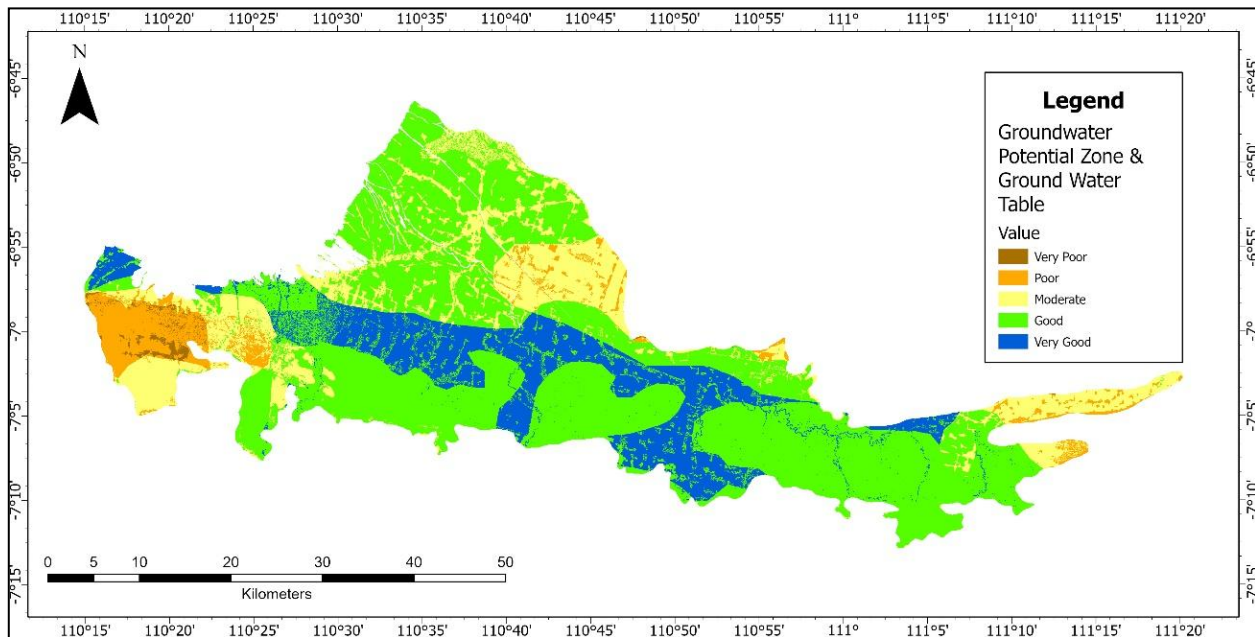


Figure 4. Groundwater Potential Zone Map of Semarang-Demak Groundwater Basin

Table 6. Groundwater Potential Zone Areas in Semarang-Demak Groundwater Basin

No.	Class Zone GWPZ	Area (Ha)
1.	Very Poor	1,084.665
2.	Poor	11,074.160
3.	Moderate	34,899.250
4.	Good	107,214.800
5.	Very Good	31,206.400
Total		185,479.300

### 3.3. Validation of Groundwater Potential Zone Identification

This study demonstrates the distribution of area based on the GWPZ classification in a specific region, totaling 185,479.3 hectares. Within this area, the "Very Poor" zone encompasses 1,084.665 hectares, approximately 0.6% of the total, indicating a minimal portion with very low groundwater potential. The "Poor" zone, covering 11,074.16 hectares, accounts for about 6.0%, suggesting limited water resource potential despite its larger area. The "Moderate" classification occupies 34,899.25 hectares, representing roughly 18.8%, indicating better conditions for groundwater management. The largest area is in the "Good" zone at 107,214.8 hectares, contributing approximately 57.8% of the total,

highlighting significant potential for water resource availability. Lastly, the "Very Good" zone, at 31,206.4 hectares, accounts for around 16.8%, reflecting very high groundwater quality areas. Thus, the GWPZ area distribution provides a clear understanding of the groundwater potential in the region, emphasizing the need for focused resource management in higher potential areas.<sup>7</sup>

The accuracy analysis between Groundwater Level and Groundwater Potential indicates a relatively good level of congruence, with a percentage reaching approximately 73.24%. Out of 72 locations analyzed, 52 show alignment between groundwater level and potential categories based on validation results. However, it is worth noting that among the measurement points examined, 63% exhibited low or very low groundwater potential, 30% fell within the moderate category, and only 7% showed good potential. While there is high congruence, particularly at locations classified as "Good" and "Moderate," where shallow groundwater levels correlate with good potential, several locations categorized as "Poor" and "Very Poor" demonstrate less optimal alignment.

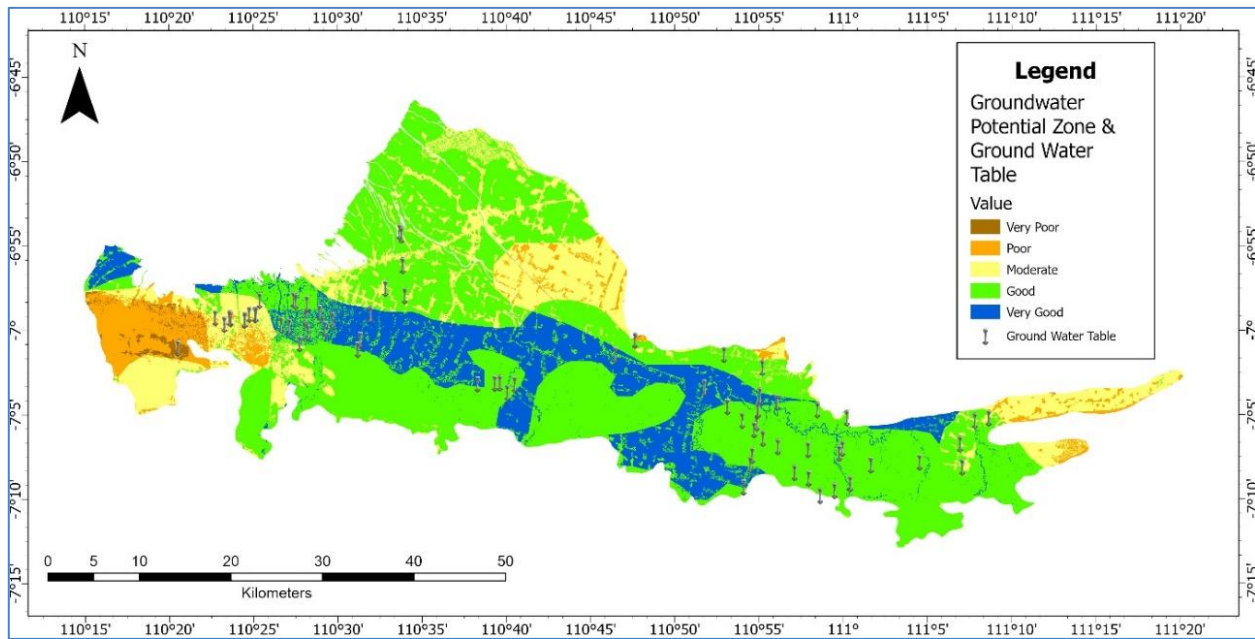


Figure 5. Overlay Groundwater Table on Groundwater Potential Zone Map of Semarang-Demak Groundwater Basin

Table 7. Validation of Groundwater Potential Zone Identification with Water Table Data

Groundwater Potential	Groundwater Level	Average depth (m)	Number of Locations
Very Poor	Very Deep	-84.91	12
Poor	Deep	-25.57	10
Poor	Shallow	-3.81	15
Moderate	Deep	-39.46	14
Moderate	Shallow	-1.49	23
Good	Shallow	-0.45	12
Good	Very Deep	-44.75	2

#### 4. Discussion

High-potential groundwater zones were primarily concentrated in the central and southwest regions, where favorable conditions such as unconsolidated sediments, high rainfall, and low slopes were prevalent. This finding aligns with the observations of Achu *et al.* (2020), who identified lithology, lineament density, and geomorphology as critical factors influencing groundwater recharge and occurrence. Furthermore, it supports the findings of Kawara *et al.* (2024), who emphasized the importance of lineament density, slope, rainfall, and drainage density in their GWPZ analyses.

The study underscores the interplay of geological, hydrological, and anthropogenic factors in shaping groundwater potential. The

central zones have very good potential and provide essential recharge areas critical for sustaining groundwater resources in Semarang and Demak. These insights can guide policymakers and stakeholders in prioritizing conservation efforts and optimizing groundwater extraction to mitigate overexploitation risks. Additionally, identifying low-potential zones highlights areas where alternative water supply strategies may be necessary, such as rainwater harvesting or managed aquifer recharge.

While the study effectively combines GIS, remote sensing, and AHP, some limitations exist. First, the accuracy of the GWPZ map depends on the resolution and quality of spatial data, which may not capture micro-level variations. Second, the validation covered only

71 locations, which might not fully represent the basin's heterogeneity. Future research could enhance validation by incorporating more extensive field data and employing advanced machine learning algorithms for GWPZ prediction.

#### 4. Conclusion

This study highlights the importance of mapping Groundwater Potential Zones (GWPZ) for sustainable water resource management in rapidly urbanizing regions like the Semarang-Demak Groundwater Basin. By employing advanced geospatial techniques and the AHP, the study integrates key parameters such as geology, geomorphology, slope, and rainfall to delineate areas with varying groundwater potential, with a total area of 185,479.3 hectares. The "Very Poor" zone covers 1,084.665 hectares (0.6% of the total area), indicating minimal groundwater potential. The "Poor" zone, spanning 11,074.16 hectares (6.0%), also has limited potential. The "Moderate" zone, comprising 34,899.25 hectares (18.8%), reflects improved conditions for groundwater management. The "Good" zone, the largest at 107,214.8 hectares (57.8%), demonstrates significant groundwater potential, while the "Very Good" zone, at 31,206.4 hectares (16.8%), highlights areas of excellent groundwater quality.

The results show that zones with very high groundwater potential are concentrated in the central part of the basin, primarily due to favorable conditions such as high rainfall, alluvial formations, and low drainage density. Conversely, low and very low groundwater potential zones are found in the south and eastern regions, characterized by less favorable geological and topographical factors. Validation using groundwater table data from infiltration wells confirms the reliability of the GWPZ map, with significant compatibility observed in many locations. These findings provide actionable insights for stakeholders to implement targeted groundwater management strategies, ensuring sustainable water resource utilization in the face of growing urbanization and environmental pressures. Future research should incorporate dynamic parameters such as climate variability and human-induced land use

changes to refine the models further and enhance their predictive capabilities.

#### Data availability statement

The data used in this research consists of primary and secondary data. The primary data were collected from field measurements conducted by the Agency for Energy and Mineral Resources (ESDM), Central Java Province, in 2023. The secondary data were obtained from official Indonesian government institutions, including the Ministry of Energy and Mineral Resources, Geospatial Information Agency (BIG), and the Indonesian Geological Agency. The rainfall data were sourced from CHIRPS, which can be accessed via the website [chc.ucsb.edu](http://chc.ucsb.edu).

#### Funding Agencies

This research was not funded by any external parties.

#### Author Contribution

**RAo** conducted the investigation, formal analysis of the literature review and preparation of the manuscript, **DR, BHS, A, EGAS, WH, RP** were involved in conceptualization as well as reviewed the manuscript. All the authors read and approved the final manuscript.

#### Acknowledgment

The authors thank the Agency for Energy and Mineral Resources (ESDM), the Geospatial Information Agency (BIG), and Center for Research and Development of Agricultural Land Resources (BBSLPD) for providing data.

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## Bioremediation of Phenolic Pollutants by Fungi: A Perspective

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Received: 2 October 2024; Accepted: 26 December 2024; Published: 20 June 2025

**Abstract:** Phenol is a priority pollutant that poses a significant risk to human health and ecological systems when released into aquatic environments. Consequently, numerous technologies have been developed and implemented to remove phenol from wastewater. These technologies can be classified into physical, chemical, and biological techniques. While conventional treatment methods can effectively remove phenol, some are more economical and less environmentally beneficial. This overview, which is based on a collation of relevant and comprehensive literatures, emphasizes various phenolic pollutants in wastewater and how mycoremediation can be implemented to address these issues. Mycoremediation research has been chiefly directed on investigating the effects of various conditions on phenol degradation and evaluating its effectiveness under controlled experiments. Moreover, mycoremediation enables a doable solution for mitigating pollution, improving water quality, and supporting biodiversity in aquatic ecosystems. These also mean that advancing mycoremediation encourages environmentally sustainable practice. However, the remaining gaps exist in current research including the toxicity assessment of degradation by-products, the application of synthetic biology methods for chassis modification, creation and development of innovative immobilization methods, improvement of remediation efficiency by integration of multiple technologies and scalability of mycoremediation for practical wastewater treatments. These areas warrant further research to advance the greater potential of mycoremediation.

**Keywords:** Fungi, mycoremediation, phenol, wastewater treatment

DOI: <https://doi.org/10.55981/limnotek.2025.8074>

### 1. Introduction

Water pollution has become a significant global concern due to several factors, including population growth, industrial expansion, urbanization, rising water usage, and agricultural practices. These issues have led to environmental degradation and contamination, adversely affecting water bodies, human health, and ecology. Phenolic compounds are a notable type of organic contaminants that significantly affect water quality due to their high toxicity and carcinogenic characteristics (Bibi *et al.*, 2023; Liu *et al.*, 2024). Phenol and its derivatives are among the most extensively

utilized organic compounds. Simple phenolic molecules serve as intermediaries in the synthesis of specific polyphenolic secondary metabolites. They are also used as starting points for the industrial synthesis of numerous other chemical compounds. Consequently, industrial effluent from manufacturing organic chemicals, oil refining, and olive processing contains phenol and its derivatives (Wu *et al.*, 2022).

Persistent pollutants, such as phenols, are resistant to degradation by physical, chemical, or biological means (Mohd, 2022). Because of this, phenol has been listed as one of 129



priority pollutants by the US EPA and the Canadian National Pollutant Release Inventory (NPRI) that must be remedied before being released into the environment (EPA, 2014). Approximately 10 million tons of phenolic compounds are released into the environment annually by agrochemicals, leather, textiles, petrochemicals, and pharmaceuticals industries (Alshabib & Onaizi, 2019). In Indonesia, for instance, phenolic compound was detected around 0.013 ng/L in the tributary of Bengawan Solo River (Khoiriyah *et al.*, 2019). Moreover, industrial processes such as making paint, paper, pulp, and pesticides are believed to release phenolic chemicals into the environment (Alshabib & Onaizi, 2019). The percentage of phenolic compounds in industrial effluents can vary from 1 mg/L to 7000 mg/L (Mohd, 2022; Bibi *et al.*, 2023).

The discharge of untreated phenolic wastewater into the environment can cause significant health issues and pollute soil, surface water, and groundwater, disrupting the natural environment equilibrium (Anku *et al.*, 2017). Moreover, it is common for phenolic wastewater to seep into the ground and contaminate surrounding lakes, rivers, water reservoirs, and agricultural areas (Panigrahy *et al.*, 2022). As a result, the EPA sets a criterion for water filtration at fewer than 1 part per billion (ppb) for phenol in surface waters. This decision complies with the standards established by the European Union.

However, the permissible discharge limits for phenolic compounds are 0,5 mg/L for surface waters and 1,0 mg/L for effluents from sewage treatment systems as specified by Italy government, law no. 152/2006 (Mohd, 2022). The Indonesian government has established permissible phenol content limits for river water quality standards, ranging from 0,002 to 0,02 mg/L depending on the intended usage (Law No. 22/2021). Phenolic compounds, even at low concentrations, can adversely affect aquatic biota, and in drinking water, they can cause unpleasant tastes and odors at level as low as 5 g/L (Panigrahy *et al.*, 2022). Thus, proper and effective treatment of phenolic wastewater is crucial before reuse or discharge (Alshabib & Onaizi, 2019).

Environmental exposure to phenol has a significant negative influence on human health

as well as ecological systems. Even at modest levels, phenol can be lethal to aquatic life (Rittmann & McCarty, 2001). The human body can quickly absorb phenol through the skin, diet, and respiratory systems. Hanafee *et al.* (2019) stated that phenol exposure can result in a variety of health issues, including catastrophic skin damage, eye irritation, severe gastrointestinal problems, cardiovascular disorders, and, in the worst cases, death. Therefore, urgent intervention is required to mitigate the risks of phenol exposure and to remediate phenolic effluents in ecosystems.

A variety of physical and chemical approaches, including distillation, membrane separation, chemical and electrochemical oxidation, ozonation, advanced oxidation, and photocatalysis, have been suggested in treating phenolic wastewater (Wu *et al.*, 2022; Bibi *et al.*, 2023). Effective removal phenolic contaminants rely on the selection of suitable membrane and adsorbent materials. However, these materials often exhibit significant losses and inadequate regeneration (Crini *et al.*, 2019; Dotto & McKay, 2020). The high chemical requirements of complex oxidation processes result in considerable running costs, even though they are frequently highly efficient (Tuan Tran *et al.*, 2022). Some treatment technologies merely transfer pollutants from the water to another medium, potentially causing secondary pollution (Hodges *et al.*, 2018). Limitations of physical and chemical wastewater treatment methods have driven the development of advanced, cost-effective, and sustainable technologies to reduce their environmental impact (Mehdi *et al.*, 2021; Wu *et al.*, 2022).

Mycoremediation is an environmental decontamination technique that utilizes fungi as a means of remediation. Fungi, alongside bacteria, are known for their diversity and remarkable capacity to decompose phenolic compounds. Unlike bacteria, fungi can thrive in ecologically challenging conditions, including environments with limited nutrient availability, reduced water activity, and low pH levels, where bacterial growth may be insufficient (Ibrahim & Al-Ghamdi, 2019). Fungi possess the capability to utilize phenol as a carbon source for their development, demonstrating remarkable adaptability to various ecosystems

and the ability to thrive in extreme conditions (Hanafee *et al.*, 2019). This review describes the various types of phenolic pollutants present in aquatic ecosystems, their environmental impacts, and the utilization of fungal-based techniques for the remediation of phenol in these environments.

## 2. Methodology

This review employs a literature review methodology to explore the presence of phenolic pollutants in aquatic ecosystems, their environmental impacts, and the application of fungal-based remediation techniques. Relevant studies were sourced from reputable scientific databases such as ScienceDirect, Web of Science, and SpringerLink. Search terms included combinations of keywords such as mycoremediation, biodegradation of phenol, phenol environmental impact, and fungi enzymatic pathways. The inclusion criteria focused on peer-reviewed journal articles published in English that addressed phenolic pollution in aquatic environments and discussed fungal-based remediation approaches. Studies unrelated to aquatic ecosystems, phenolic pollutants, or fungi-based techniques were excluded. Extracted data encompassed the types and sources of phenolic pollutants, their ecological effects, and the specific fungal mechanisms used for phenol degradation. The review synthesizes findings into three primary themes: the type of phenolic pollutants, the environmental and ecological impacts, and the potential of fungal-based bioremediation as a sustainable solution. This review focuses on highlighting the advantages of fungi in bioremediation while recognizing, but not extensively addressing, challenges and limitations such as scalability and economic viability.

## 3. Result and Discussion

### 3.1. Types of phenolic pollutants

Phenolic compounds are introduced into aquatic environments through various pathways and can be detected in multiple environmental matrices, including surface water, seawater, and riverbed sediments. Common phenolic compound found in wastewater include simple phenols (phenol and cresols), chlorophenols (2,4-dichlorophenol

and pentachlorophenol), nitrophenol (2-4 dinitrophenol and 4-nitrophenol), and bisphenol

#### 3.1.1. Simple Phenols

Simple phenols consist of a benzene ring bonded to a hydroxyl group (OH) and commonly present in significant concentrations in industrial wastewater, particularly from refineries. In some cases, phenol concentration in wastewater can reach levels as high as 10 g/L (Wu *et al.*, 2022).

- a. Phenol (C<sub>6</sub>H<sub>5</sub>OH): found in wastewater from the textile and pharmaceutical industries, phenol poses a threat to aquatic ecosystems, leading to reduced biodiversity (El-Naeb *et al.*, 2022; Ahmaruzzaman *et al.*, 2024).
- b. Cresols (C<sub>7</sub>H<sub>8</sub>O): These methylated phenols, used in wood preservatives and disinfectants, exist in three isomeric forms (o-cresol, m-cresol, p-cresol). They are harmful to both soil and water environments (Gucbilmez, 2022).

#### 3.1.2. Chlorinated Phenols

Chlorinated phenols are phenol derivatives with one or more chlorine atoms covalently bonded. This compound are typically found in lake and river sediments, tannery waste and sewage sludge (Wu *et al.*, 2022).

- a. 2,4-Dichlorophenol (C<sub>6</sub>H<sub>4</sub>Cl<sub>2</sub>O): Commonly used in pesticide production, this persistent pollutant adversely impacts both aquatic and terrestrial organisms (Gucbilmez, 2022).
- b. Pentachlorophenol (C<sub>6</sub>Cl<sub>5</sub>OH): Utilized in wood preservation and as a pesticide, pentachlorophenol is highly toxic to aquatic life and contributes to long-term soil and groundwater contamination (Kahru *et al.*, 2002).

#### 3.1.3. Nitrophenols

Nitrophenols are used in agricultural and industrial processes and are linked to respiratory and hematological health issues in humans. They also contribute to groundwater pollution (Maletta *et al.*, 2023)

- a. 2,4-Dinitrophenol (C<sub>6</sub>H<sub>4</sub>N<sub>2</sub>O<sub>5</sub>): used in the manufacture of explosives and pesticides, this compound is toxic to aquatic life and humans and posing a significant risk of environmental risk if released into water bodies (Gucbilmez, 2022).

b. 4-Nitrophenol (C<sub>6</sub>H<sub>5</sub>NO<sub>3</sub>): A by-product of industrial activities such as pesticide production, it harms aquatic organisms and accumulates in the environment (Kahru *et al.*, 2002).

#### 3.1.4. Bisphenols

Bisphenol A (BPA, C<sub>15</sub>H<sub>16</sub>O<sub>2</sub>), widely used in manufacturing plastics and epoxy resins, is recognized as an endocrine disruptor capable of interfering with hormonal functions in wildlife and humans. It contaminates water sources frequently (Kahru *et al.*, 2002; Gucbilmez, 2022).

### 3.2. Environmental Impacts

#### 3.2.1. Aquatic Ecosystems

The highwater solubility of phenolic chemicals facilitates their contamination of

aquatic habitats, thereby reducing biodiversity. These substances interfere with the growth and reproduction of aquatic organisms, primarily affecting algae, fish, and microbes. For example, phenol concentration ranging from 9 to 25 mg/L can be lethal to fish (Gucbilmez, 2022). When phenolic chemicals are present at level exceeding safe thresholds, they can alter phytoplankton diversity significantly and leading to ecological disruption (El-Naeb *et al.*, 2022). Furthermore, phenolic chemicals can disrupt the physiological functions of aquatic species, resulting in increased mortality rates and genotoxic effects (Gad & Saad, 2008). Phenolic pollutants also affect biodiversity by altering microbial populations and degrading water quality (Saratale *et al.*, 2020).

Table 1. Guidelines at both national and international levels regarding the presence of phenolic compounds in water

Compounds	National Water Quality Standards (mg/L)				Regulation of Drinking Water (mg/L)		
	Indonesia Government Regulation No. 22 of 2021				WHO	EPA 2023	EC 2020
	Class I <sup>a</sup>	Class II <sup>b</sup>	Class III <sup>c</sup>	Class IV <sup>d</sup>	Water for Human Consumption		
Phenol	0.002	0.005	0.01	0.02	0.001		0,001
Pentachlorophenol					0.009	0.001	
Dinitrophenol						0.007	0.001
2,4,6- trichlorophenol					0.2		0,025
Bisphenol							0.003

a= Water for raw drinking water and other applications that necessitate a specific level of water quality.

b= water for infrastructure and facilities, including recreational activities, freshwater fish cultivation, animal husbandry, crop irrigation, and other applications necessitating similar water quality standards.

c= water for cultivating freshwater fish, livestock management, irrigation of crops, and other applications that require identical water quality standards.

d= water for irrigation of crops and other applications that require a similar quality of water.

#### 3.2.2. Soil Pollution

Pentachlorophenol is a chlorinated phenol that adheres to soil particles and persists in the environment for extended periods while inhibiting microbial growth. It reduces soil fertility and hinders plant development. Furthermore, leaching from contaminated soils poses a threat to groundwater quality (Kahru *et al.*, 2002).

#### 3.2.3. The Effect of Bioaccumulation on the Food Chain

Many phenolic contaminants tend to bioaccumulate in organisms, transferring up the food chain and enhancing their harmful effects. This bioaccumulation can lead to long-term health risk for higher organisms, including

reproductive and developmental disorders both in humans and wildlife (Gad & Saad, 2008).

#### 3.2.4. Human Health Risk

Numerous phenolic derivatives are recognized for their -high toxicity and their tendency to produce hazardous by-products during water treatment, such as polychlorinated dibenzo-n-dioxins (Taneeva *et al.*, 2024). Humans' exposure to phenolic chemicals can result in severe health complication. Endocrine disruptors such as BPA are associated with developmental delays, cancer, and reproductive issues. Chronic exposure can also damage the kidneys and liver (Kahru *et al.*, 2002).

Chlorination of raw water increases the risks of exposure to phenolic chemicals. The



production of chlorophenols in drinking water has been primarily attributed to the chlorination process (WHO, 2003). More stringent regulations are necessary because the elevated global levels indicate that current treatment methods are insufficient to completely eliminate phenolic compounds from water, posing significant risks to public health.

Water potability recommendations provide acceptable values for water quality standards intended for human consumption. These regulations, which are currently not sufficiently stringent regarding emerging toxins such as phenolic compounds, must be enforced by public water systems (Ladeia Ramos *et al.*, 2024). Table 1 lists the phenolic compounds that have reached the Maximum Permitted Concentration (MPC) for surface and drinking water, as specified in the national guidelines of the Indonesia government (Law No. 22/2021) and relevant international standards.

### 3.3. Fungi-based strategies

Phenol compounds are organic pollutants that degrade water quality and belong to the class of aromatic organic substances with the molecular formula  $C_6H_5OH$ . These compounds can be produced naturally by various organisms or released into the environment as raw effluent by multiple industries. When water containing chlorine reacts with phenolic chemicals, it forms complexes with unpleasant tastes and odors. The substitution of chlorine enhances these undesirable characteristics with harmful consequences (Almasi *et al.*, 2019). Many phenolic compounds are dangerous due to their carcinogenic, mutagenic, teratogenic, and toxic properties. They can also disrupt the endocrine system. Even at low concentrations, they have a significant negative impact on aquatic ecosystems and human health (Alshabib & Onaizi, 2019; Singh *et al.*, 2021). Due to their persistent and resistance to degradation, phenolic contaminants represent the major environment risk and treatment challenge.

Table 2. Removal of phenolic pollutants by fungi and associated mechanisms

Fungal strain	Periods	Compound	Removal (%)	Mechanism	Reference
<i>Pleurotus ostreatus</i> ( <i>P. ostreatus</i> )	10 days	p-chlorophenol (CP)	99.2	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
		phenol	98.7		
	8 days	Nonylphenol	70.0	Biotransformation & Biosorption	(Pezzella <i>et al.</i> , 2017)
	5 weeks	Bisphenol A phenol	65.0 90.0	Enzymatic oxidation	(Ntougias <i>et al.</i> , 2015)
<i>P. dryinus</i> <i>Trametes hirsuta</i> ( <i>T. hirsuta</i> )	30 days	chlorophenols,	>95.0	Biotransformation	(Ariste <i>et al.</i> , 2019)
	10 days	p-chlorophenol (CP)	99.0	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
	30 days	phenol chlorophenols,	99.7 >80.0		
<i>T. versicolor</i>	8 days	Nonylphenol	85.0	Biotransformation & Biosorption	(Pezzella <i>et al.</i> , 2017)
		Bisphenol A	100.0		
<i>Tricoderma atroviride</i>	8 days	Phenol	92.0	Lignocellulolytic enzymes	(Kumar Vaidyanathan <i>et al.</i> , 2022)
<i>Cadophora sp.</i>	10 days	2, 4-dinitrophenol	91.0		
		p-chlorophenol (CP)	73.0	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
<i>Phanerochaete chrysosporium</i>		phenol	91.2		
	10 days	p-chlorophenol (CP)	99.2	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
		phenol	99.2		
<i>Pseudogymnoascus sp</i>	8 days	Nonylphenol	80.0	Biotransformation & Biosorption	(Pezzella <i>et al.</i> , 2017)
		Bisphenol A	60.0		
<i>Aspergillus caesiellus</i> ( <i>A. caesiellus</i> )		p-chlorophenol (CP)	91.0	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
		phenol	97.1		
		phenol	89.0	Biodegradation & Adsorption	(Batista-García <i>et al.</i> , 2017)
<i>A. awamori</i>	7-8 days	phenol	92.2	Degradation	(Stoilova <i>et al.</i> , 2006)
<i>A. biennis</i>	5 weeks	phenol	85.0 >80	Enzymatic oxidation	(Ntougias <i>et al.</i> , 2015)

Mycoremediation, a fungi-based treatment strategy, offers considerable potential for addressing phenolic. Fungi are well-known for their ability to degrade aromatic xenobiotics through their highly effective enzymatic biodegradation processes on wide range structural diverse of contaminants. Both live and dead cells, as well as their enzymes, have been studied for treating wastewater contaminated with phenolic compounds. Fungal species, including Ascomycetes, *Aspergillus fumigatus*, *Debaryomyces*, *Aspergillus niger*, dark septate endophyte fungi (DSE), and various Basidiomycetes have been studied for their ability in phenol degradation (Melati *et al.*, 2021, 2023; Mtibaa *et al.*, 2020; Jiang *et al.*, 2017; Tebbouche *et al.*, 2016; Kües, 2015). Other fungi and their phenol reduction mechanism are summarized in Table 2.

Various fungal strains from diverse taxonomic groups, have shown the ability to eliminate, break down, and completely degrade phenols in liquid environments, primarily through co-metabolism. Fungi employ several mechanisms for phenol removal, including sorption, oxidative and reductive dechlorination, conjugation, ring cleavage, mineralization, and polymerization (Tomasini & Leon-santiesteban, 2019).

Table 2 shows that white rot fungi (WRF) groups such as *Pleurotus* sp., *Trametes* sp., and *Phaenorocytes* sp. are commonly reported as phenol degraders with high removal efficiency. Similarly, filamentous fungi such as *Aspergillus* sp. are frequently studied for phenol mycoremediation.

However, several factors, including pH, temperature, oxygen availability, substrate concentration, fungal species, and immobilization methods, influence the efficiency of mycoremediation in phenolic compounds removal from aqueous solution. Understanding these parameters is crucial to enhancing fungal growth, enzyme activity, and elimination of phenolic pollutants.

#### a. The pH of the Medium

The pH levels significantly impact fungi growth and the activity of ligninolytic enzymes responsible for phenol degradation by. For examples, *Aspergillus niger* and *Trametes versicolor* thrive in neutral pH settings (6-7.2)

(Siva Kumar *et al.*, 2009; Supriya & Neehar, 2014). This neutral pH is optimal for the activity of the ligninolytic enzymes such as laccases, manganese peroxidases, and lignin peroxidases (LiPs). Conversely, extreme pH level can denature enzymes and inhibit fungal metabolic processes. Additionally, under such condition, the positive charges on amino groups in fungal cell wall, allow them to act as potential absorbers of phenolic compounds (Siva Kumar *et al.*, 2009). These findings underscore the importance of maintaining appropriate pH levels in bioremediation processes.

#### b. Temperature

Temperature strongly affects fungal phenol degradation, with optimal range varying by species. Studies indicate that temperature is essential in improving the effectiveness of phenol degradation processes. It is necessary to regulate the fungal enzyme synthesis and the metabolism (Sivasubramanian & Namasivayam, 2015). Fungi associated with phenol cleanup often exhibit their most efficient degradation rates between 25°C and 35°C. Elevated or decreased deviations from this range may cause denaturation of the enzyme or inhibit fungus growth. For instance, *Aspergillus niger* inhibits peak degradation efficiency at 35°C, while a significant reduction is observed below 25°C or above 40°C (Supriya & Neehar, 2014). Similarly, *Magnusiomyces capitatus* QWD1 and *Penicillium janthinellum* N12 P6C3 show optimal performance at 35 °C (Hanafee *et al.*, 2019; Wang *et al.*, 2019). In contrast, *Aspergillus flavus* and *Aspergillus nomius* SGFA1 achieve maximum phenol degradation at 25 and 28. 1 °C, respectively (Zanin *et al.*, 2014; Liu *et al.*, 2023).

#### c. Oxygen Availability

Optimal oxygen availability for phenol degradation depends on the fungal strain and environment conditions. Research has demonstrated that different types of fungi have distinct preferences for oxygen concentrations, which are essential to their ability to degrade phenol compounds. Aerobic environment generally enhance phenol breakdown, as demonstrated by *Candida tropicalis* SDP-1, which effectively degraded 1200 mg/L of phenol within 40 hours under optimal condition

of pH 8, 35°C, and agitation (Gong *et al.*, 2021). Similarly, *Debaryomyces* species exhibit efficient phenol degradation at pH 6.0 and 200 rpm agitation (Jiang *et al.*, 2016). Furthermore, *Magnusiomyces capitatus* QWD1 demonstrated superior performance in activated sludge environments, suggesting that oxygen levels can optimize degradation process (Wang *et al.*, 2019). These findings highlight the need for tailored oxygenation strategies in fungal bioremediation.

#### d. Phenol Concentration

High concentrations of phenol can inhibit fungal growth due to its toxic effect on cellular membranes and metabolic processes. While certain fungi tolerate moderate phenol concentrations, their degradation efficiency diminishes at elevated levels. Stoilova *et al.* (2007) investigated the biodegradation of phenols and their derivatives by *Aspergillus awamori* at various concentrations (0.3, 0.6, 1.2, and 3 g/L) and found that the fungus's ability to degrade phenol decrease significantly at concentration above 0.6 g/L. Conversely, fungal strains such as *Graphium* sp. and *Phanerochaete chrysosporium* effectively degrade phenol at concentrations of 0.3 and 0.05 g/L, respectively, (Kennes & Lema, 1994; Santos *et al.*, 2003;). Additionally, endophytic fungi demonstrated phenol tolerance and degradation at concentrations as high as 0.8% (Khalil *et al.*, 2021).

#### e. Fungal Species and Enzymes Involved

Different fungal species and strains exhibit varying levels of phenol tolerance and degradation capabilities, largely due to their enzymatic systems. Among various fungal species, Basidiomycetes have emerged as promising candidates for phenolic mycoremediation due to their ability to degrade a wide range of persistent aromatic pollutants. This ability is attributed to the secretion of intracellular enzymes, such as cytochrome P450 monooxygenases, and the activity of external peroxidases, including laccase, manganese peroxidase, and lignin peroxidase (Yan *et al.*, 2017; Mtibaa *et al.*, 2020). The enhanced capability of Basidiomycota for phenol biodegradation is largely due to their production of a diverse range of phenol-

degrading enzymes, such as laccases, tyrosinases, and various forms of manganese and lignin peroxidases (Kües, 2015). Furthermore, Ascomycetous fungi surpass white-rot fungi (WRF) in pollutant degradation due to their ability to survive under low oxygen concentrations, acidic pH, or ligninolytic substrates, condition that often influence enzymatic activity (Aranda, 2016; Mtibaa *et al.*, 2020). The degradation of endocrine-disrupting chemicals (EDCs) has also been shown to require the presence of cytochrome P450 enzymes, which act as crucial oxidizing agents (Nowak *et al.*, 2019; Mtibaa *et al.*, 2020). Additionally, the biodegradation of 2,4-dichlorophenol (2,4-DCP), nonylphenols (NPs), and 4-tert-octylphenol (OP) was assessed using the Ascomycetous fungus *Thielavia* sp. HJ22. This strain efficiently degraded 100%, 95%, and 80% of 4-tert-OP, NPs, and 2,4-DCP in less than eight hours, respectively. Cytochrome P450 monooxygenase and laccase enzyme from Ascomycetous fungi play essential roles in breaking down these phenolic pollutants (Mtibaa *et al.*, 2020).

Numerous fungi enzymatic pathways that aid in the conversion of phenolic chemicals into less hazardous forms are the main component of the fungal enzymes' process of phenol degradation. Important enzymes that use various methods to accomplish degradation, such as phenol hydroxylases, catechol dioxygenases, laccase and peroxidase are essential to this process. Phenol hydroxylase catalyzes the first hydroxylation of phenol to catechol (van Schie & Young, 2000). Then, catechol dioxygenases break down catechol via ortho or meta routes, producing non-toxic byproducts. For example, catechol 2,3-dioxygenase is used by *Nocardia hydrocarbonoxydans* for meta cleavage (Shetty & Shetty, 2016). Additionally, phenol hydroxylases can also hydroxylate catechol to produce pyrogallol. Further, phenol hydroxylase activity can be inhibited by high phenol concentrations, but catechol dioxygenase showed no effect (Radziff *et al.*, 2021). The peroxxygenase mechanism in certain fungi, such those that produce hemoglobin dehaloperoxidase, involves the Fe=O center abstracting hydrogen atoms, which aids in the

breakdown of chlorophenols (Zhang *et al.*, 2021).

Laccases efficiently detoxify phenols by catalyzing the oxidation of phenolic compounds through electron transfer processes and promote the breakdown of harmful phenolic compounds (Shanmugapriya *et al.*, 2019; Li *et al.*, 2024). Laccases are found in many fungi and are especially good at breaking down phenolic contaminants and lignin (Shanmugapriya *et al.*, 2019). Two important peroxidases enzymes that use hydrogen peroxide to oxidize phenolic compounds are lignin peroxidase (LiP) and manganese peroxidase (MnP) (Terrazas-Siles *et al.*, 2005; Dashtban *et al.*, 2010). The LiP mechanism involved oxidative cleavage, electron transfer and substrate versatility. LiP catalyzes the oxidative cleavage of C-C and C-O-C bonds in aromatic non-phenolic compounds, which are generally more resistant to degradation (Wong, 2009). According to Higuchi (2004) the enzyme oxidizes lignin directly at the protein surface by long-range electron transfer. LiP's usefulness in detoxification procedures is increased by its ability to function on a range of substrates, including those with high redox potential (Wang *et al.*, 2018). While MnP involved manganese cycling and catechol-mediated mechanism. The manganese cycle is facilitated by MnP's oxidation of Mn(II) to Mn(III), which also oxidizes mediators that aid in the breakdown of non-phenolic substrates (Wong, 2009). A catechol-mediated cycle is involved in the early phases of phenol degradation by MnP, which results in the production of several oxidation products such as benzoquinone and hydroquinone (Xu *et al.*, 2017)

#### *f. Immobilization Techniques*

Immobilized materials are of great interest in the biodegradation of phenolic pollutants due to their ability to prevent the formation of harmful by-products. Phenol biodegradation was assessed using *Debaryomyces* species encapsulated in calcium alginate beads and nanoscale Fe<sub>3</sub>O<sub>4</sub>. The results indicated that approximately 900 mg/L of phenol was degraded within 80 hours, with removal efficiency of over 99.9%. Furthermore, it was demonstrated that the encapsulated *Debaryomyces* species in the calcium alginate

beads could be reused for up to 10 cycles (Jiang *et al.*, 2017). To enhance the efficacy of mycoremediation when applied to naturally occurring wastewater, the addition of co-substrate is often required. However, this approach is typically viewed unfavorably due to the associated increased cost (Ariste *et al.*, 2019).

#### **4. Conclusion**

The present review describes the types of phenol, their impact on various environment and a fungi-based treatment. Various types of phenols are found in wastewater, including simple phenols, chlorophenols, nitrophenol, and bisphenol. They are poisonous, dangerous, endocrine disrupting, mutagenic, teratogenic, and/or carcinogenic, and seriously harm the environment. Fungi can effectively remove dangerous phenolic chemicals from wastewater. As an eco-friendly and sustainable approach, it offers a viable solution for mitigating pollution, improving water quality, and supporting biodiversity in aquatic ecosystems. Mycoremediation technology provide a more economical, sustainable, and environmentally friendly alternative to physico-chemical methods for wastewater treatment. In recent years, research has primarily focused on analyzing the effects of various conditions on the phenol degradation and evaluating mycoremediation's effectiveness under controlled experiment environments. Effort have also been directed toward optimizing the degradation processes to enhance fungal performance. Apart from being able to remove phenolic pollutant, the integration of mycoremediation into management practices can promote water sustainability on a global scale.

However, significant gaps remain in the literature, particularly regarding the toxicity assessment of degradation by-products, the application of synthetic biology methods for chassis modification, development of innovative immobilization methods, creation of innovative immobilization methods, investigation of improving remediation efficiency by integrating fungal bioremediation with other technologies, including chemical oxidation, biochar, phytoremediation or phytoremediation and scalability of



mycoremediation for practical wastewater treatments. These areas warrant further research to advance the field.

### Funding Agencies

Financial support from the Rumah Program 4 (RP 4) initiated by Biological and Environmental Research Organizations, BRIN in 2024.

### Conflict of interests

The authors affirm no recognized financial conflicts of interest or personal affiliations that might have seemingly impacted the research presented in this paper.

### Author Contribution

**Irma Melati** conducted the investigation, formal analysis of the literature review and preparation of the manuscript. **Miratul Maghfiroh**, **Nurul Setiadewi**, **Riky Kurniawan**, **Annisa Indah Pratiwi**, and **Rosidah** were involved in conceptualization as well as preparation and reviewed the manuscript. All the authors read and approved the final manuscript

### Acknowledgment

We acknowledge the Research Center for Limnology and Water Resources-Research Organization of Earth and Maritime-BRIN for providing the facility. RP 4, ORHL, BRIN 2024 for facilitating this research

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## **Mercury Dynamics in Mining-Adjacent Ecosystems: Risk Assessment of Lake Lais, Central Kalimantan, Indonesia**

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Received: 15 October 2024; Accepted: 26 April 2025; Published: 20 June 2025

**Abstract:** Mercury is a hazardous chemical that significantly impacts both the environment and human health. In Central Kalimantan, gold mining activities contribute to mercury contamination, particularly in aquatic ecosystems. Lake Lais, an oxbow lake along the Kahayan River, is potentially affected by mercury from upstream mining activities. This study aims to assess mercury contamination in Lake Lais water and sediment, and evaluate the ecological risks associated with mercury pollution. In September 2024, water and sediment samples were collected from five sites in Lais Lake. Water samples were collected from the surface using polypropylene bottles with added nitric acid to preserve mercury content. Sediment samples were taken using an Ekman grab and analyzed for mercury using Atomic Absorption Spectrophotometry (AAS) at *Balai Standarisasi Pelayanan Jasa Industri* in Banjarbaru, South Kalimantan. The results showed that water mercury concentrations ranged from 0.00 to 0.002 mg/l, which is within the limits set by the Indonesian government (PP No. 22/2021). However, sediment mercury levels ranged from 0.14 to 0.47 mg/kg, which can negatively impact the ecosystem based on international standards. Lais Lake was classified as having mild to moderate mercury contamination, with an ecological risk ranging from moderate to very strong. The Risk Quotient (RQ) exceeded 1, indicating high ecological risk. These findings highlight the need for regular monitoring and stricter regulation of mercury use in mining. Future research should focus on long-term impacts and seasonal variations in mercury levels to better assess ecological risks.

**Keywords:** Index Geoaccumulation (I-Geo), Mercury Contamination, Risk Ecology (RI), Risk of Quotient (RQ), Gold Mining

DOI: <https://doi.org/10.55981/limnotek.2025.8184>

### **1. Introduction**

Mercury or mercury (Hg) is a type of metal as an organic and inorganic compound. This type of metal is found in nature and is widely distributed in rocks, ore, soil, water, and air. Mercury (Hg) is liquid, grey in colour and odourless. At room temperature, mercury has liquid properties and has high electrical conductivity, which causes mercury to be widely used in industry, mining and laboratories. Mercury is very dangerous for both human life and other aquatic biota in

waters containing mercury (Marsyalita et al., 2012). In general, mercury enters the waters in the form of elemental Hg (HgO) and has a high density. This element can be converted into organic mercury caused by the action of methane bacteria, namely methyl mercury. This element has toxic properties and strong binding properties, as well as good solubility in the body of fish and other aquatic biota (Elvince et al., 2008).

In Central Kalimantan, mercury has been using in gold processing since the 2000s.

Mercury left over from gold processing can enter the environment from several processes, namely the disposal of liquid waste that still contains mercury, tailings and smoke from the gold refining process. All of these processes can enter the surrounding environment and the aquatic environment. Mercury waste that enters the waters can settle into the sediments at the bottom of the waters and then mercury is converted into the very dangerous methylmercury. Methyl mercury is known to be one of the most dangerous types of mercury in fish. If fish containing methylmercury is consumed by humans, human health may be compromised. The presence of mercury in the aquatic environment can pose risks that can cause harm. Risk of ecology related to mercury have been conducted by some researchers such as Guo, et al (2010), Mulyaningsih, T.R, and Suprapti, S. (2015), Abdullah et al (2020), Kho, et al. (2022), Handayani, et al (2024)

Mercury contamination of river waters can occur in several ways, one of which is the extraction of gold from illegal gold mines located along river banks. Liquid mercury is used in gold extraction to form amalgam. Amalgam is then melted through a combustion process releasing mercury in gaseous form into the atmosphere. Mercury released into the atmosphere will be reabsorbed through various media by plants, animals and humans causing health problems (Nakazawa et al., 2021). Hg (0) emitted to the atmosphere is oxidized, trapped by the rain, and deposited in environmental waters; Hg is also discharged directly into environmental waters. These processes may increase Hg concentrations in aquatic organisms such as fish, potentially increasing the risk of Hg intake among people who eat fish on a daily basis. (Nakazawa, et al., 2016)

According to the US EPA (2024), ecological risk assessments can be used to predict the likelihood of future impacts (prospective); or to evaluate the likelihood that observed effects are due to past or ongoing exposure to specific stressors (retrospective). In addition, ecological risk assessments are used to support various types of actions, including regulation of hazardous waste disposal sites, industrial chemicals, and pesticides, watershed management; protection of ecosystems from

chemical, physical, or biological stressors. Information from ecological risk assessments can be used by risk managers to communicate with interested parties and the general public; limit exposure to ecological stressors; negotiate remediation options with stakeholders; or develop monitoring plans to ensure risk reduction and ecosystem restoration.

Some research related to ecological risk of mercury in Indonesia has been conducted by some researchers (Maulana, et.al., 2023; Mulyaningsih and Suprapti, 2015; Nugrayani, et al., 2023; Astuti, et al., 2023)

Lake Lais is one of the lakes located in Tanjung Sangalang Village, Kahayan Tengah Sub-district, Pulang Pisau Regency, Central Kalimantan. Lais Lake is utilized by the community as a fishing ground area. As a source of protein, fish is one of the sources of mercury entry into the human body. This study aims to assess the ecological risk of mercury (Hg) concentrations in water and sediment in Lake Lais.

## **2. Materials and Method**

### **2.1. Sampling Location**

This research was conducted in Lake Lais on 03 September 2024. The lake located in Tanjung Sangalang Village, Kahayan Tengah District, Pulang Pisau Regency, Central Kalimantan Tengah (Figure 1). The lake is one of the oxbow lakes in Central Kalimantan with an area of 5,4 ha.

### **2.2. Research Procedure**

Water and sediment sampling was carried out at each station (5 stations) by following procedures:

- i. Water samples were taken from each station at the surface of the water using a 50 ml polypropylene bottle at each station. Then the water samples taken were added with nitric acid (HNO<sub>3</sub>) to stabilize the mercury concentration in the samples and put in a box filled with ice before the samples sent to the Balai Standarisasi dan Pelayanan Jasa Industri, Banjarbaru, South Kalimantan.
- ii. Sediment samples were collected using an Ekman Grab at each sampling station and the sediment samples were put into a 50 ml polypropylene bottle that had been prepared and labelled, then put into

Styrofoam box before the samples were sent to the Balai Standarisasi dan Pelayanan Jasa Industri, Banjarbaru, South Kalimantan.

- iii. Mercury analysis for both samples were analysis using Atomic Absorption Spectrometry (AAS) method.

### 2.3. Data analysis

#### *Pollutant accumulation index*

Index of Geographic Accumulation (I-Geo) is used to assess mercury pollution of Lake Lais. In order to obtain the I-Geo value or the level of mercury pollution, the formula is used as follows (Nugraha et al., 2022).

$$\text{I-Geo} = \text{Log}_2 (\text{Cn}/1.5 \text{ Bn}) \quad \dots \text{Eq.1}$$

where:

Cn: Heavy metal concentration in sediment

Bn: Normal concentration of metal (background) in nature 0.08 mg/kg (Panggabean et al., 2022).

Table 1. Classification of Geoaccumulation Index

I-Geo Range	Pollution Class
I-Geo ≤ 0	Unpolluted
0 < I-Geo ≤ 1	Unpolluted to moderately polluted
1 < I-Geo ≤ 2	Moderately polluted
2 < I-Geo ≤ 3	Moderately to heavily polluted
3 < I-Geo ≤ 4	Heavily polluted
4 < I-Geo ≤ 5	Heavily to extremely polluted
I-Geo > 5	Extremely polluted

The CF was calculated by dividing the metal concentration in the sediment by the background value (Huang, et al., 2023)

$$\text{CF} = \text{C (contaminants)/C (background)} = \text{Ci} / \text{Cb} \quad \dots \text{Eq.2}$$

where:

Ci: the Hg concentration in the sediment sample

Cb: Cb is background concentration of metal in nature 0.08 mg/kg (Panggabean et al., 2022).

#### *Risk of Ecology (RI)*

The RI evaluating the degree of contamination of sediments according to the toxicity of pollutants was calculated:

$$\text{RI} = \sum E_{ir} = \text{T}_{ir} \times \text{C}_{if} \quad \dots \text{Eq.3}$$

where:

RI : Total environmental potential index

$\sum E_{ir}$ : Sigma of the potential environmental index

$E_{ir}$  : index of potential environmental risk of one heavy metal element

$T_{ir}$  : toxic response factor (Hg = 40)

Table 2. Classification of ecological risk index

RI Range	Ecological Risk Level
RI ≤ 40	Low ecological risk
40 < RI ≤ 80	Moderate ecological risk
80 < RI ≤ 160	Strong ecological risk
160 < RI ≤ 320	Very strong ecological risk
RI > 320	Extremely high ecological risk

To describe the potential risk of toxic pollutants, the following formula (Huang et al., 2023) was used:

$$\text{RQ} = \text{Ci}/\text{PNEC} \quad \dots \text{Eq.4}$$

where RQ is the Risk Quoitient; Ci is the Hg concentration in the sediment sample and PNEC is the concentration predicted to have no effect. According to Portezuela et. al. (2019), the PNEC of Hg is estimated at 0.04 mg/kg. RQ ≥ 1 indicates high risk, and RQ < 1 indicates low risk (Huang et al., 2023).

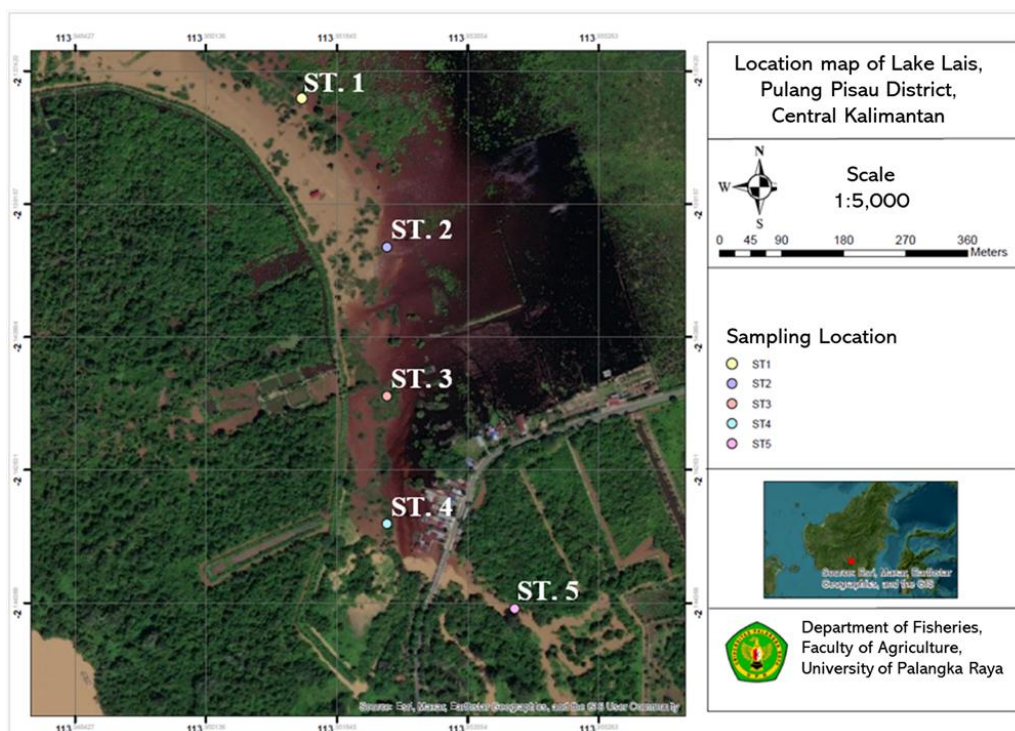


Figure 1. Location map of Lake Lais showing the sampling stations, Pulang Pisau District, Central Kalimantan, Indonesia

### 3. Result and Discussion

#### 3.1. Mercury Content in Water and Sediment

The presence of mercury in water and sediment in Lais Lake might come from several pathways, such as rainwater containing mercury, particles from the air entering the lake and also from the Kahayan River water entering the lake. The sources of mercury to the environment consist of natural and anthropogenic sources (Guentzel et al., 2007; Mao, et al. 2023). Natural sources of mercury include volcanic eruptions and marine emissions. Anthropogenic (human-caused) emissions include mercury released from fuels or raw materials or industrial products or processes (US EPA, 2024).

Mercury released from natural and anthropogenic sources enters aquatic ecosystems primarily through river flow and atmospheric deposition. It is then converted to highly toxic methylmercury (MeHg) through methylation by microbes in sediments and the water column. Sediments are considered a repository of pollutants and environmental changes. Therefore, the aquatic environment is critical to the Hg cycle and human exposure (Mao, et al. 2023).

Based on the results presented in Table 2, the mercury content in Lais Lake water ranged from 0.001 to 0.005 mg/l. The mercury content in lake water is still within the prescribed range by Republik Indonesia (2021). Compared to another research location in Central Kalimantan, the mercury content in Lais Lake water is much higher than the mercury content in Tilap Lake water, which was below 0.000007 mg/l (Elvince, et al. 2008) and Payawan Lake (0,000069 mg/l) (Indrajaya and Virgiyanti, 2019). Therefore, the mercury content in Lais Lake water was lower than that in water from Limboto Lake (0.0146 mg/l) (Niode, et al. 2021).

Mercury content in Lais Lake sediments ranged from 0.142 to 0.473 mg/kg. Mercury content in sediments can have a negative impact on organisms in the water. Sediment mercury can be converted by microorganisms into methylmercury, a highly toxic chemical that builds up in fish, shellfish and animals that eat fish (US EPA, 2024). Some countries set the mercury content in sediments to prevent the adverse effects of mercury presence in sediments. The quality standard value of sediment Hg from ANZECC & ARMACANZ is divided into two categories, namely low (0.15



mg/kg) which indicates low toxicity effects but needs further attention and high (1.00 mg/kg) indicates the reference value as an evaluation of the adverse effects experienced by half the exposed population. Meanwhile, Canadian quality standards in the CCME also show an Interim Sediment Quality Guideline (ISQG) value of 0.13 mg/kg as the limit of negative effects experienced by biota and a Probable Effect Level (PEL) of 0.70 mg/g as an adverse biological effect experienced by biota due to exposure to contaminants. In addition, the

National Oceanic and Atmospheric Administration (NOAA) sets quality standards based on the Effect Range Low (ERL) value of 0.15 mg/kg and Effect Range Medium (ERM) of 0.71 mg/kg which shows the criteria for the relationship between chemical concentrations in sediments and indicators of biological damage caused (Haryati, et. al., 2022). Based on the references, the mercury toxicity in sediment from Lais Lake indicates low adverse effects.

Table 3. Mercury Concentration in Water and Sediment Samples from Lake Lais

Station	Mercury in Water (mg/L)	Mercury in Sediment (mg/kg)	Quality Standards
ST.1	0.001	0.473	<b>Water:</b> 0.0001–0.0005 (PP No. 22/2021) <b>Sediment:</b> - 0.15–1.00 (ANZECC & ARMCANZ) - 0.1–0.7 (Canadian Guidelines) - 0.1–0.7 (NOAA, US)
ST.2	0.002	0.317	
ST.3	0.002	0.217	
ST.4	0.001	0.263	
ST.5	0.001	0.142	

### 3.2. Ecological Risk Assessment of Mercury in Lake Lais

Ecological risk assessment is used to understand the likelihood and consequences of these impacts on ecosystem receptors (Kho, et al., 2022). The geo-accumulation index (Igeo) is widely used to assess pollution levels and sediment health status by comparing the current concentration status with pre-industrial levels (Abdullah, et al., 2020). The geo-accumulation index (Igeo) is calculated to

determine the adsorption rate of heavy metals in sediments. It is determined by comparing sediment heavy metal concentrations and initial concentrations (Ananga et al., 2023).

Based on Table 4, the geoaccumulation index of mercury in Lake Lais is categorized as lightly to moderately polluted. ST. 1, ST. 2 and ST. 4 are included in the moderately polluted ( $1 < I\text{-Geo} < 2$ ), while ST. 3 and ST. 5, are categorized in the lightly polluted criteria ( $0 < I\text{-Geo} < 1$ ).

Table 4. Contamination Factor (CF), Geoaccumulation Index (I-Geo), Risk Index (RI), and Risk Quotient (RQ) Calculated from Mercury Content in Sediments of Lake Lais

Station	Ci	CF	I-Geo	RI=Eir	RQ
ST.1	0,473	5,91	1,979	236,50	11,83
ST.2	0,317	3,96	1,401	158,50	7,93
ST.3	0,217	2,71	0,855	108,50	5,43
ST.4	0,263	3,29	1,132	131,50	6,58
ST.5	0,142	1,78	0,243	71,00	3,55

Ecological risk assessment is a process to evaluate how likely it is that the environment will be impacted as a result of exposure to one or more environmental stressors, such as chemicals, land use change, disease, and invasive species (US EPA, 2024). Table 5 shows

the ecological risk caused by the presence of mercury in sediments from each station. ST. 1 has a very strong ecological risk ( $160 < RI < 320$ ), ST. 2, ST.3 and ST.4 have strong ecological risk ( $80 < RI < 160$ ) and ST. 5 has moderate ecological risk ( $40 < RI < 80$ ). In general, Lake

Lais appears to have a strong ecological risk from the presence of mercury. Some mercury risks to human health and ecology are reproductive disruption, genotoxicity, endocrine disruption, carcinogenicity, and immunosuppression (Huang, et al., 2023).

The risk quotient (RQ) index is estimated to describe the possible hazards of mercury contaminants in the ecosystem (Maulana, et al., 2023). Based on Table 3, the RQ value of each has exceeded 1 ( $RQ > 1$ ), which means that mercury content in sediments in Lais Lake has a high risk to the ecosystem.

#### 4. Conclusion

The findings of this study indicate that mercury content in water samples from Lake Lais (0.001–0.002 mg/L) remains within the permissible limits established by the Government of Indonesia under Regulation PP No. 22 of 2021. In contrast, mercury concentrations in sediment samples, ranging from 0.01 to 0.47 mg/kg, approach or exceed the threshold values recommended by international guidelines (e.g., ANZECC, NOAA, and Canadian sediment quality standards), highlighting a potential ecological risk.

According to the Index of Geoaccumulation (I-Geo), Lake Lais is classified as lightly to moderately polluted. The Ecological Risk Index (RI) categorizes mercury contamination as posing a low to very strong ecological risk. Additionally, Risk Quotient (RQ) values greater than 1 further underscore that mercury contamination in the sediments of Lake Lais represents a significant ecological threat to the aquatic environment.

Future research should focus on identifying pollution sources, expanding spatial and temporal coverage, and assessing bioaccumulation in aquatic biota to evaluate potential human health impacts. Policymakers are encouraged to use this data to develop mitigation strategies, particularly by implementing stricter controls on gold mining and industrial runoff. Ongoing monitoring, public awareness, and integrated management are essential for protecting this vulnerable ecosystem.

#### Data availability statement

Data will be made available on request.

#### Funding Agencies

This study was funded by Faculty of Agriculture, University of Palangka Raya, Indonesia

#### Conflict of interests

The authors declare they have no competing interests

#### Acknowledgment

We would like to thank the Faculty of Agriculture, University of Palangka Raya for supporting the data collection in this research

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## **Microplastic Contamination in Fish, Water and Sediment from Milkfish Ponds: Environmental Insights from Kasemen District, Banten Province, Indonesia**

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Received: 01 November 2024; Accepted: 19 May 2025; Published: 20 June 2025

**Abstract:** Plastic waste discarded into the environment can easily enter water bodies. One as such for an example is the milkfish farming ponds in Kasemen Subdistrict, which have become a dumping ground for household waste. This has led to the entry of plastic waste into the pond waters, which can be degraded into microplastics. This study aims to identify the content and abundance of microplastics in fish and other fish parts such as: flesh, intestines, stomach, gills, water, and sediment, as well as to measure the water quality in the milkfish farming ponds in Kasemen Subdistrict, Serang City, Banten Province. A total of 12 milkfish were collected from 6 stations, with 2 fish taken from each station using the random sampling method across 6 hectares of the farming ponds. The quantity and types of microplastics identified in the samples were analyzed using descriptive statistical analysis with software brand Meiji Techno. The results showed that the milkfish farming ponds in Kasemen Subdistrict were contaminated with microplastics, found in the fish's flesh, gills, intestines, stomach, water, and sediment. The microplastics identified were fiber, film, and fragment types. FTIR test results indicated that the microplastics found were made of polypropylene (PP) polymer.

**Keywords:** milkfish (*Chanos chanos*), microplastics, polypropylene, pond

DOI: <https://doi.org/10.55981/limnotek.2025.8622>

### **1. Introduction**

Waste has become a problem for humans and the environment. Waste comes in various types, one of which is plastic waste (Suprijanto *et al.*, 2021). Plastic waste takes a relatively long time to decompose. Due to this extended period, plastic in aquatic environments will undergo a degradation process, breaking down into smaller particles known as microplastics (Tuhumury and Ritonga, 2020). Microplastics are plastic particles smaller than 5 mm. Microplastics have been studied for differences in molecules, structure, size, shape, color, and

source, making them diverse contaminants or pollutants (Rochman *et al.*, 2019). Water, sediments, plants and aquatic animals can be contaminated by microplastics at varying concentrations (Cole *et al.*, 2011).

The milkfish (*Chanos chanos*) farming ponds in Kasemen Subdistrict have become a dumping ground for household waste. The surrounding community disposes of household waste, especially plastic waste, around the edges of the ponds. This contributes to the entry of plastic waste into the ponds, where it can be degraded into microplastics, allowing

the Kasemen ponds to accumulate microplastics, leading to the contamination of aquatic organisms. Furthermore, these farming ponds are located close to the sea, where plastic waste is present, allowing water flow to carry plastic waste into the aquaculture environment.

The entry of microplastics into water can occur through two categories: 1) primary microplastics, which are small plastic particles that enter marine environments directly, and 2) secondary microplastics, larger particles resulting from the fragmentation of plastic pieces (Ambarsari and Anggiani, 2022). Fish can ingest microplastics accidentally during feeding because their shape is similar to the fish's food, or because the prey itself has been contaminated by microplastics (Yona *et al.*, 2020). Monitoring efforts of microplastics in water, sediment, and organisms (both qualitatively and quantitatively), field sampling techniques, and laboratory tests (identification and quantification of particles) have been extensively conducted to enhance understanding of the negative impacts of microplastics on the environment (Wang and Wang, 2018).

Milkfish is a leading brackish water fish commodity, providing good nutritional value at an affordable price ranging from Rp. 36.000 to Rp. 40.000 for 1 kg, and is popular among the Indonesian population (Akhmadi *et al.*, 2019). The production of milkfish in Banten Province in 2020 reached 12,585 tons (Eris *et al.*, 2020). The location of the milkfish farming ponds, which are close to both the sea and residential areas, makes it possible to find microplastic content accumulated in the body organs of milkfish.

The objective of this research is to identify the content and abundance of microplastics in fish, sediment, and water, as well as to measure water quality in the milkfish farming ponds. Since research on microplastic accumulation in milkfish farming ponds is still limited, the result of this research is expected to provide important information for relevant parties regarding the presence of microplastic accumulation in fish, sediment, and water in the milkfish farming ponds in Kasemen Subdistrict, Serang City.

## 2. Materials and Method

### 2.1. Time and Location of Sampling

The research was conducted from March to April 2023. Samples of fish, sediment, and water were collected, and water quality measurements were taken at the milkfish farming ponds in Kasemen Subdistrict, Serang City, Banten Province. The analysis of microplastic content in milkfish samples was carried out at the Aquaculture Laboratory, Sultan Ageng Tirtayasa University. Additionally, Fourier Transform Infrared (FTIR) ATR (Attenuated Total Reflectance) tests were conducted at the Integrated Laboratory and Research Center (ILRC), University of Indonesia.

### 2.2. Sample Collection

Fish sampling was carried out in the milkfish farming ponds, located in Kasemen Subdistrict, Serang City. A total of 12 milkfish were collected from 6 stations, with two fishes taken from each station using the random sampling method across 6 hectares of the farming ponds. This method refers to the research conducted by Sawalman *et al.* (2021). Simple random sampling, also is a method of selecting samples from the population randomly and straightforwardly, ensuring each population member has an equal chance of being chosen (Harahap *et al.*, 2018).

Sampling was conducted weekly: week 1 (station 1), week 2 (station 2), week 3 (station 3), week 4 (station 4), week 5 (station 5), and week 6 (station 6). The samples taken from these 6 points include the inlet, middle, and outlets areas. Stations 1 and 2 represent the inlet area, stations 3 and 4 represent the middle area, and stations 5 and 6 represent the outlet area.

Sediment samples were collected from each station using a 2-inch PVC pipe. Sediment sampling was conducted at a depth of 16 cm from the surface. The sediment samples were labeled with identification tags. Meanwhile, water samples were taken from each station using a plankton net at the water surface and stored in sample bottles. Water quality parameters (pH, temperature, and salinity) were measured at each station. According to Humaerah and Rasyid (2024), pH, temperature, and salinity are significant factors affecting microplastics.

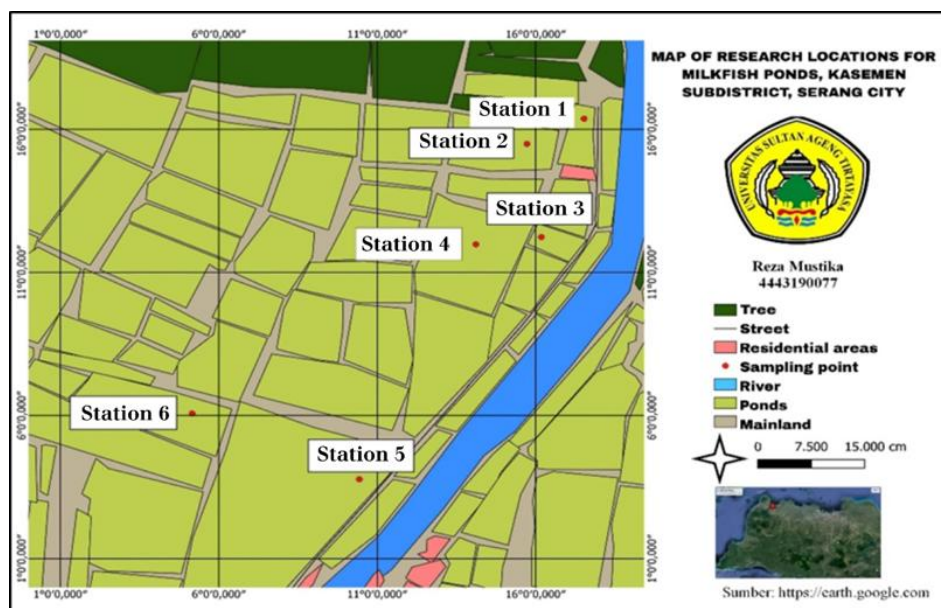


Figure 1. Map of the research location for sampling in the milkfish aquaculture pond

## 2.3. Microplastic Sample Analysis

### 2.3.1 Milkfish Sample Analysis

The total length of the milkfish was measured using a 30 cm ruler, and the weight was recorded using a digital scale. The fish were dissected from the anus to the dorsal area, following the lateral line to the head and downward to expose the stomach and internal organs. The flesh, gills, and digestive organs (intestines and stomach) were removed. Each organ was weighed and placed in labeled sample bottles 100 ml.

The sample extraction method followed Purnama *et al.* (2021), the samples were placed in sample bottles, then a 10% v/w KOH solution was added at a 1:3 sample volume ratio. The bottles were sealed and incubated at room temperature for 24 hours. Once the organs had dissolved, the samples were filtered using Whatman No. 42 filter paper and dried in an oven at 75°C for three hours. The dried samples were observed using a stereo microscope, and documentation was done with a camera. Identified microplastics were placed into a tube with a sample weight of 1 gram for FTIR ATR testing.

After collecting sample data, the abundance of microplastics was calculated. According to Purnama *et al.* (2021), microplastic abundance can be calculated using the following formula:

$$K = \frac{N_i}{N} \quad \dots \text{Eq.1}$$

where:

- K = microplastic abundance (particles/individual)  
 $N_i$  = number of microplastic particles found (particles)  
 $N$  = number of fish (individuals)

### 2.3.2 Sediment Sample Analysis

Sediment samples were dried in an oven at 70°C for 48 hours. Once dried, the sediment was ground and sieved using Whatman No. 42 filter paper, and 25 grams of the sample were placed in a sample bottle (Layn *et al.*, 2020). The filtered samples were mixed with NaCl solution at was added at a 1:3 sample volume ratio and left for 24 hours. Afterward, the samples were observed using a stereo microscope. The formula to calculate microplastic abundance in sediment is as follows (Putro, 2021):

$$\text{Microplastic abundance} = \frac{\text{The number of microplastics in sediment (particles)}}{\text{Sediment weight (grams)}} \quad \dots \text{Eq.2}$$

### 2.3.3 Water Sample Analysis

The water samples, stored in 150 ml sample bottles, were mixed with NaCl solution at a ratio of 1 part water to 3 parts NaCl solution and left for 24 hours to separate any remaining plastic contaminants. After 24 hours, 1 ml of the water sample was taken using a pipette and observed under a stereo microscope (Hasibuan *et al.*, 2021). Chemical substances like NaCl can separate polymers with lower density since NaCl has a high density (Sutanhaji *et al.*, 2021). The formula to calculate microplastic abundance in water is as follows (Syachbudi, 2020):

$$C = \frac{n}{v} \quad \dots \text{Eq.2}$$

where:

C = microplastic abundance (particles/ind)  
n = number of microplastic particles in water  
v = filtered water volume (mL)

### 2.3.4 Fourier Transform Infrared (FTIR) Test

The Fourier Transform Infrared (FTIR) test aims to determine the type of plastic polymer contained in the microplastics found in the digestive organs of milkfish. The identified

microplastics were placed in tubes for classification based on type (film, fiber, and fragment), texture (hard, soft, thick, and thin), and color of the microplastics found in the milkfish samples.

## 3. Results and Discussion

### 3.1. Abundance of Microplastic Types

The identification of microplastics using a stereo microscope on milkfish, sediment, and water in aquaculture ponds revealed the presence of film, fiber, and fragment types (Figure 2). The presence of microplastics in an environment is influenced by oceanographic factors such as currents and waves, allowing them to travel vast distances from their pollution sources (Yona *et al.* 2020). This milkfish aquaculture pond is located close to the sea and residential areas, which affects the abundance of microplastics found in this research. Visually, during data collection in the field, various types of plastic waste could be seen in the area. According to Ayuningtyas *et al.* (2019), the entry of microplastics from the sea into the water column is caused by current. Microplastics carried by currents will accumulate in the aquaculture pond in Kasemen District.

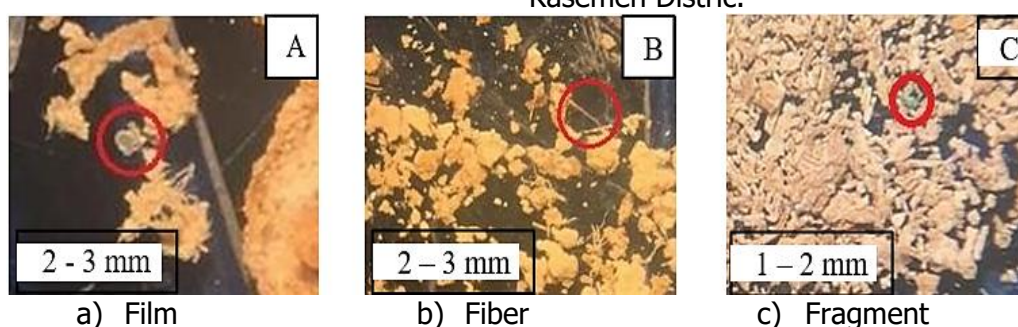


Figure 2. Types of microplastics found in the milkfish pond

In figure 2, film-type microplastics are found in the flesh organ, fiber-type microplastics are found in the gill organ, and fragment-type microplastics are found in the intestine organ. Film-type microplastics have characteristics resembling sheets or plastic fragments and are transparent in color. They are commonly used in the production of plastic bags or packaging. Amin *et al.*, (2020) stated that film-type microplastics have a lower density compared to other types of microplastics, allowing them to float in the water column. Consequently, when fish feed, microplastics may unintentionally

enter their mouths. Fiber-type microplastics are typically characterized by fibers or shapes resembling fishing nets, often colored red or black. Fragment-type microplastics appear as shards from bottle waste, jars, mica, or small pieces from PVC pipes, and they are typically blue, green, or yellow (Ambarsari and Anggiani 2022). Fragment-type microplastics are frequently found in fish intestines due to their widespread dispersion in the water column (Amin *et al.*, 2020). Microplastics enter aquatic environments from the sea, carried by currents (Ayuningtyas *et al.*, 2019).



### 3.2. Microplastic Abundance in Milkfish

Microplastic abundance in milkfish was identified in the gills, flesh, stomach, and intestines. At stations 1, 4, and 5, microplastic abundance was dominated in the gills, with 22 particles/ind, 6 particles/ind, and 11

particles/ind, respectively. Meanwhile, at stations 2, 3, and 6, the flesh had the highest abundance, with 8 particles/ind, 6 particles/ind, and 6 particles/ind, respectively. This may be due to the milkfish consuming microplastics either intentionally or unintentionally.

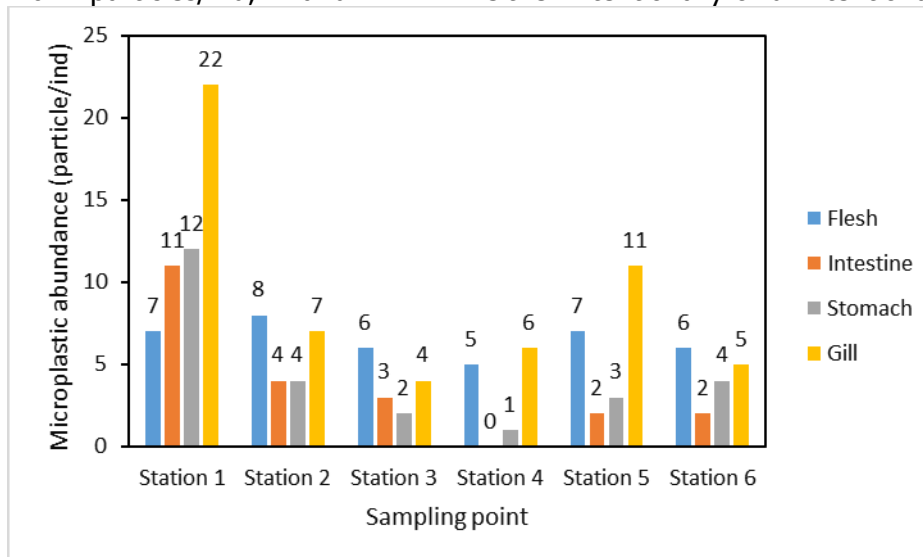


Figure 3. Microplastic abundance in milkfish (flesh, intestines, stomach, and gills)

Microplastics can accumulate in various fish organs, including flesh, gills, and the digestive system (intestines and stomach). The digestive system is where microplastics accumulate as part of the fish's feeding process. Research shows that flesh and gills contain higher microplastic levels than the digestive system (Figure 3). This aligns with Yona *et al.* (2020), who found that microplastics accumulate more in the gills than in the digestive system. Utomo *et al.* (2022) explained that microplastics consumed by fish enter the gastrointestinal tract. If not excreted through urine or feces, microplastics can pass through the gastrointestinal walls and spread to other tissues or organs via the bloodstream. The flesh is one tissue where microplastics can accumulate. Utomo *et al.* (2022) stated microplastics consumed by fish then enter the gastrointestinal tract. If microplastics are not

excreted from the body through urine and feces, they can pass through the gastrointestinal tract walls and spread to body tissues or other organs via the bloodstream. Annas (2023) added that aquatic organisms may stop feeding due to a false sensation of fullness caused by indigestible microplastics in the stomach, potentially leading to death from starvation. Yona *et al.* (2020) stated that microplastics entering fish gills come directly from the water as part of the fish's respiration process.

### 3.3. Correlation Between Milkfish Weight and Microplastic Abundance

The relationship between milkfish weight and microplastic abundance was studied to determine whether fish weight affects microplastic abundance. The relationship between fish weight and microplastic abundance is presented in table 1.

Table 1. Relationship between milkfish weight and microplastic abundance

Model Summary			Anova		Coefisien	
R	R Square	df	F calculated	Significance	t calculated	Significance
0.255	0.065	1	1.525	0.230	1.235	0.230
		22				
		23				

The correlation coefficient ( $R$ ) is 0.255, and the coefficient of determination ( $R^2$ ) is 0.065. With a significance value of  $0.230 > 0.05$ , this indicates that fish weight has no significant effect on the number of microplastics. Fachrudin (2022) concluded that fish weight does not influence the amount of microplastics. The amount of microplastics entering fish bodies is more influenced by feeding habits. Microplastics have a size of less than 5 mm. Hermawan *et al.* (2022) stated that fish may unintentionally consume microplastic particles while foraging. Margaretha *et al.* (2022) found that the presence of microplastics in fish is

related to the species studied, habitat, feeding habits, plastic particle density, and the presence of microplastics in the aquatic environment.

### 3.4. Microplastic Abundance in Aquaculture Pond Water

The study results show that microplastic abundance in aquaculture pond water is dominated by the film type. Station 3 had the highest number of microplastics, with 6 film-type particles/mL, while station 1 had the fewest, with 1 film-type particle/mL. Fiber-type microplastics were the least common in pond water.

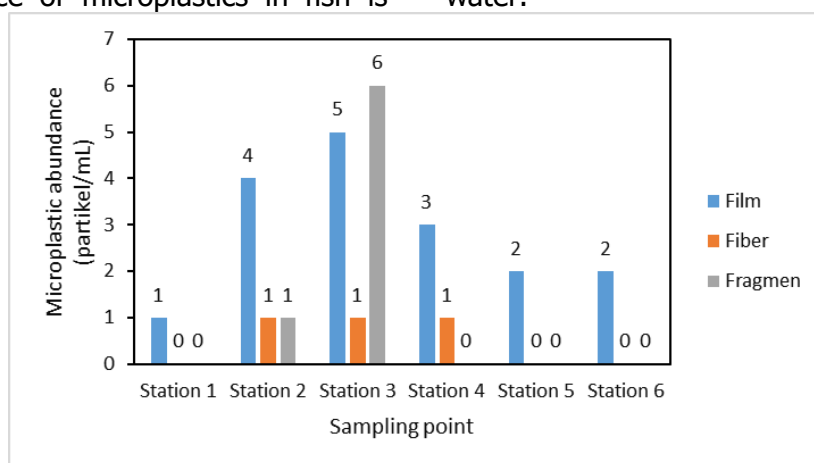


Figure 4. Microplastic abundance in aquaculture pond water

The abundance of film microplastics in aquaculture ponds suggests the presence of macro debris, such as plastic bags, bottles, plastic wrap, and other waste. The dominance of film-type microplastics in pond water is due to human activity, such as disposing of food packaging near ponds or coastal areas close to the ponds. These wastes are carried into the ponds by currents or waves. At stations 2 and 3, fragment-type microplastics were found. This corresponds to the condition of stations 2 and 3, where a significant amount of plastic bottle and mica waste was carried by currents from the sea. Amin *et al.* (2020) stated that film-type microplastics have a lower density than other types, allowing them to float in the water column. Ayuningtyas *et al.* (2019) noted that film-type microplastics consist of thin sheets from packaging, are transparent, and have a lower density, making them easily

transportable. This accounts for the dominance of film-type microplastics in aquaculture pond water. Putri (2021) stated that microorganisms and other particles can cause film microplastics to sink, accumulating at the bottom of the water and potentially being ingested by fish.

### 3.5. Microplastic Abundance in Aquaculture Pond Sediment

Microplastic abundance in pond sediment was dominated by fragment-type microplastics at all stations. Station 1 had the highest number, with 14 fragment-type particles/g, while station 4 had the fewest, with 1 fragment-type particle/g (Figure 5). Station 1 is the station located near the sea, which may result in a large accumulation of plastic waste carried by currents from the sea to station 1. This leads to a very high abundance of fragment-type microplastics at station 1.

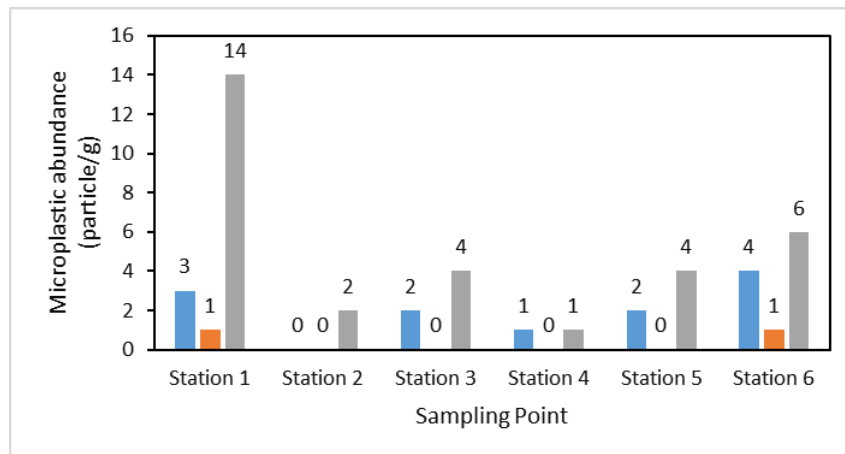


Figure 5. Microplastic abundance in aquaculture pond sediment

Azizah *et al.* (2020) explained that microplastics entering water bodies eventually settle in sediment. The presence of microplastics in sediment poses ecological risks. Fragment-type microplastics dominate sediment due to human activities, particularly the disposal of plastic bottles and mica-based waste near aquaculture ponds. Layn *et al.* (2020) stated that the high abundance of fragment-type microplastics results from the dominant plastic bottle waste along water bodies. Sarasita *et al.* (2020) noted that fragment-type microplastics have higher density, making them more likely to sink to deeper waters or sediment. Yona *et al.* (2019) found high levels of fragment-type

microplastics in aquatic sediment. Another factor that can influence the presence of microplastics in fish ponds is the local residents disposing of waste in the pond area, as well as the currents from the sea flowing into the pond. As a result, the fish in the pond can accumulate microplastics.

### 3.6. Aquaculture Pond Water Quality Measurement

Water quality measurements in aquaculture ponds were conducted at each research station, assessing parameters such as temperature, pH, and salinity. Water quality parameters were measured at the same time as fish and sediment sampling. The results are shown in Figure 6.

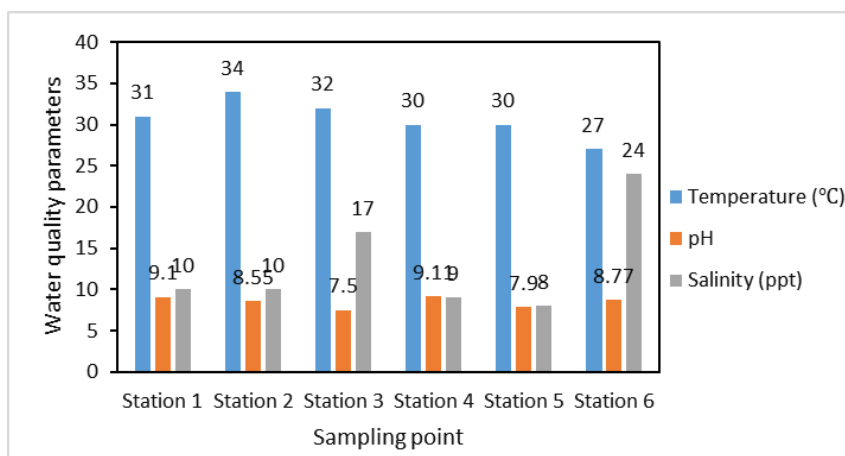


Figure 6. Aquaculture pond water quality

Milkfish can survive in temperatures of 27–35°C. A pH range of 6–9 is ideal for milkfish farming, and the fish can live in salinity levels of 0–35 ppt (Wahyuni *et al.*, 2020). This shows that the water quality in aquaculture ponds remains within normal limits for milkfish farming. Water quality affects the survival,

growth, and development of aquatic organisms. If water quality is good, it supports the survival of aquatic life such as fish.

Water temperature in ponds fluctuates more than in coastal waters due to the smaller water volume and larger surface area, making pond water heat and cool more quickly (Jefri *et al.*,

2020). pH fluctuations occur because of rainwater input, which lowers pH levels. The pH may rise when measured during the day due to photosynthesis, which reduces CO<sub>2</sub> levels and

increases pH (Faizin, 2018). Yunarty *et al.*, (2022) noted that salinity fluctuations are due to evaporation during the day.

Table 2. Interpretation of FTIR Test on the Digestive Organs of Milkfish

Sample Name	Peak Result (cm <sup>-1</sup> )	Functional Groups	Polymer Characterization
Station 1 and 2	3374.91	N - H stretch	<i>polypropylene</i> (PP)
	2918.95	C - H stretch	
	2585.02	C - H stretch	
	1628.9	C = O stretch	
	1371.82	CH <sub>2</sub> bend	
	997.4	CH <sub>3</sub> rock, CH <sub>3</sub> bend, CH bend	
	909.88	Aromatic ring stretch or CH bend	
	830.16	Aromatic CH out-of-plane bend	
	792.96	Aromatic CH out-of-plane bend	
	752.69	CH <sub>2</sub> rock, C = O bend	
Station 3 and 4	3361.3	N - H stretch	<i>polypropylene</i> (PP)
	2921.55	C - H stretch	
	2620.72	C - H stretch	
	2582.78	C - H stretch	
	1620.77	C = O stretch	
	1391.66	CH <sub>2</sub> bend	
	829.36	Aromatic CH outof-plane bend	
	699.96	Aromatic CH out-of-plane bend	

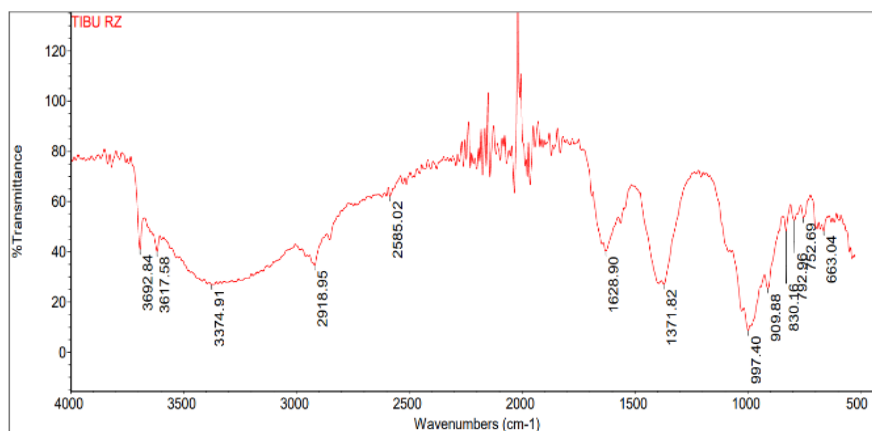


Figure 7. FTIR Test Results of Intestinal Samples from Stations 1 and 2

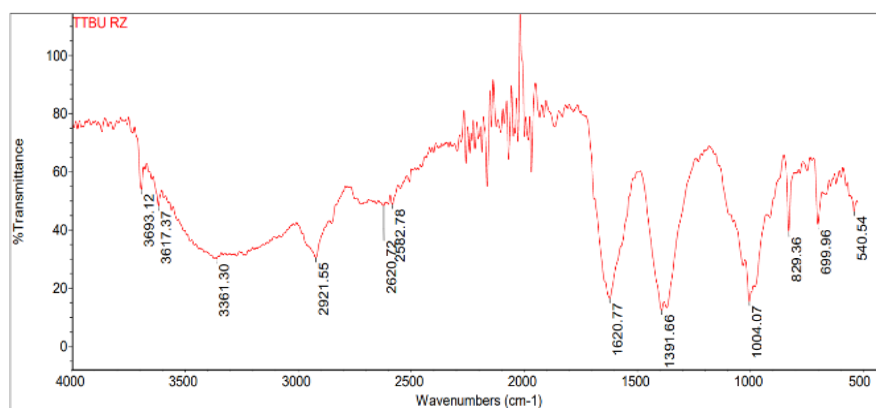


Figure 8. FTIR Test Results of Intestinal Samples from Stations 3 and 4



### 3.7. FTIR Test on Milkfish Digestive Organs

FTIR (Fourier Transform Infrared) tests were conducted to identify the type of polymer in the samples. The polymer identification used FTIR ATR. Seftianingrum *et al.*, (2023) stated that FTIR ATR spectra are obtained from an electro-nexus spectrophotometer equipped with a Diamond Smart Orbit. Murtadho (2023) added that ATR (Attenuated Total Reflectance) is suitable for analyzing thick or highly absorbent solid materials.

The estimation of microplastic polymers can be determined based on wavelength analysis, the estimation can be compared based on the research by Jung *et al.*, (2018). The results of the FTIR test on the digestive organs of milkfish indicated the estimation of the polymer type as polypropylene (PP).

The type of microplastic found in the FTIR test of the digestive organs of milkfish is PP. Plastic is a synthetic polymer made from petroleum, characterized by its flexibility and difficulty in biodegradation. The polymer most commonly used in plastic production is PP (Murtadho, 2023). PP has strong characteristics, low vapor permeability, excellent resistance to fats, durability at high temperatures, and lightweight properties. Additionally, this type of polymer is resistant to corrosion and has a high melting point. Due to these properties, PP is widely available in the market. Polypropylene is commonly used in household plastic products such as funnels, glass bottles, buckets, plastic bags, milk bottles, detergent packaging, margarine packaging, water pipes, trash bins, and various types of reusable containers (Faqih 2022). The results of the FTIR analysis are consistent with the conditions of the aquaculture pond in Kasemen District, which is very close to human activities and the sea, where the surrounding population disposes of used waste near the pond.

### 4. Conclusion

The types of microplastics found in milkfish, water, and sediment include film, fiber, and fragments. Based on the data from the research conducted, it can be concluded that the abundance of microplastics in the organs of milkfish from Kasemen pond was dominated by film-type microplastics. The abundance of

microplastics in the sediment of the aquaculture pond in Kasemen District is dominated by fragment-type microplastics. The types of microplastics found in milkfish, water, and sediment include film, fiber, and fragments. The FTIR test results on the digestive organs of milkfish indicated the dominant polymer type as PP. The quality of the water, including temperature, pH, and salinity in the aquaculture pond of Kasemen District, remains within normal limits for milkfish farming.

### Data availability statement

Data used in this study can be requested from the author.

### Conflict of interest

The authors declare there is no conflict of interest.

### Authors contribution

**RM, DH, DA, and MBS:** idea concept, data collection, conceptualization, data analysis, and writing the original draft. **RK:** review and editing.

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**Microplastics in Sediment and Digestive Tract of Amazon Sailfin Catfish  
(*Pterygoplichthys* Spp.) in the middle segment of the Citarum River,  
Karawang, West Java, Indonesia**

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Received: 09 November 2024; Accepted: 26 December 2024; Published: 20 June 2025

**Abstract:** Microplastic pollution, primarily driven by industrial, residential, and agricultural activities, is a growing concern in the middle section of the Citarum River, Indonesia. Microplastics research was conducted three times in the central Citarum watershed from February to April 2022. This study investigates the types, abundance, and polymer composition of microplastics found in the sediment and digestive tract of the Amazon sailfin catfish (*Pterygoplichthys* spp.). Four types of microplastic-pellets, films, fibers, and fragments were identified, with an average abundance of  $602.22 \pm 563.87$  particles/kg in sediment and  $90.6 \pm 40.7$  particles/individual in catfish. The majority of particles were smaller than 0.3 mm. Identified polymers included Polyamide (PA), Polystyrene (PS), Polypropylene (PP), Cellulose Acetate (CA), Acrylonitrile Butadiene Styrene (ABS), Polyvinyl Chloride (PVC), Melamine, Ethylene Vinyl Acetate (EVA), Polyethylene (PE), Low-Density Polyethylene (LDPE), High-Density Polyethylene (HDPE), and Polyethylene Terephthalate (PET). Statistical analysis showed no significant difference in microplastic pollution among industrial, densely populated residential, and agricultural areas. These findings underscore the widespread distribution of microplastics in the Citarum River and highlight the need for comprehensive mitigation strategies.

**Keywords:** Citarum River, microplastics, pollution, polymers, sediment, Amazon sailfin catfish, *Pterygoplichthys* spp

DOI: <https://doi.org/10.55981/limnotek.2025.8790>

## 1. Introduction

The increasing production and use of plastics has certainly led to a growing amount of plastic waste (Mauludy *et al.*, 2020). As the number of people using plastics in their daily activities increases, the accumulation of plastic waste in the natural environment (Eriksen *et al.*, 2014; Kuncoro, 2018;) has become an environmental hazard (van Emmerik & Schwarz, 2020). An estimated

80% of plastic waste comes from land-based sources such as residential, commercial, public places, agriculture and industry (Ambarsari & Anggiani, 2022). Plastic waste has accumulated in the terrestrial environment, open ocean, coastlines, remote islands, and deep sea (Barnes *et al.*, 2009; Jakovljevic *et al.*, 2020). Plastic debris can carry toxic chemicals (Barnes *et al.*, 2009), including heavy metals (Hidalgo-Ruz *et al.*,



2012), which can adversely affect growth and reproduction (Cera *et al.*, 2020). Plastic is not biodegradable (Arisandi *et al.*, 2020; Mavuso & Singwane, 2020), and degradation is a slow process, so that plastic particles can persist in aquatic environments for long periods (Barnes *et al.*, 2009). When plastic degradation peaks, the abundance of microplastics and nanoparticles will increase and far outnumber plankton (Jovanović, 2017). Microplastics are pollutants commonly found in aquatic environments. (Nurhasanah *et al.*, 2021).

Microplastics are plastic particles that have a size of less than 5 millimeters, with the lower size limit of microplastics not determined (Masura *et al.*, 2015). According to its source, microplastics come from primary microplastics produced as resin pellets or as additives to personal care products (e.g., soaps and scrubs in cosmetics) and secondary microplastics are degradation products of larger plastic objects, which are broken down by biological degradation, UV radiation and physical abrasion into smaller fragments (Whitehead *et al.*, 2021). The presence of microplastics in water bodies, sediments, and aquatic organisms, including fish, has been studied extensively in various regions around the world. Aquatic organisms, including fish, can ingest microplastics containing toxic metals and become vectors for the spread of pathogens and microbes that tend to accumulate in the gills, liver, mantle and muscles, and digestive glands through various routes of exposure, such as drinking contaminated water, eating prey contaminated with microplastics, or ingesting microplastics through the gills. In fish, microplastics cause intestinal damage with rupture of the villus and rupture of enterocytes, leading to death, while in humans, microplastic hazards cause genomic instability and increased DNA damage, which can lead to infertility and cancer (Mandal & Mishra, 2023).

The Citarum River, a vital waterway in West Java, Indonesia, is among the most polluted rivers globally. Stretching 297 km (Hidayat *et al.*, 2022, it provides water for nearly 27 million people (MenPUPR, 2016). The river is divided into upstream, middle, and downstream sections, with its middle section,

particularly in Karawang, West Java, characterized by agricultural, industrial, and densely populated residential areas. Despite studies focusing on the upstream and downstream sections, research on microplastics in the middle section remains scarce. It is one of the most polluted rivers in the world. The release of plastic waste as a secondary source of microplastics in the Citarum River is still quite high, reaching more than 1000 kg per hour (Hariyadi *et al.*, 2022). Polymer types found in previous studies in the Citarum River include Polyethylene Terephthalate, Polystyrene, High-Density Polyethylene, Low-Density Polyethylene, Polyvinyl Chloride, and Polypropylene (Aisyah *et al.*, 2022). Microplastics with a lower density than water float, while those with a higher density sink and settle in the sediment (Laksono *et al.*, 2021). The middle Citarum River has sediments from ancient volcanic activity, containing silt, clay, sand and gravel (Citarum, 2016), and the higher density of plastic compared to water causes microplastics to accumulate at the bottom of the river water (Azizah *et al.*, 2020). The slope of the river also accelerates the deposition of microplastic particles.

This study aims to assess the abundance and characteristics of microplastics present in the sediments of the middle section of the Citarum River in Karawang, West Java, and to identify the abundance and characteristics of microplastics found in the digestive tract of Amazon sailfin catfish inhabiting the same area. The results of this study are expected to provide important information on the current state of microplastic pollution in the Citarum River system and its potential impact on local aquatic life.

## 2. Material and Methods

### 2.1. Sampling Area

The sampling was collected in three repetitions from February to April 2022 in the heavily polluted middle Citarum River, Karawang district (see Table 1 and Figure 1). This area is a densely populated, industrialized and agricultural area. This leads to considerable pollution from plastics and other wastes (Hidayat *et al.*, 2022). Water levels were relatively stable during these

three months, allowing for consistent sampling. This study aimed to assess the abundance and composition of microplastics

in the sediment and digestive tract of Amazon sailfin catfish, a common invasive species found in the Citarum River.

Table 1. Sampling location.

Station	Location	Coordinates	Description
Station 1	Wadas, Teluk Jambe Raya	S 06°19'37.41", E 107°18'41.81"	Represents industrial areas with factories and warehouses.
Station 2	Kp. Pasir Panggang, East Teluk Jambe	S 06°18'39.71", E 107°16'34.83"	Densely populated residential area with hotels, restaurants, and showrooms.
Station 3	Sumedangan, Teluk Jambe Timur	S 06°16'47.63", E 107°16'27.83"	Agricultural area representing farming activities.

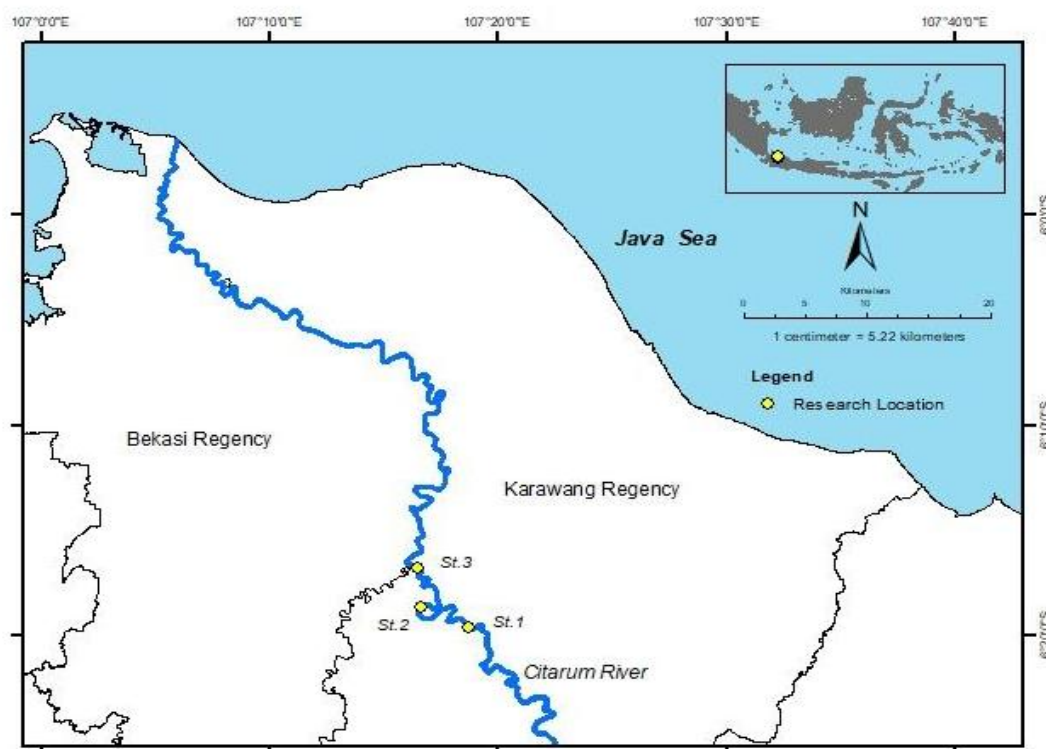


Figure 1. Map of sampling sites in the middle segment of the Citarum River, Karawang, West Java, Indonesia.

## 2.2. Sample collection

Sediment sampling was carried out by inserting a sediment core made of a transparent acrylic tube with a 2-inch diameter into a sediment column at a depth of 10 cm. The collected sediment sample was placed and labeled in an aluminum Ziplock container with 500 grams (wet weight).

Amazon sailfin catfish (*Pterygoplichthys* spp.) were collected from catches of local fishers specializing in Amazon sailfin catfish (*Pterygoplichthys* spp.) from study sites representing a range of community activities,

including industrial areas, densely populated settlements, and agriculture. The number of samples was set at five fish to represent the number of fish caught each month (Julia *et al.*, 2020), with February samples identified as samples 1 through 5, March samples as samples 6 through 10, and April samples as samples 11 through 15, respectively. Samples were then stored in a refrigerator before analysis.

## 2.3 Sample Preparations

### a. Sediment Sample

A 300-gram wet sediment sample was dried in an oven at 50 °C for three days until completely dry (Rochman *et al.*, 2015). The dry sediment was weighed, and 50 grams of it was placed into a beaker. Subsequently, 50 ml of saturated NaCl solution (prepared by dissolving 300 grams of NaCl in 1 liter of distilled water) was added and stirred until homogeneous. NaCl is used to separate the plastic waste from the sediment by density separation, where the plastic waste floats and separates from the sediment that settles due to the difference in density. The part separated from the sediment is called the supernatant. The mixture was covered with aluminum foil and left to settle for 24 hours to form a clear supernatant. The supernatant was filtered through a 5 mm sieve to remove particles larger than 5 mm and then filtered again through a 0.3 mm sieve to remove particles smaller than 0.3 mm. The filter results from the 0.3 mm filter were transferred to a beaker for further analysis.

The next step was adding 20 mL of 0.05 M Fe (II) solution and 20 mL of 30% hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) into the sample's beaker. The mixture was left at room temperature for 5 minutes, then heated on a hot plate to 70°C until gas bubbles appeared. The sample was removed and allowed to cool slightly. After that, it was reheated to 70°C for approximately 30 minutes (Brugge *et al.*, 2020). If organic material is still present in the sample and visible with the naked eye, an additional 20 ml of 30% H<sub>2</sub>O<sub>2</sub> could be added until the organic material is fully degraded (Mauludy *et al.*, 2019). These steps were repeated as necessary to ensure complete decomposition of organic material. The sample was then filtered using a 0.3 mm filter to separate microplastics from degraded organic material, assisted by spraying with distilled water. Samples that are free from organic material are then refiltered using GFF filter paper and a vacuum pump. The ready-filter paper was further identified using a stereo microscope and FTIR microscopy.

### b. Fish Sample

The microplastic content of the fish was analyzed through a two-step process. First, the fish samples were cleaned with distilled water to remove contamination from impurities and

microplastics. The total length and weight of the fish were measured. The fish were dissected, and the digestive tract was weighed and transferred to a sterile beaker.

In the second step, 10% KOH solution was added to the fish gut samples in a beaker three times the sample volume and incubated overnight at 60°C to degrade the organic matter (Rochman *et al.* 2015). To further degrade the remaining fish intestines, 20 ml of 30% H<sub>2</sub>O<sub>2</sub> solution was added. The sample was heated at 70°C until the salt dissolved and left overnight at room temperature. The sample was then filtered using Whatman GFF paper with the help of a vacuum pump, and the filtrate was further identified using a stereo microscope, the same as the sediment sample.

## 2.4 Microplastic analysis

Observations of microplastic particles in sediment and fish digestive tracts were carried out using a Nikon SMZ2B Stereo microscope with an additional 51-megapixel Koppache microscope camera, with a zigzag scanning pattern from left to right to ensure the entire sample area was thoroughly covered. This ensured all particles were detected and analyzed. During observations, microplastic particles were identified visually based on the following criteria: Resistance to torn with tweezers (particles have sufficient strength and durability, preventing them from being torn using tweezers); particles have a lack of organic structure (plastic particles do not have a structure associated with organic materials, such as wood fiber or other organic materials) (Peng *et al.*, 2017). Microplastic particles identified and met those criteria were measured using a Nikon SMZ2B microscope, with a Koppache camera and software. In this study, only particles smaller than 5 mm were measured. Particles exceeding the size ranges were isolated and excluded from further analysis.

Fourier Transform Infrared Spectroscopy (FTIR) analysis was conducted at the Advanced Characterization Laboratory, National Research and Innovation Agency (BRIN) in Cibinong. The samples were analyzed using FTIR device employing the Attenuated Total Reflectance (ATR) method. The obtained spectrum was compared with the reference spectrum from

the database, with matches accepted at a confidence level of 75% or higher.

## 2.5 Data analysis

The classification of microplastics is based on their types-pellets, films, fibers and fragments and their size, categorized as <0.3 mm, 0.3-<0.5 mm, 0.5-<1 mm and 1-5 mm (Liu *et al.* 2021). The abundance of microplastics in sediment was calculated based on the sample weight of 50 g, conserved to 1 kg (Hidalgo-Ruz *et al.* 2012). Microplastic concentration was expressed as the number of particles per kilogram of dry sediment (Claessens *et al.*, 2011). For microplastics detected in the digestive tract of Amazon Sailfin Catfish (*Pterygoplichthys* spp.), the abundance was calculated in particles per individual (Gresi *et al.*, 2021).

The statistical analysis method used was the Shapiro-Wilk normality test, followed by the Kruskal-Wallis test, to determine the differences in microplastic abundance across three stations in the middle segment of the Citarum River. Data was analyzed using Microsoft Excel 2019 and IBM SPSS Statistics 25.

## 3. Result and Discussion

### 3.1 Microplastics found in sediment

This study identified 21,680 microplastic particles from 9 sediment samples from industrial, residential and agricultural areas (see Table 2 and Figure 2). The average total abundance was  $602.2 \pm 563.9$  particles/kg. The particles consisted of four types: pellets, films, fibers, and fragments. Fragments were the most common type in industrial areas, while fibers dominated in residential and agricultural

areas. The particle sizes were predominantly less than 0.3 mm, followed by 0.3-0.5 mm and 0.5-1 mm. The highest concentrations were observed in March 2022 for industrial areas and in February 2022 for residential and agricultural areas.

At Station 1, the most common type of microplastics was fiber, with an average total abundance of  $1533 \pm 1307.3$  particles per kilogram. At Station 2, fragments were the most common type, with an average total abundance of  $686.7 \pm 98.5$  particles per kilogram. At Station 3, fiber was the predominant type, with an average total abundance of  $1120 \pm 429.7$  particles per kilogram.

Microplastic particles were detected sediment samples in the middle section of the Citarum River. These particles found were in the form of pellets, films, fibers and fragments. The total abundance of plastic particles which dominantly found at Station 1 was fiber. The high abundance might be caused by the wastewater from washing process in garment factories in the area (Ali *et al.*, 2021). Fiber particles can also derive from glass fibers (Sutanhaji *et al.*, 2021).

At Station 2, which represents a densely populated residential area, microplastics enter the waters mainly through household waste and runoff from residential areas. The highest form of microplastic found at this station was the fragment. Fragment-shaped microplastics might come from items, such as drinking bottles, jars, buckets, mica folders, parallel pipes, containers/jerry cans, irrigation pipes, and plastic pots (Sutanhaji *et al.*, 2021).

Table 2. Summary of microplastic data identified from sediments

		Station 1	Station 2	Station 3	Total
Average Number of MPs $\pm$ SD (Particles/kg)		658.3 $\pm$ 828.5	446.7 $\pm$ 264.7	701.7 $\pm$ 431.5	602.2 $\pm$ 563.9
Total number of MPs		7,900	5,360	8,420	21,680
Total number of MPs by type of debris	Pellet	400	1,020	1,660	3,080
	Film	1,000	300	660	1,960
	Fiber	4,600	1,980	3,360	9,940
	Fragment	1,900	2,060	2,740	6,700
Total number of MPs by size (mm)	<0.3	5,900	2,440	4,100	12,440
	0.3 - < 0.5	880	1,840	2,480	5,200
	0.5 - <1	1,020	1,040	1,440	3,500
	1-5	100	40	400	540



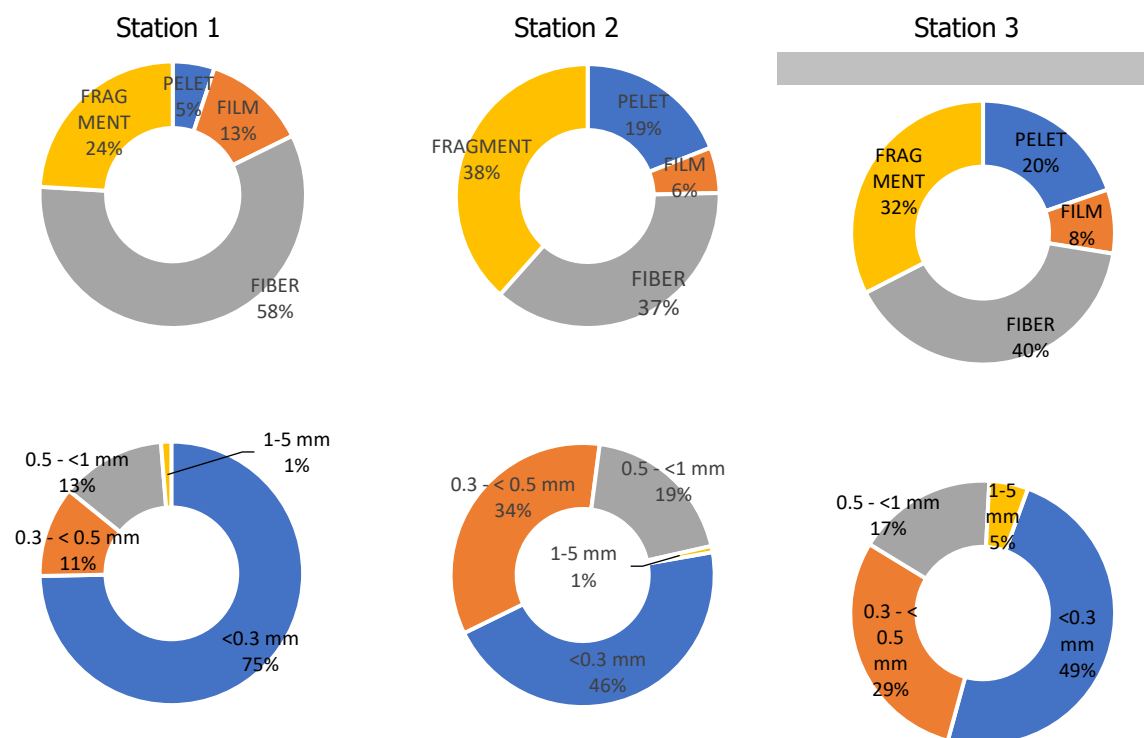


Figure 2. Distribution of microplastic particles by type, and size collected in sediments at three study sites Station.1 (Industrial Area); Station 2 (Densely Populated Residential Area); Station 3 (Agricultural Area)

At Station 3, an agricultural area, fiber was the highest form of microplastic particles. Similar to Station 1, fiber in these agricultural areas is likely derived from materials such as clothing, rope, cloth, mulch, plastic sacks, fiberglass raffia, and others (Sutanhaji *et al.*, 2021).

Comparing the sediment abundance average in the middle section of the Citarum River of  $602.22 \pm 563.87$  particles/kg with several other rivers, microplastics in sediments in the middle section of the Citarum River are smaller than microplastics in sediments in the Opak River, Progo River of  $1,799.33 \pm 1,430.87$  particles/kg and  $645.33 \pm 405.94$  particles/kg, respectively. The average microplastic abundance in sediments in the middle section of the Citarum River is  $602.22 \pm 563.87$  particles/kg. Compared to other rivers, this is lower than the concentration in the Opak River and Progo River, which are  $1,799.33 \pm 1,430.87$  particles/kg and  $645.33 \pm 405.94$  particles/kg, respectively (Utami *et al.*, 2022). However, it is higher than the levels in Muara Badak, Kutai Kartanegara Regency, which range from 69.3 to 90.12 particles/kg (Dewi *et*

*al.*, 2015), and the Sei Kambing River at 32.3 particles/100g (Hasibuan *et al.*, 2020).

The particle size of microplastics in sediments from three sampling sites: industrial areas, densely populated settlements, and agricultural areas, is predominantly less than 0.3 mm, with the least represented size range being 1-5 mm. The size differences are influenced by the duration of the fragmentation process in the aquatic environment, where a longer process results in a smaller size of microplastics. Besides that, UV radiation also plays a role in breaking down larger plastics into smaller particles (Azizah *et al.*, 2020). Differences in microplastic concentrations among sampling sites are caused by variation in anthropogenic activities, sources of microplastic input, and the influence of natural factors such as water currents, wind, and river transport dynamics (Fitriyah *et al.*, 2022).

Based on the results of FTIR analysis on sediments (see Figure 3), MPs particles of polyamides, polystyrene, polypropylene, cellulose acetate, acrylonitrile butadiene styrene, and polyvinyl chloride were found. Microplastics in organisms can interfere with

digestive enzymes that convert nutrients into forms that animals can use for energy. This can slow growth, delay maturation, and reduce reproduction (Trestrail *et al.*, 2021). Microplastics found in organisms may also transfer to humans, who risk exposure by consuming contaminated species. Whole organisms consumed pose a greater risk than those with digestive tracts removed. Potential

human health impacts include skin, breathing, and digestive problems, as well as heart disease, reproductive issues, and cancer (Carbery *et al.*, 2018).

Based on statistical analysis using the Kruskal Wallis Test on sediment, the results show no significant difference in microplastics abundance among the sampling stations, with a significance value of 0.332 ( $p > 0.05$ ).

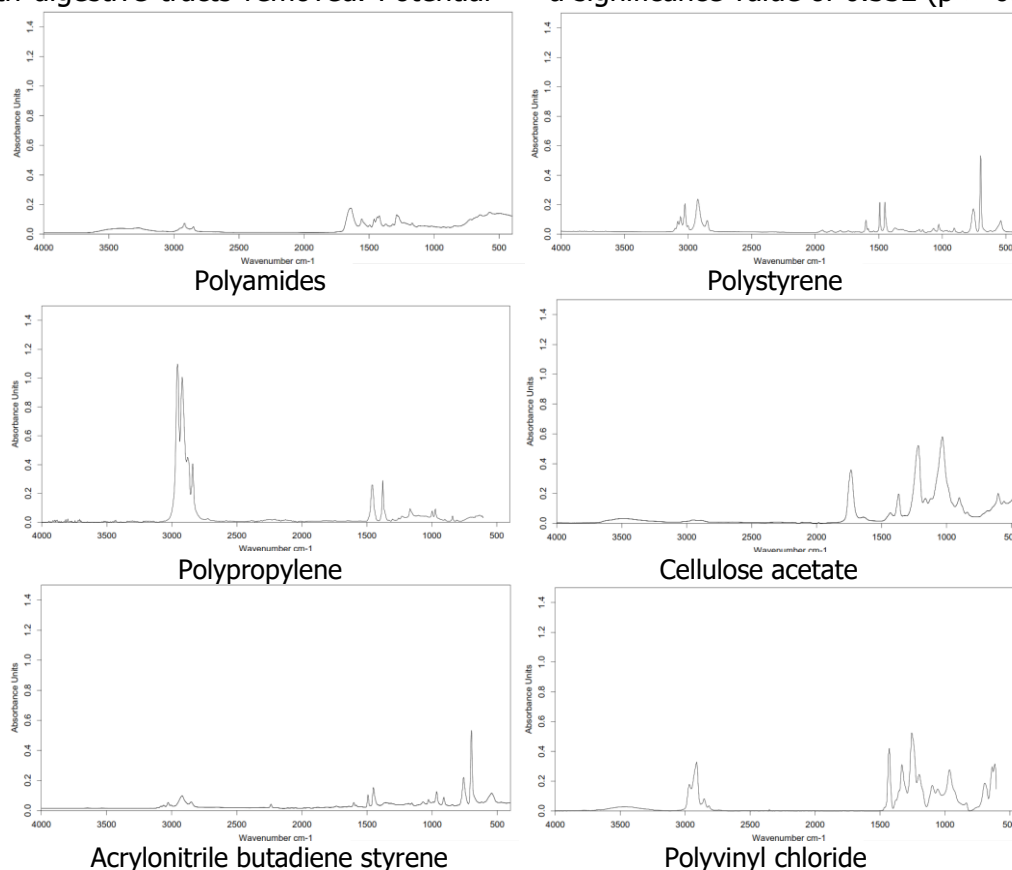


Figure 3. Results of analysis using FTIR on sediment samples

Based on the results of FTIR analysis on sediments (see Figure 3), MPs particles of polyamides, polystyrene, polypropylene, cellulose acetate, acrylonitrile butadiene styrene, and polyvinyl chloride were found. Microplastics in organisms can interfere with digestive enzymes that convert nutrients into forms that animals can use for energy. This can slow growth, delay maturation, and reduce reproduction (Trestrail *et al.*, 2021).

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Based on statistical analysis using the Kruskal Wallis Test on sediment, the results show no significant difference in microplastics abundance among the sampling stations, with a significance value of 0.332 ( $p > 0.05$ ).

### 3.2. Microplastics found in Amazon Sailfin Catfish (*Pterygoplichthys* spp.)

A total of 15 digestive tract samples from Amazon Sailfin catfish (*Pterygoplichthys* spp.) contained 1359 microplastic particles, which were pellets, films, fibers and fragments (see Table 3 and Figure 4).

This study found a significant presence of microplastics in the digestive tract of Amazon Sailfin Catfish (*Pterygoplichthys* spp.) sampled from the middle section of the Citarum River in Karawang, West Java, Indonesia. Fiber was the most abundant type of microplastic found.

In February, an average of  $79.4 \pm 9.9$  microplastic particles per individual was observed across all samples, with the highest concentration recorded in sample 1, suggesting a potential localized source of pollution. In March, the average increased to  $107.2 \pm 58.7$

particles per individual, indicating a notable rise in microplastic abundance; the highest concentration during this period was detected in sample 10. By April, the average decreased slightly to  $85.2 \pm 36.4$  particles per individual, with the peak concentration observed in sample 11. This fluctuation in monthly averages and varying spatial distribution across sampling points highlights the dynamic nature of microplastic contamination in the study area.

Table 3. Summary of data collected from Microplastics in the digestive tract of Amazon sailfin catfish (*Pterygoplichthys* spp.) in February-April 2022.

		Feb	March	April	Total
Average Number of MPs $\pm$ SD (Particles/kg)		79.4 $\pm$ 9.9	85.2 $\pm$ 36.4	107.2 $\pm$ 58.7	90.6 $\pm$ 40.7
Total number of MPs		397	426	536	1,359
Total number of MPs by type of debris	Pellet	60	60	152	272
	Film	14	23	17	54
	Fiber	196	259	187	642
	Fragment	127	84	180	391
Total number of MPs by size (mm)	<0.3	313	329	289	931
	0.3 - < 0.5	48	48	153	249
	0.5 - <1	32	37	88	157
	1-5	4	12	6	22

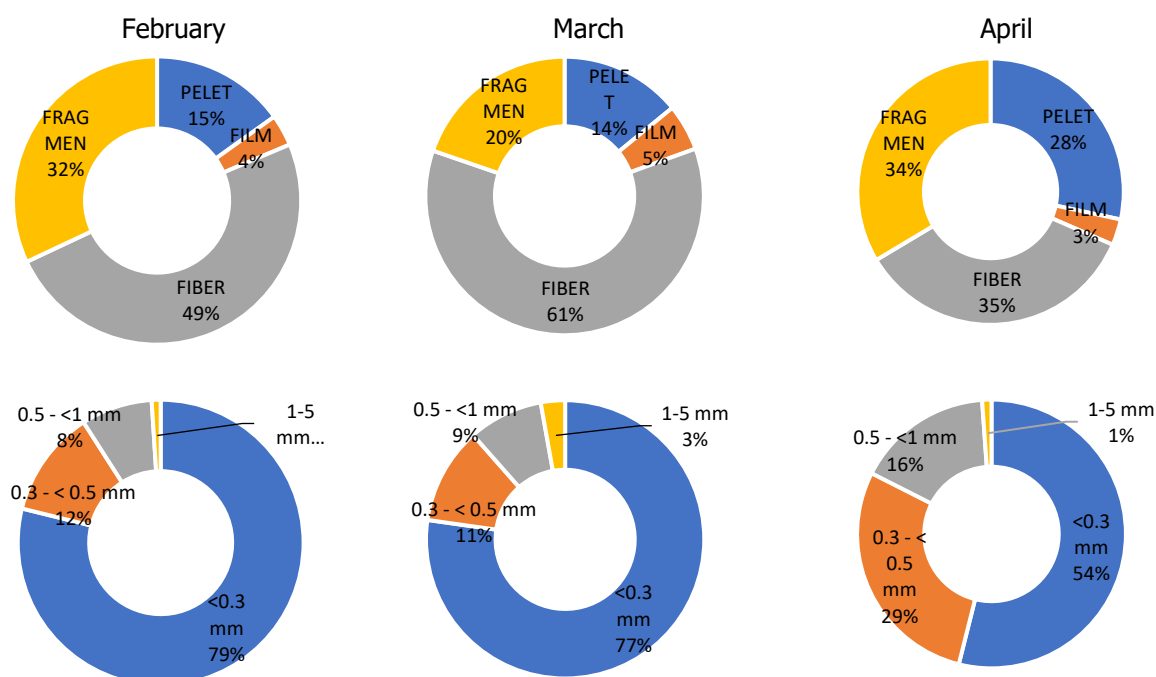


Figure 4. Distribution of microplastic particles by type and size collected from the digestive tract of Amazon Sailfin Catfish (*Pterygoplichthys* spp.) in the middle section of the Citarum River.

From a total of 15 Amazon Sailfin Catfish (*Pterygoplichthys* spp.) samples collected from February-April 2022, a total of 1359 microplastic particles were obtained, with an average total abundance of  $90.6 \pm 40.7$  particles/individual (Table 3). There are 4 types of particles obtained, namely pellets, films, fibers, and fragments, with the largest number of fibers at 47%, fragments at 29%, pellets at 20%, and films at 9%, respectively (Figure 4). In the overall microplastic particle size, sizes

<0.3mm were obtained by 68%, sizes 0.3-<0.5mm by 18%, sizes 0.5-<1mm by 12%, and sizes 1-5mm by 2%, respectively.

The results of FTIR analysis show that the total types of polymers obtained from all research locations in the middle Citarum River are Polyamides, Low-density polyethylene, High-density polyethylene, Ethylene vinyl acetate, Polystyrene, Acrylonitrile butadiene styrene, Polypropylene, Polyvinyl chloride, and Polyethylene terephthalate (Figure 5).

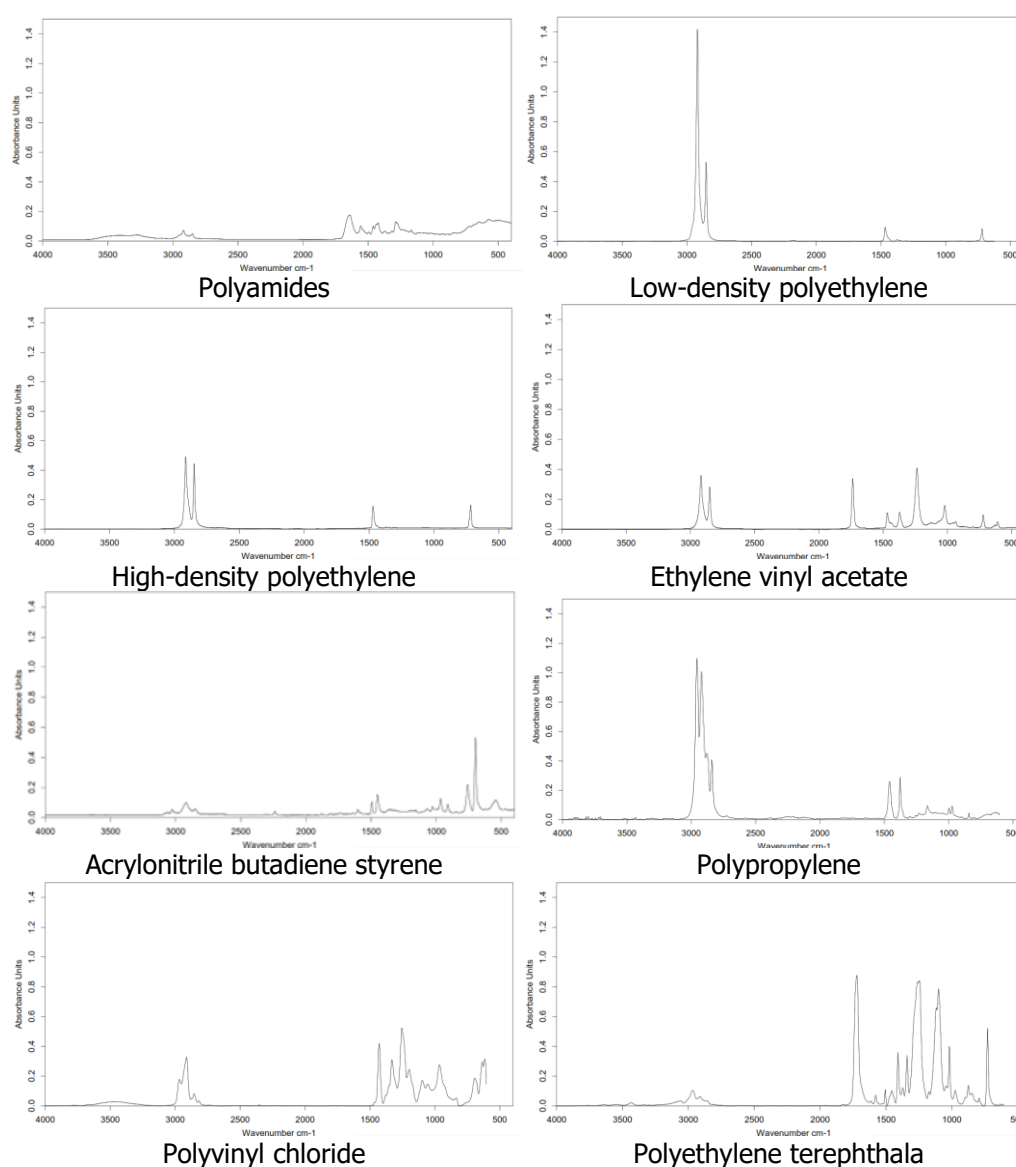


Figure 5. Results of analysis using FTIR on Amazon Sailfin Catfish digestive tract samples  
Amazon Sailfin Catfish

The Amazon Sailfin Catfish (*Pterygoplichthys* spp.) is one of the organisms found in the Citarum River. The Amazon Sailfin Catfish group is native to the South American Amazon

River with the following classification; Class Actiopterygii; Nation Siluriformes; Tribe Loricariidae; Clan: *Pterygoplichthys*; Species: *Pterygoplichthys ambrosettii*, *P. anisitsi*, *P.*



disjunctivus, *P. etentaculatus*, *P. gibbiceps*, *P. joselimaianus*, *P. lituratus*, *P. multiradiatus*, *P. pardalis*, *P. parnaibae*, *P. punctatus*, *P. scrophus*, *P. undecimalis*, *P. weberi*, *P. xinguensis* and *P. zuliaensis* (Wahyudewantoro, 2018), is considered an invasive species (Qoyyimah *et al.*, 2021). They have a remarkable adaptability, making them thrive in polluted waters. Their diet includes insect larvae, benthic algae, algae, detritus, and worms. Due to their small size and buoyancy, fish easily ingest microplastics. Fish ingest microplastics by mistaking the particles as natural prey or accidentally swallowing them. Microplastics can be unintentionally swallowed by fish or by mistake for natural prey (Jovanović, 2017). The presence of microplastics in aquatic organisms can damage the digestive tract, leading to internal injuries and, in severe cases, death. In addition, it may cause blockages in the digestive system, reducing the fish's ability to feed, which may result in starvation and energy depletion. Their accumulation in the digestive tract can also impair growth, lower steroid hormone levels, inhibit enzyme production, and negatively affect reproduction.

The microplastic abundance in the digestive tract of Amazon sailfin catfish in the middle Citarum River was  $90.6 \pm 40.7$  particles per individual. This is relatively lower than the levels found in the digestive tracts of Amazon Sailfin Catfish from the Krukut River, which contain 486 particles per individual (Sandra & Radityaningrum, 2021), as well as the abundance from the Ciliwung River at MT. Haryono (5,344 particles/individual) and at the West Tanjung area (5,581 particles/individual) (Deriano *et al.*, 2021).

## 5. Conclusions

This study investigated the presence and composition of microplastics in the sediment and digestive tract of Amazon catfish from the middle section of the Citarum River, West Java, Indonesia. The results indicate that microplastics were pervasive in the sediment and fish samples examined. Microplastics found in sediment samples were mainly fragments, fibres and films, ranging in size from <0.3mm to 5mm. Interestingly, raising concerns about the potential for trophic transfer and

bioaccumulation of these contaminants within the aquatic food web, the majority of microplastics identified in fish digestive tracts were less than 1 mm in size. The results of this study highlight the urgent need for comprehensive strategies to address plastic pollution, particularly microplastics, in freshwater ecosystems to mitigate the adverse impacts on aquatic organisms and human health.

## Acknowledgment

This research was funded by the ASEANO Project research, Earth and Maritime Research Organization, National Research and Innovation Agency (BRIN). The author would like to thank M.S. Syawal and A. Nurhidayat from the Limnology and Water Resources Research Center - National Research and Innovation Agency, T. Prabowo, M. Aristawidya, and D.A. Hediando from IPB University, and other parties involved in Microplastic research in the middle segment of the Citarum River, Karawang, West Java. The author also expresses his gratitude for the facilities and scientific and technical support from the Cibinong Advanced Characterization Laboratory - Bioproduct Integrated Laboratory, National Research and Innovation Agency, through E-Services Science, National Research and Innovation Agency.

## Author Contributions

**ISH** contributed to research design, collecting, identification and analysis of data. **H**, **SH** and **T** contributed to the survey and evaluated the paper. **GPY** contributed to the paper evaluation and proofreading. **TS** contributed to the identification and paper writing. **E.T** contributed to the collection and preparation. All authors read and approved the final paper.

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## **Efficient Removal of Indigosol Blue Using Activated Carbon from Kepok Banana (*Musa paradisiaca* x *balbisiana*) Peels**

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Received: 2 October 2024; Accepted: 19 May 2025; Published: 20 June 2025

**Abstract:** Indonesia's textile industry has experienced significant growth, yet it faces environmental challenges from dye waste, such as Indigosol Blue, which is classified as hazardous and toxic, requiring special treatment. The use of activated carbon in the adsorption process has proven effective, and the Kepok banana (*Musa paradisiaca* x *balbisiana*) peel, with its high cellulose content, is a potential raw material for activated carbon. This research aims to produce adsorbents from Kepok banana peel, activated by HCl, to adsorb Indigosol Blue using an adsorption column. The production of activated carbon was conducted through carbonization and activation processes. HCl with a concentration of 37% was used as the activator, where the soaking process lasted for 24 hours, and the activation temperature was set at 90°C for 120 minutes. The activated carbon was characterized using UV-Vis Spectrophotometry, Scanning Electron Microscopy (SEM) and Fourier Transform Infrared (FTIR). Based on the results of SEM analysis that illustrates the differences in topography and morphology of kepok banana peel powder before and after activation using various concentrations of activating agents. The results of FTIR analysis, the presence of O-H and C-O bonds indicates that activated carbon derived from kepok banana peel has more polar characteristics. If the initial colorant is increased resulting in higher adsorption efficiency. The results of this study indicated that the optimal wavelength for the Indigosol Blue dye solution, was 624 nm. The best adsorption efficiency was 88.5%, and the maximum adsorption capacity of 0.24 mg/g was obtained with an adsorbent concentration of 30 ppm.

**Keywords:** adsorption, activated carbon, Kepok banana (*Musa paradisiaca* x *balbisiana*) peel, Indigosol Blue dye

DOI: <https://doi.org/10.55981/limnotek.2025.8823>

### **1. Introduction**

The textile industry holds a central position as one of the rapidly growing industrial sectors. Globally, the textile industry generates approximately 700 tons of dye waste each year (Munasir & Ramadiani, 2023). The negative

impact of the textile industry's growth has led to environmental changes through pollution processes. The textile industry is a major contributor to waste in aquatic ecosystems, with the production of dye waste posing potential hazards to living organisms. These



wastes will continue to be generated in large quantities as the textile industry grows. The complex aromatic chemical compounds present in dye waste inhibit the ability of microbes to decompose, often making biological degradation processes difficult. The presence of dye waste also poses health risks to humans (Irawati *et al.*, 2018). Various types of dyes and solvents used in the textile industry have been shown to possess mutagenic and carcinogenic properties when in direct contact with the skin (Oktaviani and Risanti, 2022).

Indigosol Blue is a synthetic pigment commonly used in the textile industry due to its low cost and aesthetic properties. However, according to Government Regulation No. 19 of 1994, waste from the textile industry is classified as hazardous and toxic waste (B3), which refers to industrial or human activity residues that pose a threat to environmental sustainability (Kusuma, 2024). Indigosol Blue contains toxic and carcinogenic substances that pose dangers to both humans and the environment. Treatment processes are required before this waste can be released into the environment. Various technologies have been employed to treat wastewater containing dyes, including reverse osmosis, coagulation, membrane filtration, ozonation processes, chemical oxidation, and biosorption (Septiariva *et al.*, 2021). One of the strategies that can be applied to mitigate pollution from the textile sector is the adsorption technique. The adsorption approach has proven efficient in eliminating waste pigments derived from dye compounds.

Activated carbon is frequently used as a common adsorbent due to its larger surface area compared to other adsorbents. The production of activated carbon can be carried out from various carbon-containing materials, especially those with significant cellulose content (Fitriansyah *et al.*, 2021). Agricultural waste has significant potential to advance wastewater treatment technology due to its abundant availability in the natural environment and the relatively low cost of obtaining it. Additionally, it contains several biochemical elements such as cellulose, hemicellulose, chlorophyll pigments, and pectin substances (Arifiyana and Devianti, 2020).

One material with high cellulose content is the banana peel (*Musa paradisiaca* L.), which contains several chemical elements, including nitrogen, sulfur, and organic substances such as carboxylic acids, cellulose, hemicellulose, chlorophyll pigments, and pectin components containing galacturonic acid, arabinose, galactose, and rhamnose. In general, banana peels are not fully utilized and are often used only as animal feed or discarded. One variety of banana peels with high cellulose content is the Kepok banana peel (*Musa paradisiaca* x *balbisiana*). Kepok banana peels have a higher carbon content compared to other banana varieties, amounting to 44.2% (Septiana, 2023). Kepok banana peels are easier to process into activated carbon, even with simple methods, and they are one of the most commonly consumed varieties in Indonesia (Widyaningsih, 2022). According to the Indonesian Ministry of Agriculture in 2021 (Kementerian Pertanian Republik Indonesia, 2021), the production of Kepok bananas in Indonesia reached 2.5 million tons. With banana peel waste accounting for 20-30% of the total, the amount of Kepok banana peel waste generated is approximately 500,000 to 750,000 tons per year.

Previous studies have indicated that Kepok banana peels are capable of adsorbing methylene orange dye at a rate of 94.2370% with a contact duration of 120 minutes, using 0.15 M HCl as the activator, and an adsorption capacity of 0.0283 mg/g in a batch process (Budiawan, 2021). Based on this background, this research will utilize Indigosol Blue dye as the adsorbed substance in an adsorption column method, as this method offers greater adsorption capacity, higher adsorption efficiency, and can be operated continuously. This research contributes to improving existing methods by evaluating the use of activated carbon from Kepok banana peels in a column adsorption method. Additionally, this research emphasizes sustainability in wastewater treatment by utilizing agricultural waste as a low-cost and easily obtainable adsorbent. The key experimental conditions in this study include optimizing adsorbent mass and contact time to achieve maximum adsorption efficiency.

The aim of this study is to evaluate the effectiveness of Kepok banana peel carbonization in absorbing Indigosol Blue dye. Additionally, this research seeks to analyze the influence of variations in adsorbent mass and contact time on the adsorption capacity of the dye. Furthermore, this study systematically demonstrates that the column adsorption method exhibits higher efficiency compared to other methods in the absorption process of Indigosol Blue.

## 2. Materials and Method

The research was conducted from April to July 2024 at the Integrated Laboratory Unit of UNS, the Biology Laboratory of the Faculty of Mathematics and Natural Sciences (FMIPA) UNS, and the Integrated FMIPA Laboratory UNS. The equipment used included glassware, pestle and mortar, an analytical balance, an oven, and an adsorption column measuring 6 cm in width and 50 cm in height. The characterization instruments used were a UV-Vis Spectrophotometer (Hitachi UH5300, Japan), Fourier Transform Infrared (FTIR, Shimadzu Prestige-21), and Scanning Electron Microscopy (SEM, JEOL\_JSM\_6510). The materials used were kepok banana peel, distilled water, HCl, Indigosol Blue dye, and aluminum foil.

### 2.1 Preparation of Activated Carbon from Kepok Banana Peel

The production of activated carbon from kepok banana peels aims to activate the carbon by expanding its pores through high-temperature heating, thereby increasing the surface area of the carbon. The kepok banana peels are washed and sun-dried for two days. After drying, the banana peels are cut into approximately 2 cm pieces (Saputro *et al.*, 2023). A total of 250 grams of dried banana peels are placed in a 1000 ml beaker, followed by the addition of 1000 ml of HCl solution with varying concentrations of 0.15 M, 0.2 M, and 0.25 M, for a duration of 24 hours. After soaking, the banana peels are filtered and rinsed with distilled water until reaching a neutral pH level. Next, the banana peels are activated in an oven at 90°C for 12 hours.

### 2.2 Preparation of Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) Dye Solution

The preparation of the Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) dye standard solution was carried out with meticulous and precise procedures. The first step involved weighing the Indigosol Blue powder using an analytical balance to ensure accuracy. After weighing, the obtained masses of the Indigosol Blue powder were 15 mg, 20 mg, 25 mg, and 30 mg. The powder was then dissolved in distilled water to a total volume of 1 liter. As a result, the concentrations of the resulting solutions were 15 ppm, 20 ppm, 25 ppm, and 30 ppm.

### 2.3 Adsorption of Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) Dye by Activated Carbon from Kepok Banana Peel

The Adsorption Test of Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) Dye was conducted in a glass aquarium with a rectangular shape, having a diameter of 6 cm and a height of 50 cm. In the initial stage, the adsorbent, which had been activated with HCl solution in concentrations of 0.15 M, 0.2 M, and 0.25 M, was placed in the glass aquarium in the amount of 250 grams with contact times of 60 minutes and 120 minutes. Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) solution with concentrations of 15, 20, 25, and 30 ppm was flowed downwards through the column. Samples of the solution exiting the adsorption column were collected according to the predetermined contact times. The adsorption process results were then analyzed using UV-Vis and FTIR spectrophotometers.

### 2.4 Determination of the Optimum Wavelength (λ) of Indigosol Blue Dye

Standard solutions of Indigosol Blue (C<sub>16</sub>H<sub>10</sub>N<sub>2</sub>O<sub>2</sub>S) dye were produced with varying concentrations of 15, 20, 25, and 30 ppm. Subsequently, the optimum wavelength (λ) of Indigosol Blue dye was determined using spectrophotometry with a UV-Vis spectrophotometer. First, 10 mL of the Indigosol Blue dye solutions at concentrations of 15, 20, 25, and 30 ppm was taken using a micropipette. The absorbance of these solutions was then measured using the UV-Vis spectrophotometer. The absorbance measurements from the UV-Vis spectrophotometer can be taken in the visible wavelength area, specifically within the range of 400-760 nm. This wavelength determination aims to assess the absorption rate in relation to the concentration of the solution.

## 2.5 Calibration Curve Creation Using UV-Vis Spectrophotometry

Standard solutions of Indigosol Blue dye were prepared with varying concentrations of 15, 20, 25, and 30 ppm by dissolving 15, 20, 25, and 30 mg of Indigosol Blue dye in a 1000 mL volumetric flask. The dye was then diluted with distilled water up to the mark and homogenized. Subsequently, the absorbance of the standard solutions of Indigosol Blue dye at these varying concentrations was measured using a UV-Vis spectrophotometer. Based on the absorbance data obtained, a standard curve was constructed, plotting the relationship between absorbance and concentration.

## 2.6 Data Analysis

The data from the variations in contact time and concentration were plotted to determine the optimum contact time and concentration using Excel software:

(a) Determination of Adsorption Capacity (Q):

$$Q = \frac{(C_0 - C_1) \cdot V}{M} \quad \dots \text{Eq.1}$$

description:

Q = adsorption capacity (µg/g adsorbent)

C<sub>0</sub> = initial dye concentration

C<sub>1</sub> = dye concentration after adsorption

V = volume of solution (mL)

M = maximum adsorbent mass (g)

(b) Determination of Freundlich and Langmuir Parameters. The Freundlich parameters were determined by plotting the curve of log  $q_e$  vs log  $C_e$ . The Langmuir parameters were determined by plotting the curve  $\frac{C_e}{q_e}$  vs  $C_e$ .

## 3. Result and Discussion

### 3.1. Production of Activated Carbon

Activated carbon derived from Kepok banana peels through the processes of carbonization and activation results in charcoal, as shown in Figure 1. The carbonization process produces charcoal from Kepok banana peels at a temperature of 90°C, over 12 hours. The activated carbon yield from the banana peels was 50% of the sample weight. From an initial 250 grams of sample, 125 grams of charcoal were obtained after the carbonization process. Temperature plays a significant role in the resulting charcoal. An increase in temperature leads to a reduction in the amount of charcoal formed, as some of the formed charcoal transforms into ash. However, the amount of liquid and gas produced tends to increase with the rise in temperature. Therefore, the temperature used needs to be adjusted according to the type of sample being analyzed.

To enhance the adsorption capacity of the charcoal, an activation process was conducted to expand the pores of the adsorbent. This process involves breaking hydrocarbon bonds or oxidizing surface molecules, which alters the physical and chemical properties of the charcoal. Consequently, the surface area of the adsorbent increases, which enhances the adsorption. The activation process involves the use of inorganic chemicals added to the raw material to reduce or eliminate organic compounds during carbonization or calcination.

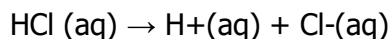


Figure 1. (a). raw materials before treatment, (b). After the carbonization process, (c). After the activation process, and (d) Powders used in the experiment.

This study utilized a chemical activation process by soaking the Kepok banana peels in a 37% HCl solution. Higher concentrations of the activating solution have a greater impact

on binding the residual carbonization compounds, which are released through the micro-pores of the carbon (Qi *et al.*, 2021). This causes the carbon surface to become

more porous, thereby increasing the adsorption capacity of the activated carbon (Heidarinejad *et al.*, 2020). The following reaction occurs during the interaction of the activator with the charcoal in the solution:



During the activation process,  $\text{H}^+$  ions drive out non-volatile substances, such as alkali and alkaline earth ions, that remain on the surface of the charcoal. The high concentration of  $\text{H}^+$  ions replaces the alkali and alkaline earth ions bonded to the charcoal surface, releasing them. As a result, bonds between the carbon and  $\text{H}^+$  ions form on the surface of the activated charcoal.

This study involved soaking Kepok banana peels in an HCl solution for 24 hours. The soaking duration is one of the factors that influence the effectiveness of the activation process, as it affects the formation of lignin. The purpose of soaking in HCl is to dissolve the organic compounds in the charcoal, thereby opening the pores on the surface of the activated carbon and increasing its adsorption capacity.

After the 24-hour soaking period, the charcoal's color changed from dark gray to deep black. The charcoal was then filtered using Whatman filter paper to ensure that any remaining activator or residue was removed. Afterward, the drying process was carried out in an oven at  $90^\circ\text{C}$  for 12 hours. The purpose of this drying process was to reduce the remaining water content in the charcoal so that the final product was only activated carbon that had undergone the activation process.

### 3.2. Determination of the Optimum Wavelength ( $\lambda$ ) of Indigosol Blue Dye

The determination of the optimal wavelength for the Indigosol Blue solution aims to find the wavelength used to measure the absorbance of the solution. At the optimal wavelength, maximum absorbance is achieved, allowing accurate measurements even when the solution has low or dilute concentrations.

Based on Figure 2, the optimal wavelength for Indigosol Blue is 624 nm. The wavelength range of 400-800 nm was selected because the complementary color of Indigosol Blue is

blue, which has a light spectrum within the visible range between 500-650 nm (Fitriansyah *et al.*, 2021). Therefore, the optimal wavelength was taken from that range and will be used in subsequent absorbance measurements using a UV-Vis spectrophotometer.

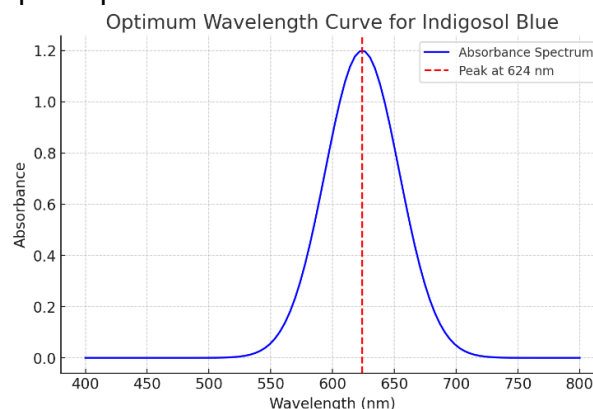


Figure 2. Curve for Determining the Optimal Wavelength for Indigosol Blue Solution

### 3.3 Preparation of the Standard Curve for Indigosol Blue Dye Solution

The calibration curve for varying concentrations, both high and low, was measured using the optimal wavelength. For the Indigosol Blue solution, the optimal wavelength is 624 nm.

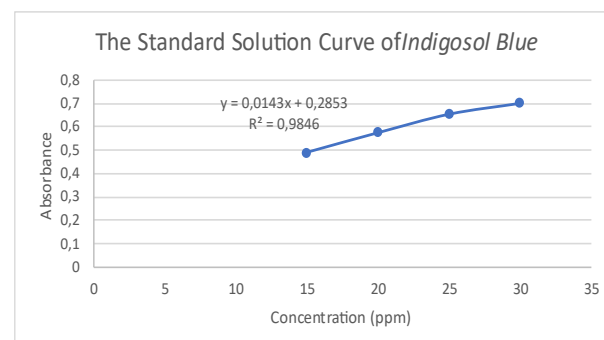


Figure 3. Calibration Curve for Indigosol Blue Solution

In Figure 3, the calibration curve shows the relationship between the concentration and absorbance of the Indigosol Blue solution at various concentration levels. The correlation coefficient ( $R^2$ ) for the solution ranges from 0.9 to 1, meeting the criteria for a standard curve. The curve exhibits an excellent correlation, approaching a value of 1, and forms a straight line with a positive gradient. Therefore, the resulting regression equation can be used to measure the concentration of



the dye solution during optimization processes as well as in subsequent adsorption activities.

### 3.4. Characterization of Activated Carbon using SEM

SEM analysis is applied to examine the topography of the activated carbon, including surface analysis and the texture of the produced activated carbon. The results of the SEM analysis can be seen in Figure 4, which depicts the differences in topography and morphology of the kepok banana peel powder before and after activation using various concentrations of activating agents. Figure 3(a) shows the kepok banana peel before the activation process, with a magnification of 3000x. In this image, the surface of the banana peel appears denser, and the pores are still closed. This indicates the presence of organic matrices that remain abundant and have not completely decomposed during the carbonization process, as well as the possibility that impurities were not fully removed during carbonization, potentially affecting the adsorption.

Figures 4(b), 3(c), and 3(d) depict the surface morphology of banana peel after the activation process using HCl at different

concentrations. Figure 3(b) shows banana peel activated with 0.15 M HCl, while figure 3(c) presents banana peel activated with 0.2 M HCl, and figure 4(d) illustrates banana peel activated with 0.25 M HCl. The SEM sample testing was conducted at a magnification of 3,000x, with visualizations of the pores in the activated carbon from the kepok banana peel before and after activation using HCl. The results show that the pores in the activated kepok banana peel sample became larger and more open. This is consistent with the research by Wardani *et al.* (2018), which explains that as the pore size of activated carbon increases after activation, the adsorption efficiency also increases. The comparison among the three samples activated with different HCl concentrations, as shown in figures 4(b), 4(c), and 4(d), demonstrates a tendency for an increase in pore size and number as the acid concentration increases. Kepok banana peel activated with 0.15 M HCl (Figure 4(b)) exhibits a more open pore structure and contains more cavities compared to kepok banana peel activated with 0.2 M HCl (Figure 4(c)) and 0.25 M HCl (Figure 4(d)).

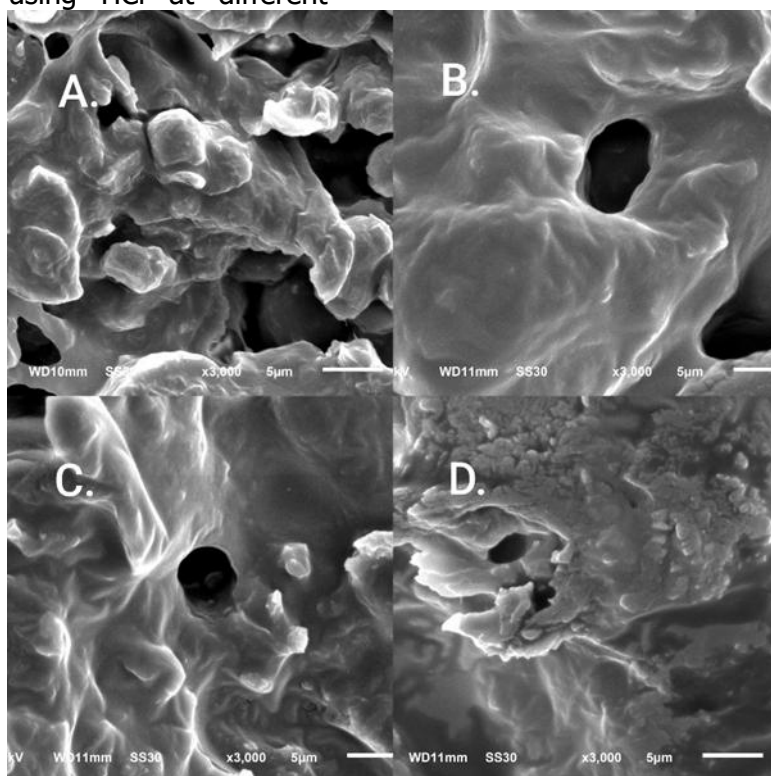


Figure 4. SEM images of Kepok banana peel powder before activation (a), after activation with 0.15 M HCl (b), after activation with 0,2 M HCl (c), and after activation with 0,25 M HCl (d)

The sample activated with 0.15 M HCl showed more uniform and evenly distributed pore formation compared to the samples activated with higher HCl concentrations. This suggests that overly high concentrations of HCl can lead to excessive dissolution of the carbon structure, ultimately reducing the material's specific surface area. Therefore, 0.15 M HCl is the recommended concentration for optimizing pore structure.

### 3.5. Characterization of Activated Carbon using FTIR

Based on the FTIR analysis results for the kepok banana peel adsorbent before activation (non-activated) and after activation (0.15M, 0.2M, and 0.25M), as shown in Figure 5, significant peaks were observed at 3481, 2927, 2856, 1736, 1458, 1374, and 885  $\text{cm}^{-1}$  within the wavenumber range of 4000 to 500  $\text{cm}^{-1}$ . The increase in HCl concentration enhances pore formation, expands surface area, and modifies functional groups, contributing to increased adsorption capacity. FTIR analysis shows differences in peak intensity and position, particularly in the O-H, C=O, and C-H groups, indicating structural changes due to acid treatment. The peaks at 3481 and 2927  $\text{cm}^{-1}$

correspond to aliphatic C-H stretching, indicating the presence of compounds containing methyl or methylene groups. The strong peak at 1736  $\text{cm}^{-1}$  indicates the presence of a carbonyl group (C=O), which typically appears in compounds such as esters, aldehydes, or ketones. The peak at 1458  $\text{cm}^{-1}$  suggests the possible presence of C-H bending vibrations associated with aliphatic bonds. The peak at 1374  $\text{cm}^{-1}$  may indicate the bending vibration of C-H in methyl groups. The lower peak at 885  $\text{cm}^{-1}$  indicates the out-of-plane bending of aromatic C-H, confirming the presence of aromatic rings within the activated carbon matrix.

The presence of O-H and C-O bonds suggests that the activated carbon derived from kepok banana peel has more polar characteristics, which enhances its adsorption capability for dye molecules containing hydrophilic functional groups (Das & Kalyani, 2023). The high intensity peaks related to oxygenated functional groups indicate the potential contribution of surface chemistry in adsorption mechanisms, particularly in hydrogen bonding and electrostatic interactions (Suhaimin *et al.*, 2022).

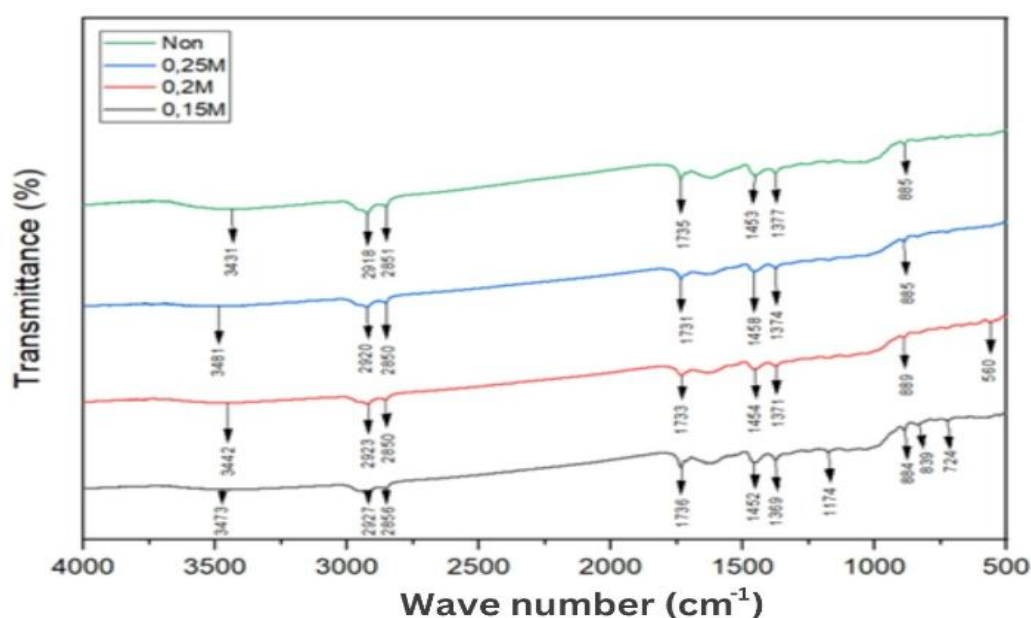


Figure 5. FTIR Test Results of Kepok Banana Peel

Studies have also shown that the presence of oxygen containing functional groups, such as hydroxyl and carboxyl groups, improves the adsorption of organic dyes due to increased

surface polarity (Yang *et al.*, 2021). Furthermore, the functional groups identified in the FTIR spectra indicate that the activated carbon underwent successful activation, as

evidenced by the enhancement of oxygenated surface groups. The presence of a strong carbonyl peak at  $1736\text{ cm}^{-1}$  is consistent with previous studies on biomass-derived activated carbon, which report the formation of carboxyl and lactone groups after chemical activation (Nazir *et al.*, 2023). These functional groups contribute to the adsorption efficiency by providing additional active sites for dye interaction. The adsorption of Indigosol Blue increases with higher HCl concentrations, with the highest efficiency at 0.25 M. This indicates that activation with 0.25 M HCl produces activated carbon with optimal adsorption properties, making it more effective in removing dye from aqueous solutions.

### 3.6. Adsorption Efficiency

Based on the graph, it can be observed that the adsorption efficiency of Indigosol Blue by kepok banana peel activated carbon treated with HCL at a concentration of 0,25M increases as the initial dye concentration increases. This indicates that at low initial concentrations, more active sites on the activated carbon are available to bind the dye. As the initial dye concentration increases, more dye molecules are bound to the activated carbon, resulting in higher adsorption efficiency. The highest adsorption efficiency reached 88.5% at an initial concentration of 30 ppm, indicating that activated carbon from kepok banana peel is effective in adsorbing Indigosol Blue dye, at low initial concentrations (Figure 6).

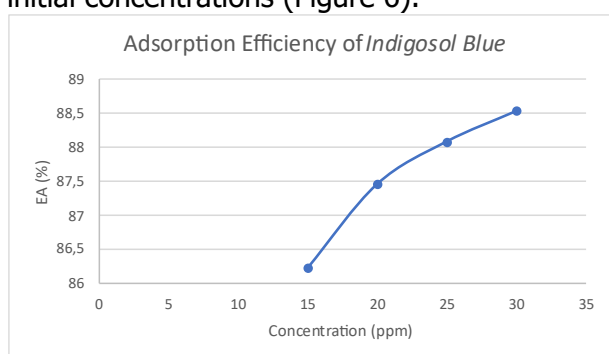


Figure 6. Adsorption Efficiency of Indigosol Blue Dye

The maximum adsorption capacity ( $q_e \text{ max}$ ) is the highest  $q_e$  value that can be achieved at a specific initial Indigosol Blue concentration. In the Figure 7,  $q_e \text{ max}$  is reached at an initial Indigosol Blue concentration of 35 ppm, with a  $q_e \text{ max}$  value of approximately 0.24 mg/g. The

$q_e \text{ max}$  value can be used to estimate the amount of activated carbon needed to adsorb Indigosol Blue from a solution with a specific initial concentration. The higher the  $q_e \text{ max}$  value, the less activated carbon is required to achieve the desired adsorption efficiency. Based on the above analysis, it can be concluded that kepok banana peel activated carbon treated with HCL at a concentration of 0,25M can be used effectively to adsorb Indigosol Blue. The adsorption capacity of Indigosol Blue by kepok banana peel activated carbon increases with increasing initial Indigosol Blue concentration. The  $q_e \text{ max}$  value in the figure is approximately 0.24 mg/g. The relationship between the initial Indigosol Blue concentration and the adsorption capacity of Indigosol Blue by kepok banana peel activated carbon can be modeled using the Langmuir equation (Figure 8).

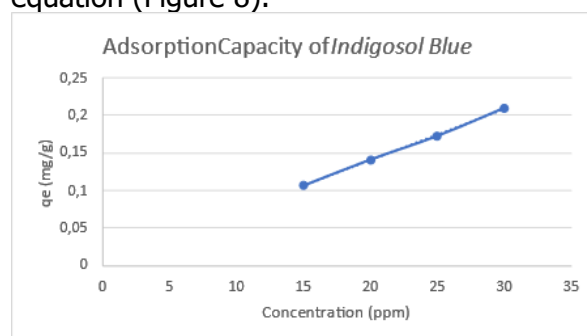


Figure 7. Adsorption Capacity of Indigosol Blue Dye

### 3.7. Adsorption Isotherms

Various concentrations of 15, 20, 25, and 30 ppm were prepared and applied at optimal contact time. In this step, activated carbon samples treated with HCL at 0,25M were used to evaluate their adsorption performance. An isotherm describes the equilibrium relationship between the adsorbate in the solid and liquid phases. Adsorption isotherm determination was carried out to study the adsorption mechanism between kepok banana peel activated carbon and Indigosol Blue adsorbate.

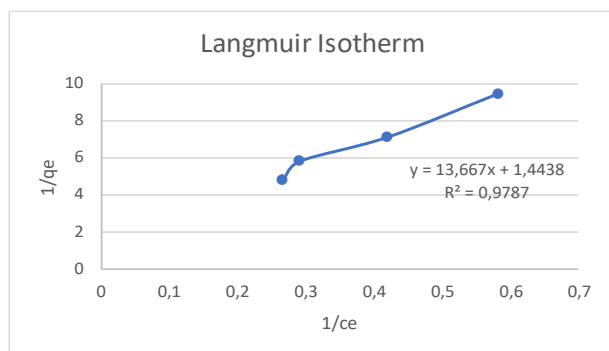


Figure 8. Langmuir Isotherm of Kepok Banana Peel Adsorbent

To assess the appropriate isotherm model, Langmuir and Freundlich isotherm models were applied to study the adsorption pattern of Indigosol Blue dye on the surface of activated carbon. The Langmuir isotherm equation is illustrated with a graph of  $C_e/q_e$  versus  $C_e$ , while a graph of  $\log q_e$  vs.  $\log C_e$  was created based on the Freundlich isotherm equation. From the analysis of both isotherms, a comparison between the Langmuir and Freundlich isotherms for kepok banana peel adsorbent was obtained, as shown in Figures 8 and 9.

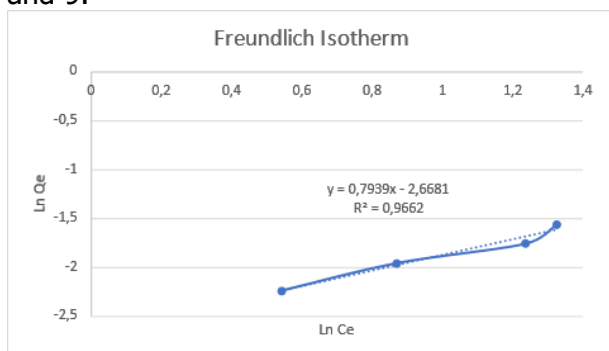


Figure 9. Freundlich Isotherm of Kepok Banana Peel Adsorbent

Figures 8 and 9 display curves representing the adsorption isotherms of kepok banana peel activated carbon for Indigosol Blue dye solution. The isotherm model that best fits the research data was analyzed using simple linear regression, considering the correlation coefficient ( $R^2$ ) values to determine the contribution of the variables. Based on the linear regression coefficient ( $R^2$ ) values from each graph, the Langmuir isotherm graph shows an  $R^2$  value almost similar to the Freundlich isotherm graph, which are 0.97 and 0.96, respectively. This indicates that the

adsorption of Indigosol Blue dye on the surface of activated carbon follows both isotherm patterns. However, it should be noted that the closer the  $R^2$  value is to 1, the greater the influence between variables, indicating a better relationship. Referring to the data shown in the figures, the adsorption characteristics of kepok banana peel adsorbent appear to better match the Langmuir isotherm, as the correlation coefficient ( $R^2$ ) obtained is higher and closer to 1 compared to the Freundlich isotherm. The different models assume different adsorption mechanisms.

The effectiveness of activated carbon derived from kepok banana peels in adsorbing Indigosol Blue dye is also reflected in its high adsorption capacity, reaching 88.5%. This reinforces the conclusion that adsorption occurs in a single layer (monolayer) and that the surface is homogeneous, as assumed by the Langmuir model. Each pore of the activated carbon can only absorb a single dye molecule, further strengthening the efficient adsorption characteristics. The linear equation of the Langmuir isotherm curve is used to identify Langmuir parameters, such as  $K_L$  (Langmuir Equilibrium Constant), which serves as a key indicator in determining the efficiency of this adsorption process. Beyond its theoretical contributions, these findings also have significant practical implications for aquatic ecosystem restoration. Activated carbon from banana peels can be applied through floating adsorption units or integrated into biological filters in water channels and small rivers. These units are expected to treat up to 500 liters of water per day per unit and reduce dye concentrations by up to 90% after a single usage cycle. A study by Harimu *et al.* (2020) found that using this adsorbent in natural waters can increase.

#### 4. Conclusion

This study aimed to evaluate the adsorption efficiency of activated carbon derived from Kepok banana peels in removing Indigosol Blue from wastewater. The findings demonstrate that activated carbon treated with 0.25M HCl can achieve an impressive adsorption efficiency of 88.5% at an initial dye concentration of 30 ppm. The optimal adsorption conditions were determined to be an adsorbent mass of 125



grams and a contact time of 120 minutes, ensuring maximum dye removal. These findings contribute to the development of sustainable, low-cost adsorbents for industrial wastewater treatment, particularly in reducing aquatic environmental pollution. By utilizing agricultural waste as a viable adsorbent, this research aligns with broader efforts to enhance eco-friendly wastewater treatment technologies.

Additionally, this study builds upon previous research by demonstrating the potential of banana peel-derived activated carbon in continuous adsorption systems. The column adsorption method exhibited a maximum adsorption capacity ( $q_e$  max) of 0.24 mg/g, suggesting its effectiveness in treating wastewater containing Indigosol Blue. Despite its promising results, this study acknowledges certain limitations. Factors such as adsorbent regeneration and long-term performance require further investigation. Future research should explore modifications to the adsorbent surface properties and conduct pilot-scale studies to assess its feasibility and scalability for industrial applications. In conclusion, this study highlights the potential of Kepok banana peel-derived activated carbon as an efficient and sustainable adsorbent for dye removal. Further advancements in adsorption techniques and process optimization could enhance its applicability in real-world wastewater treatment scenarios.

### Funding Agencies

This research was funded by the Directorate General of Learning and Student Affairs (Belmawa) of the Ministry of Education, Culture, Research, and Technology of the Republic of Indonesia, as well as supported by funding from the associated university. We express our gratitude for the financial support that made this study possible.

### Conflict of interests

The authors declare that there are no conflicts of interest related to this study. All research activities were conducted with integrity and transparency, ensuring that personal or financial relationships did not influence the results or interpretation of the data. Any potential conflicts have been

addressed, and the authors affirm their commitment to ethical research practices.

### Data availability statement

To enhance the scientific credibility of this study, all data generated or analyzed during this research are available upon reasonable request from the corresponding author. The datasets include all relevant experimental results, methodology details, and supplementary information. There are no sensitive data involved in this project, ensuring full transparency and accessibility for further research.

### Acknowledgment

The authors wish to acknowledge the significant contributions of individuals and institutions that facilitated this research. We extend our gratitude to the personnel at the PKM Center of Universitas Sebelas Maret for their essential support and collaboration throughout the study. Furthermore, we recognize the financial assistance provided by the Directorate General of Learning and Student Affairs (Belmawa) of the Ministry of Education, Culture, Research, and Technology of the Republic of Indonesia, as well as the funding from Universitas Sebelas Maret. Their commitment to advancing research in this field has been instrumental in the successful completion of this project.

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## **The Effect of Land Cover Changes on Water Quality in the Cisadane River Basin, West Java, Indonesia**

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Received: 28 November 2024; Accepted: 19 May 2025; Published: 20 June 2025

**Abstract:** The availability of water and clean water quality are increasingly affected by population growth. As the human population expands, the demand for goods and services rises, often leading to intensified in water use and pollution. This study examines the correlation between land cover changes and water quality in the Cisadane River Basin. Water quality data were collected as a time series from 2019 to 2022, focusing on eight parameters: TDS, TSS, BOD<sub>5</sub>, COD, DO, TP, NO<sub>3</sub>-N, and NH<sub>3</sub>-N. Water quality was classified using the Pollution Index (IP) method. Land cover changes were visually interpreted using ArcGIS at a 1:10,000 scale. Multiple linear regression analysis was used to assess the influence of land cover on river water quality. The results indicate that the overall water quality in the Cisadane River Basin falls under the 'slightly polluted' category, with noticeable influence from land cover changes. Between 2019 and 2022, built-up land increased by approximately 5.5%, contributing to elevated BOD<sub>5</sub> levels in several locations, such as Babakan and Genteng, indicating rising organic pollution from domestic waste and urban activities. Conversely, a decrease in vegetation cover by around 6.3% at various observation points correlated with decreased DO levels, especially in downstream areas such as Tanjung Burung and Vihara, potentially disrupting aquatic ecosystem balance. These findings suggest that vegetation cover plays an important role in maintaining water quality and ecological stability, while the expansion of built-up and agricultural areas tends to elevate pollutant loads in river water. Therefore, the results of this study can serve as a reference for policymakers in spatial planning of the Cisadane River Basin, especially in developing land conservation and waste management strategies to improve water quality and maintain the sustainability of water resources in the region.

**Keywords:** Cisadane River Basin, Land Cover Changes, Multiple Linear Regression Analysis, Pollution Index, Water Quality

DOI: <https://doi.org/10.55981/limnotek.2025.8944>

### **1. Introduction**

The increasing demand for clean water continues to pose a challenge alongside population growth (Sasongko et al. 2014). Population growth not only escalates the need for water but also heightens the risk of water pollution due to waste from industrial and domestic activities. Improperly managed waste

contains pollutants that flow into water bodies, thereby degrading water quality (Effendi et al., 2021; Syawal et al., 2016). Furthermore, population growth drives changes in land use, such as the conversion of natural vegetation into built-up or agricultural areas, which contribute to increased pollutant loads in water systems (Chu and Yu 2002; Kemp 2004).

In river basin areas, the impacts of land cover changes are significant. Studies by Effendi (2016, 2017) and Effendi et al. (2015, 2018) demonstrate that human activities altering land cover around river basins are closely associated with water pollution, as evidenced by the decline in various water quality indicators (Effendi, 2016, 2017; Effendi et al., 2015, 2018). The Cisadane River Basin, one of Indonesia's national priority river basins, has had significant land cover changes over the years due to economic activities and population growth (Dawud et al. 2016). The community's increasing demand for land for infrastructure and settlements has led to land degradation, ultimately reducing both the quantity and quality of water in the Cisadane River Basin. This, in turn, affects the watershed's capacity to meet community water needs and increases the pollution burden (Suwari et al, 2011, Yusuf et al., 2021).

Previous studies have established a relationship between land cover and water quality in the Cisadane river basin. Iqtashada and Febrita (2023) found that changes in land cover around the Cisadane River significantly affected water quality, particularly with increased Total Suspended Solids (TSS) and Nitrite levels in Bogor City (Iqtashada and Febrita 2023). Similarly, Nilda et al. (2015) reported that the conversion of vegetation into built-up areas directly impacted water quantity and exacerbated pollution, leading to a decline in water quality in the upstream regions of the Cisadane River Basin (Nilda et al. 2015). In contrast to previous studies by Iqtashada & Febrita (2023) and Nilda et al. (2015), this study includes a more comprehensive analysis of the entire Cisadane River Basin area, including upstream, middle, and downstream, thus providing a more holistic picture of the impact of land cover changes on water quality.

This study aims to conduct a more comprehensive analysis of the correlation between land cover and water quality in the Cisadane River Basin, covering the upstream,

middle, and downstream areas. This study focuses on the effect of land cover variations on water quality in the Cisadane River Basin. Understanding the relationship between land cover change and water quality is essential to support environmental sustainability and effective water management policies, especially in areas under pressure from population growth and economic activity. This knowledge is not only relevant for water resource management, but also for broader environmental conservation and rehabilitation efforts. By understanding these dynamics, stakeholders can take more appropriate and informed actions to maintain water quality and ecosystem sustainability. This analysis uses a regression analysis approach, which effectively identifies relationship patterns between variables. This method is very useful in identifying patterns and trends in data and providing insight into how variations in land cover contribute to changes in water quality (Hamidi & Kamulyan, 2022; Wariunsora et al., 2024). This study is expected to provide a more comprehensive understanding of the effect of various land cover classes on water quality, and the results can be a basis for stakeholders in making more effective and sustainable Cisadane River Basin management policies.

## **2. Materials and Method**

### **2.1 Time and Location of Study**

The study was conducted in the Cisadane River Basin, located between 6°72' to 6°76' S and 106°58' to 106°51' E. Administratively, the Cisadane River Basin spans 518 villages across 44 sub-districts in five regencies/cities: Bogor Regency, Bogor City, Tangerang Regency, Tangerang City and South Tangerang City. The research took place from October 2023 to April 2024. The data used in this study included spatial and temporal water quality test results from the past four years and land cover data surrounding the Cisadane River Basin. The research location can be seen in Figure 1.



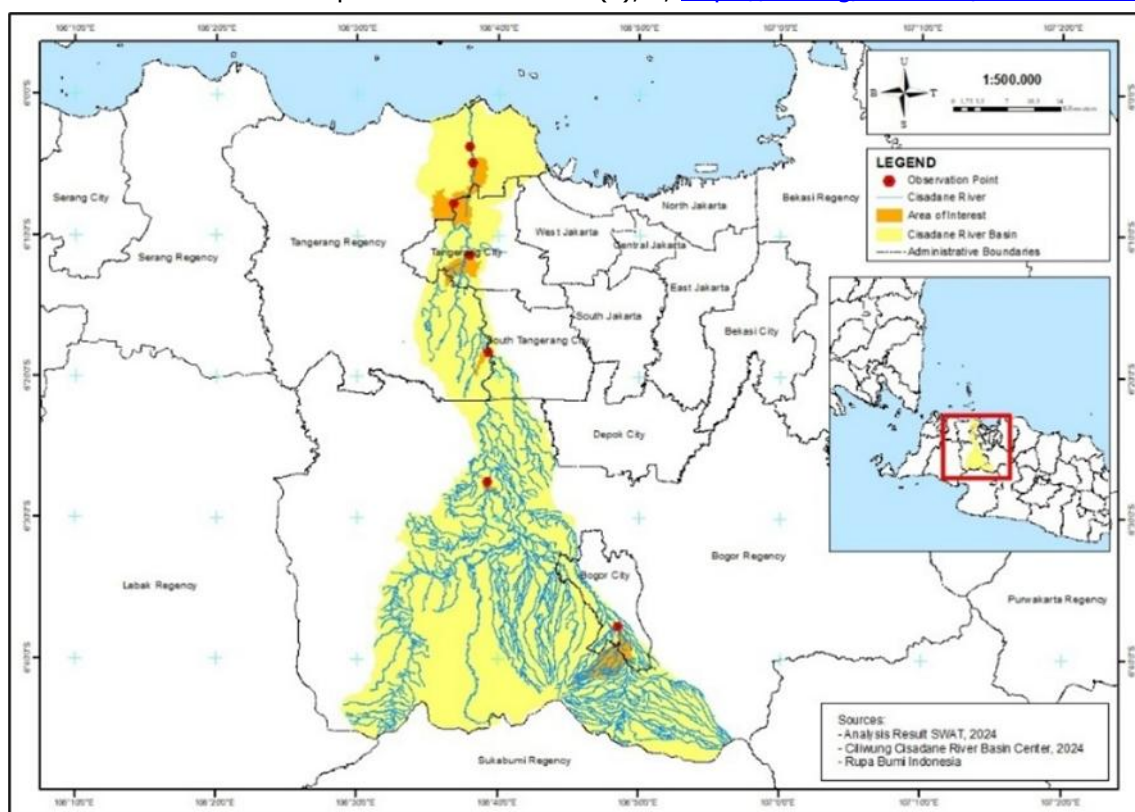


Figure 1. Study area map showing the Cisadane River Basin, Indonesia, including sampling points and watershed boundaries.

## 2.2 Water Quality

Water quality data were obtained Ciliwung Cisadane River Basin Center and Tangerang Regency Environmental and Sanitation Service. The data used in the study covered the period from 2019 to 2022 and included eight water quality parameters: Total Dissolved Solids

(TDS), Total Suspended Solids (TSS), Biochemical Oxygen Demand (BOD5), Chemical Oxygen Demand (COD), Dissolved Oxygen (DO), Total Phosphate (TP), NO<sub>3</sub>-N, NH<sub>3</sub>-N. The study analyzed water quality at seven observation points along the Cisadane River, as can be seen in Table 1.

Table 1. List of observation points with corresponding sub-districts and regency/city in the Cisadane River Basin.

Observation Point*	Sub-district	Regency/City
Genteng	South Bogor	Bogor City
Batu Beulah	Ciseeng	Bogor Regency
Serpong	Cisauk	Tangerang Regency
Babakan	Tangerang	Tangerang Regency
Kedaung	Neglasari	Tangerang Regency
Vihara	Teluk Naga	Tangerang Regency
Tanjung Burung	Teluk Naga	Tangerang Regency

\*) sorted from upstream to downstream

The determination of the Pollution Index (PI) value is based on the Decree of the Minister of Environment No. 115 of 2003, which evaluates water quality conditions by measuring and/or testing specific parameters using established methods and comparing the

results with water quality standards. The water quality standards referenced in this study align with Government Regulation of the Republic of Indonesia Number 22 of 2021, attachment VI for Class II River Water Quality Standards. Class II standards apply to water designated for

recreational facilities, freshwater fish farms, livestock, irrigation, and similar uses. The Pollution Index calculation in this study was conducted at seven observation points using eight water quality parameters: TDS, TSS, BOD<sub>5</sub>, COD, DO, TP, NO<sub>3</sub>-N, and NH<sub>3</sub>-N. The Pollution Index value is determined using the following equation:

$$P_j = \sqrt{\frac{\left(\frac{C_i}{L_{ij}}\right)_M^2 + \left(\frac{C_i}{L_{ij}}\right)_R^2}{2}} \quad \dots \text{Eq 1}$$

Where  $P_j$  is the Pollution Index,  $C_i$  is the concentration of water quality parameter  $i$ ,  $L_{ij}$  is the concentration of water quality parameter  $i$  as specified in water quality standard  $j$ ,  $M$  is the maximum value of the pollution index,  $R$  is the average value of the pollution index. Determination of water status based on the classification of pollution index values refers to the Decree of the Indonesia Minister of Environment No. 115 of 2003 concerning Guidelines for Determining Water Quality Status, as shown in Table 2.

Table 2. Methods Used for Water Quality Parameter Analysis in the Cisadane River by DLHK Tangerang Regency and BBWS Ciliwung Cisadane

Parameter (unit)	DLHK Kab. Tangerang	BBWS Ciliwung Cisadane
TDS (mg/L)	IKM.KHT-27 (Electrometry)	LAB/IK/KIM-ENV/61
TSS (mg/L)	IKM.KHT-41 (Spectrophotometry)	SNI 6989.3 : 2019
BOD <sub>5</sub> (mg/L)	SNI 6989.72:2009	APHA 5210 B 2012
COD (mg/L)	SNI 6989.2:2009	SNI 6989.2:2009
DO (mg/L)	SNI 06-6989.14-2004	LAB/IK/KIM-ENV/88
Total P (mg/L)	IKM.KHT-61 (Spectrophotometry)	SNI 06 - 6989.31 - : 2005
NO <sub>3</sub> -N (mg/L)	IKM.KHT-22 (Spectrophotometry)	SNI 06- 6989.79 - : 2011
NH <sub>3</sub> -N (mg/L)	SNI 06-6989.30-2005	SNI 06- 6989.30 - 2005

### 2.3 Multi-temporal Land Cover

The image data used for land cover classification were obtained from Maxar Technologies for 2019-2022. Maxar Technologies is one of the satellite used by Google. This data was obtained from <https://earth.google.com/>, and it is open to the public. Land Cover classification was performed using a visual interpretation method, with delineation of each land cover class conducted in ArcGIS software at a scale of 1:10,000. The visual interpretation analysis method has advantages, including being able to analyze images with high spatial resolution and having good knowledge of conditions in the field (Kosasih *et al.*, 2019). The land cover types were categorized into five classes: water bodies, open land, built-up land, agricultural land and vegetation. The classification of each land cover type followed the SNI 7654:2010 Land Cover Classification standards. Before determining land cover, an Area of Interest (AOI) was defined to identify areas directly affecting water quality at the observation points. The AOI determination was conducted using ArcSWAT software, with input data including

Digital Elevation Model (DEM) and the flow path of the Cisadane River. During the AOI delineation process, model outlet points were set at each water quality observation point. In this study, the AOI was restricted to areas with a direct impact on water quality measurement at the observation point, ensuring the selected region are those in direct contact with the research observation points.

### 2.4 Variable Selection

Before conducting the regression analysis, a classical assumption test was performed. Classical assumption testing is an essential step preceding linear regression analysis (Masiaga *et al.* 2022). The tests conducted included multicollinearity, heteroscedasticity, normality, and autocorrelation test. These tests aim to ensure the accuracy and reliability of the regression equation in estimating relationships (Alita *et al.* 2021; Nurcahya *et al.* 2024). These tests ensure the regression model meets the necessary assumptions for valid and reliable analysis.

## 2.5 Regression Analysis

The regression analysis in this study was conducted using SPSS 225 software. Each water quality parameter was a dependent variable, while land cover data were used as independent variables. The regression analysis involved eight dependent variables: TDS, TSS, BOD5, COD, DO, TP, NO3-N, NH3-N. The regression method applied was multiple linear regression analysis. The regression model equation used in this study is as follows:

$$Y = \alpha + \beta_0 + \beta_1X_1 + \beta_2X_2 + \beta_3X_3 + \dots + \beta_kX_k + \epsilon_i \quad \dots \text{Eq 2}$$

where Y is water quality parameters (mg/l),  $\alpha$  is constant or intercept of the regression model,  $\beta_1 - \beta_5$  is the regression coefficient for each independent variable, X1 is water bodies, X2 is agricultural land, X3 is built-up land, X4 is open land, X5 is vegetation, and  $\epsilon_i$  is error term representing unobserved factors.

## 3. Results and Discussion

### 3.1. Variable Selection

#### a. Multicollinearity Test

Based on the test results, all independent variables meet the multicollinearity criteria, with VIF values below 10 and tolerance values above 0.10. This indicates that there is no multicollinearity present in the data, confirming that the independent variables are sufficiently independent for further regression analysis (Ainiyah et al. 2016). The detailed results of the multicollinearity test are presented in Table 3.

Table 3. Multicollinearity Test Results

Model	Collinearity Statistics	
	Tolerance	VIF
Water bodies	0,181	5,515
Agricultural land	0,202	4,958
Built-up land	0,104	9,635
Open land	0,125	7,992
Vegetation	0,635	1,576

#### b. Heteroscedasticity Test

The heteroscedasticity test was conducted by analyzing the scatterplot for each variable. If the points on the scatterplot are randomly distributed and do not form a specific pattern, it can be concluded that there is no heteroscedasticity in the regression model, indicating the absence of residual variance similarity across observations (Purba et al. 2021). Based on the test results, the scatter plot shows no discernible pattern in the distribution of points. Therefore, it can be concluded that the data do not exhibit heteroscedasticity. The scatterplot results of the heteroscedasticity test are presented in the Figure 2.

#### c. Normality Test

The normality test results in data processing indicate that the histogram of each variable forms a perfect bell curve. This suggests that the data for each variable are normally distributed. These findings align with previous research stating that a perfect bell-shaped histogram signifies normally distributed data (Purba et al. 2021). The histogram obtained from the normality test is presented in Figure 3.

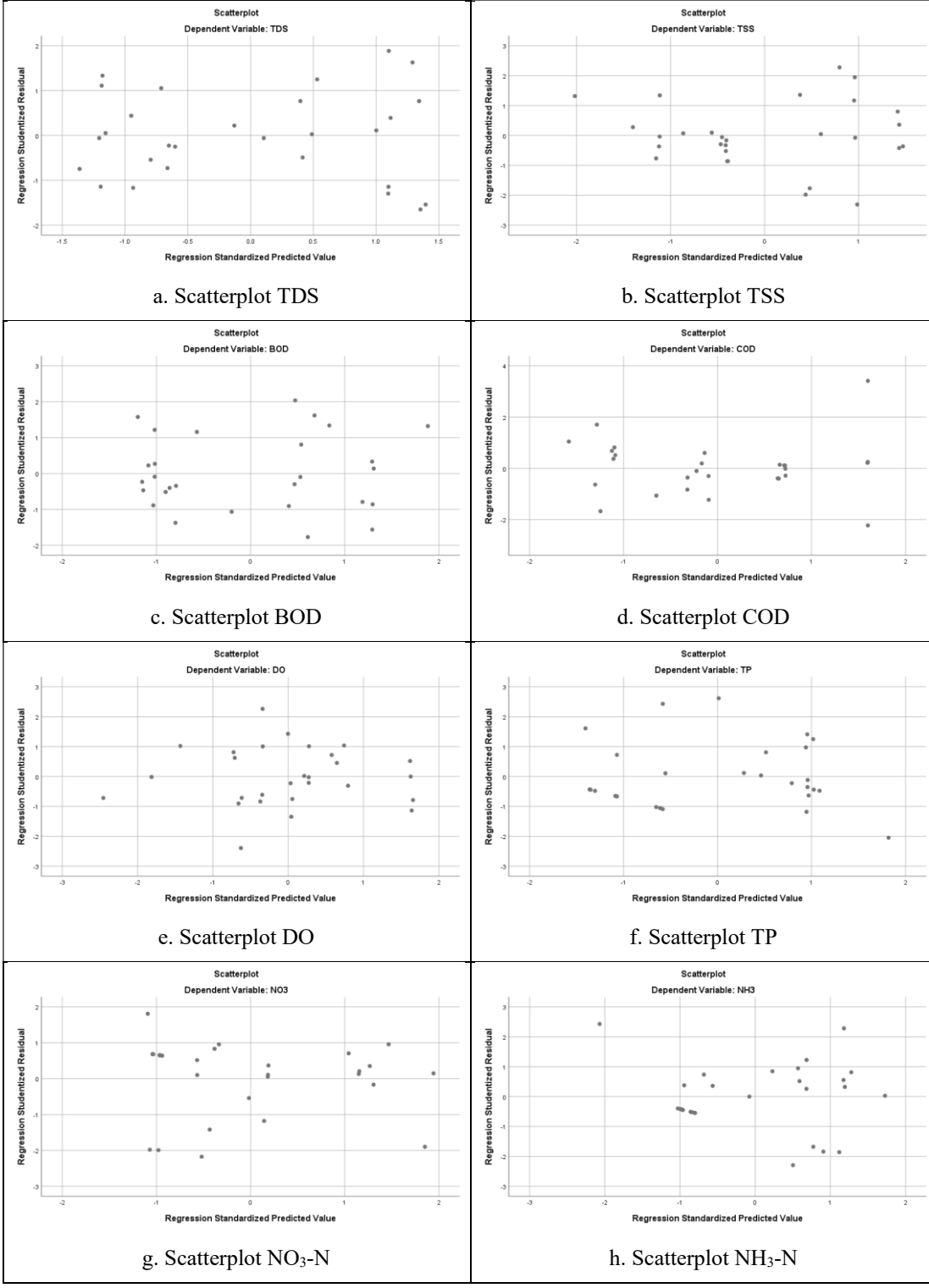


Figure 2. Heteroscedasticity Test Results



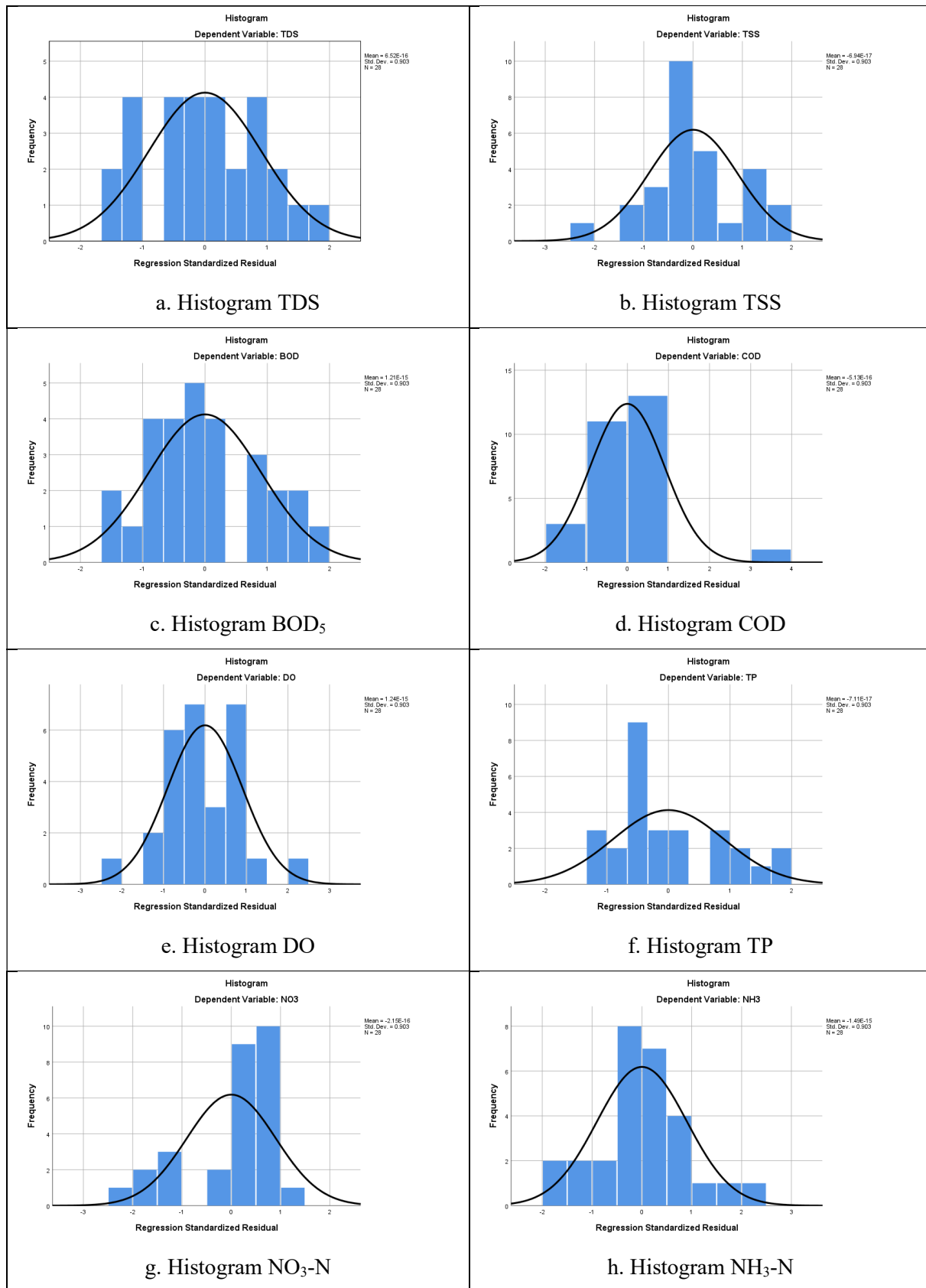


Figure 3. Normality Test Results

#### d. Autocorrelation Test

The result of the autocorrelation test in data processing indicates that the Durbin-Watson (DW) value for each variable falls outside the range of the lower limit (dL) and upper limit (dU). This implies that there is no autocorrelation in the model. The dL and dU values used in the analysis refer to the Durbin-

Watson Table (Junaidi 2010). These findings are consistent with previous research, which states that if the calculated DW value lies outside the dL and dU limits, the regression model is free from autocorrelation (Ainiyah et al. 2016). Details of the Durbin-Watson values are presented in Table 4.

Table 4. Autocorrelation Durbin-Watson Test Results

	TDS	TSS	BOD <sub>5</sub>	COD	DO	TP	NO <sub>3</sub> -N	NH <sub>3</sub> -N
Durbin-Watson	2.060	2.029	2.171	2.025	1.856	2.131	2.072	1.884
dL	1.028	1.028	1.028	1.028	1.028	1.028	1.028	1.028
dU	1.850	1.850	1.850	1.850	1.850	1.850	1.850	1.850

### 3.2 Water Quality Status

In 2019, the overall water quality of the Cisadane River at various observation points was within acceptable limits for several parameters, except for BOD<sub>5</sub> and DO, which mostly exceeded the quality standard limits. By 2020, the BOD<sub>5</sub> levels showed improvement, as observed at the Kedaung and Vihara locations, which met the quality standards after exceeding the limits in 2019. However, the DO value at the Genteng location declined in 2020, failing to meet the standard it adhered to in 2019. The TP value at the Batu Beulah location also decreased in quality compared to 2019, exceeding the quality standard. In 2021, the TSS values generally declined in quality compared to 2020, with several locations surpassing the quality standard of 50 mg/l. At the same time, the COD levels at the Genteng location increased beyond the quality standard, reaching 27.32 mg/l. The NH<sub>3</sub>-N levels also rose, with several locations exceeding the quality standard limits. The BOD<sub>5</sub> values at upstream locations, including Genteng, Batu Beulah, Serpong and Babakan, consistently exceeded the quality standard during this period. By 2022, the water quality of the Cisadane River showed overall improvement compared to 2021, as several locations met the quality standards. However, the TP values deteriorated further compared to 2021, particularly at Genteng, Batu Beulah, Serpong, and Babakan, where the values exceeded the quality standards.

Based on various parameters, the water quality in the Cisadane River Basin demonstrates variations across the years. TDS at all observation points, the TDS value

consistently remains below the quality standard of 1000 mg/l, indicating that the concentration of dissolved solids has not reached levels that could significantly disrupt water quality (Vigiak et al. 2019). TSS mostly meets the standard of 50 mg/l, though spikes occurred at Genteng and Batu Beulah in 2021 and 2022. Elevated TSS levels can decrease water clarity, reducing light penetration necessary for photosynthetic organisms (Bilotta and Brazier 2008). BOD<sub>5</sub> at many locations, particularly upstream areas such as Genteng, Batu Beulah, and Serpong, frequently exceed the standard of 3 mg/l. This suggests high concentrations of organic matter requiring oxygen for decomposition, which reduces dissolved oxygen availability and adversely affects aquatic ecosystems (Abdullahi et al. 2021). COD generally remains within the standard limit of 25 mg/l. However, locations such as Genteng experienced increased, indicating pollution from organic substances that elevate oxygen consumption and degrade water quality (Prilly and Harisuseno 2019). Dissolved oxygen levels vary significantly.

While upstream locations such as Batu Beulah and Serpong maintain values above the minimum 4 mg/l threshold supportive of aquatic life, downstream locations like Babakan and Kedaung show low DO values. These low levels can impair the growth and health of aquatic organisms (Susilowati et al. 2018; Samaras et al. 2023). Total Phosphate concentration at several points, including Batu Beulah and Serpong, occasionally exceed the safe limit of 0.2 mg/l. Elevated levels increase the risk of eutrophication, which can trigger algal blooms and deteriorate overall water quality (Akinawo 2023). NO<sub>3</sub>-N and NH<sub>3</sub>-H

are generally within safe limits. However, NH<sub>3</sub>-N concentrations spike at specific locations, such as Babakan and Genteng, during certain years. High NH<sub>3</sub>-N levels indicate domestic waste contamination, potentially causing oxidative stress and health issues for aquatic organisms (Li et al. 2023; Lin et al. 2023).

In more detail, the results of water quality tests in the Cisadane River in 2019-2022 obtained from the Ciliwung Cisadane River Basin Center and the Tangerang Regency Environmental and Sanitation Service can be seen in Table 5.

Table 5. Cisadane River Water Test Results 2019-2022

Station*	TDS (mg/l)	TSS (mg/l)	BOD <sub>5</sub> (mg/l)	COD (mg/l)	DO (mg/l)	Total P (mg/l)	NO <sub>3</sub> -N (mg/l)	NH <sub>3</sub> -N (mg/l)
BM II	1000	50	3	25	4	0,2	10	0,2
<b>2019</b>								
Genteng <sup>1</sup>	67	10	7	27	4,2	0,1	0,3	0,01
Batu Beulah <sup>1</sup>	70	12	7	17	4,1	0,1	0,5	0,01
Serpong <sup>1</sup>	81	40	7	22	4	0,1	1	0,01
Babakan <sup>1</sup>	83	15	10	21	3	0,08	0,5	0,01
Kedaung <sup>2</sup>	106	13	3,2	19	3,1	0,05	0,1	0,01
Vihara <sup>2</sup>	105	10	3,2	18	2,8	0,1	0,1	0,01
Tanjung Burung <sup>2</sup>	104	13	2,5	18	1,7	0,04	0,1	0,01
<b>2020</b>								
Genteng <sup>1</sup>	80,83	38	6,77	11,37	3,8	0,074	0,846	0,169
Batu Beulah <sup>1</sup>	70,1	35	7,13	12,8	5,9	0,241	1,37	0,038
Serpong <sup>1</sup>	80,61	38	5,59	11,8	5,3	0,041	1,29	0,127
Babakan <sup>1</sup>	88,7	19	5,35	8,3	3,8	0,1	1,56	0,183
Kedaung <sup>2</sup>	103	20	2,6	21	2,6	0,01	1	0,01
Vihara <sup>2</sup>	102	13	2,2	18	3	0,01	1	0,01
Tanjung Burung <sup>2</sup>	116	16	2,4	17	2,6	0,01	0,7	0,01
<b>2021</b>								
Genteng <sup>1</sup>	99,6	128	5,46	27,32	5,2	0,13	0,885	0,223
Batu Beulah <sup>1</sup>	92,9	70	4,32	15,21	4,9	0,096	1,52	0,144
Serpong <sup>1</sup>	88	60	4,63	16,26	3,8	0,414	1,21	0,212
Babakan <sup>1</sup>	105,7	17	3,74	11,53	2,7	0,088	2,56	0,322
Kedaung <sup>2</sup>	185	18	2,4	22	3,6	0,01	1	0,01
Vihara <sup>2</sup>	158	16	2,1	18	6,9	0,01	1	0,01
Tanjung Burung <sup>2</sup>	146	18	2,3	19	4	0,01	1	0,01
<b>2022</b>								
Genteng <sup>1</sup>	103,7	80	4,7	84	6	0,7	1,1	2
Batu Beulah <sup>1</sup>	104,9	110	2,9	12	5	0,6	1,8	0,08
Serpong <sup>1</sup>	108,1	77	3,4	19	4	0,6	1,5	0,1
Babakan <sup>1</sup>	130,5	23	3,7	19	2	0,4	1,8	0,3
Kedaung <sup>2</sup>	141	21	2	22	5,8	0,01	1	0,03
Vihara <sup>2</sup>	183	38	2,5	22	4,8	0,01	1	0,06
Tanjung Burung <sup>2</sup>	130	22	2	20	4,2	0,01	0,2	0,04

Locations such as Genteng experienced a significant increase in Pollution Index (PI) values, indicating a rise in pollution loads potentially caused by increased domestic or industrial activities. This may result from urbanization or intensified industrial activity around the river. Similarly, Anh et al. (2023) highlighted that residential and industrial land use, along with urbanization factors such as population growth and the expansion of impermeable surfaces, are primary contributors to river water pollution in urban areas. In contrast, locations like Vihara and Tanjung Burung showed a significant decrease in PI values, suggesting improved water quality linked to reduced pollution sources or enhanced environmental conditions. This improvement

could be attributed to diminished local pollution activities or increased environmental carrying capacity to manage loads. Feng et al. (2023) emphasized that increased vegetation cover offers substantial benefits in reducing water pollution by absorbing nutrients and filtering pollutants (Feng et al. 2023). Other locations exhibited stability or fluctuations in PI values, reflecting the complex dynamics of water pollution influenced by various environmental factors and human activities. Overall, the Cisadane River's pollution index generally falls under the "slightly polluted" category. The detailed PI values and water quality status of the Cisadane River from 2019 to 2022 are presented in Table 6.

Table 6. Water Quality Status in the Cisadane River

Observation point	2019		2020		2021		2022	
	IP value	Status	IP value	Status	IP value	Status	IP value	Status
Genteng	2,07	S	2,05	S	2,28	S	4,49	S
Batu Beulah	2,06	S	2,08	S	1,36	S	2,50	S
Serpong	2,08	S	1,71	S	2,00	S	2,53	S
Babakan	2,65	S	1,68	S	2,11	S	2,58	S
Kedaung	1,75	S	2,10	S	1,28	S	0,64	G
Vihara	1,98	S	1,82	S	0,51	G	0,69	G
Tanjung Burung	2,59	S	2,10	S	0,76	G	0,63	G

where G is Good, S is Slightly Polluted, M is Moderately Polluted, H is Heavily Polluted.

### 3.3 Multi-temporal Land Cover Status

Watershed delineation in the SWAT model is a procedure that divides areas based on topography and river networks. The largest Area of Interest (AOI) is located in Genteng, covering an area of 2,267.85 hectares. This location is situated in the topography and steep slopes. In contrast, the smallest AOI is located in Tanjung Burung, covering an area of 63.81 hectares. Tanjung Burung is situated in the downstream part of the watershed, where the slope is relatively low. This observation aligns with previous studies, which reported that the upstream areas of the Cisadane River Basin reach altitudes of up to 3,000 meters above sea level with slopes of up to 40%. Meanwhile, the downstream areas are predominantly flat to gently undulating (Sudinda 2021). The AOI for each observation point is illustrated in Figure 2.

The Genteng location is predominantly covered by agricultural land and vegetation. From 2019 to 2022, agricultural land decreased significantly from 43.16% (978.89 ha) to 40.64% (921.67 ha), indicating substantial agricultural land conversion. Vegetation cover also declined, albeit slightly, from 30.47% (691.04 ha) to 30.24% (685.74 ha). The Batu Beulah location is similarly dominated by vegetation and agricultural land. During the same period, agricultural land significantly increased from 35.66% (52.14 ha) to 46.21% (67.56 ha), suggesting land conversion toward agriculture. Conversely, natural vegetation experienced a sharp decrease from 37.57% (54.93 ha) to 30.09% (43.99 ha), reflecting vegetation degradation in the area.



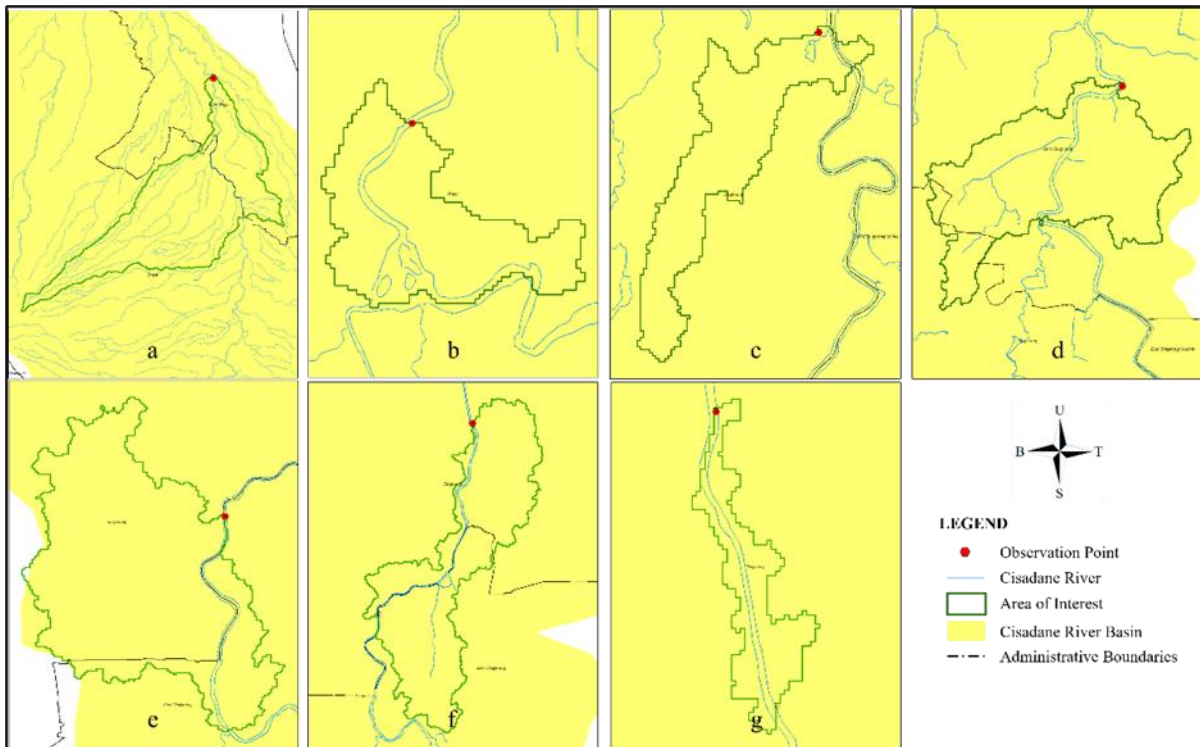


Figure 2. AOI of Each Observation Point (a. Genteng; b. Batu Beulah; c. Serpong; d. Babakan; e. Kedaung; f. Vihara; g. Tanjung Burung)

At the Serpong location, the dominant land covers are open land and built-up land. Built-up land increased from 32.74% (70.46 ha) to 34.99% (75.32 ha), signifying notable urban expansion. Meanwhile, open land, though still the largest land cover category, decreased slightly from 47.06% (101.29 ha) to 45.66% (98.27 ha), likely due to conversion for development purposes.

The Babakan location is primarily characterized by built-up land, which saw a significant rise from 70.79% (759.36 ha) in 2019 to 86.13% (923.93 ha) in 2022, indicating rapid urbanization. At the Kedaung location, agricultural land and built-up land are the dominant types of land cover. Agricultural land, the largest category, declined from 39.51% (428.31 ha) to 36.41% (394.69 ha), while built-up land increased gradually from 33.41% (362.11 ha) to 34.69% (376.00 ha), suggesting expanding urban development. The Vihara location was dominated by built-up land and agricultural land. Built-up land increased slightly from 39.41% (672.01 ha) to 40.04% (682.91 ha), reflecting growth in development. Agricultural land, however, decreased from 36.92% (629.66 ha) to 34.39% (586.47 ha), indicating a shift toward built-up land or other

uses. Finally, the Tanjung Burung location was primarily dominated by built-up land cover, which rose from 36.38% (23.48 ha) in 2019 to 38.49% (24.84 ha) in 2021 but then decreased to 35.45% (22.88 ha) in 2022. This fluctuation indicates relative stability in the development of built-up areas over time.

The analysis of land cover changes from 2019 to 2022 shows a general trend of increasing built-up land in most locations, except for Tanjung Burung, which is likely influenced by government regulations restricting construction along riverbanks. Agricultural land has decreased in areas such as Kedaung and Vihara, while vegetation has significantly declined, particularly in Babakan. Conversely, open land has increased across nearly all locations, indicating extensive land-clearing activities. These changes suggest a shift in land use from agriculture and vegetation to open or built-up land, potentially disrupting natural ecosystems and altering land use dynamics in the future. These results are in accordance with a study conducted by Basu, R & Das, A (2024) which states that rapid urbanization causes the conversion of agricultural land and vegetation to built-up land, which has a negative impact on ecosystem

services and environmental balance. Previous studies have also highlighted that rapid urbanization can exacerbate water quality issues, as observed in Nanjing, China, where

urbanization has led to increased water pollution and reduced sustainability of water resources (Ma et al. 2022).

Table 7. Land Cover Classification in the Cisadane Watershed (2019–2022)

Classification	2019		2020		2021		2022	
	Ha	%	Ha	%	Ha	%	Ha	%
<b>Genteng</b>								
Water Bodies	28,429	1,25	28,429	1,25	28,429	1,25	28,429	1,25
Agricultural Land	978,891	43,16	941,879	41,53	929,443	40,98	921,672	40,64
Built-Up Land	212,170	9,36	219,410	9,67	227,907	10,05	231,522	10,21
Open Land	357,324	15,76	387,694	17,10	393,199	17,34	400,492	17,66
Vegetation	691,039	30,47	690,443	30,44	688,876	30,38	685,739	30,24
Total	2,267,854	100	2,267,854	100	2,267,854	100	2,267,854	100
<b>Batu Beulah</b>								
Water Bodies	14,958	10,23	15,115	10,34	16,374	11,20	16,480	11,27
Agricultural Land	52,139	35,66	60,188	41,17	61,902	42,34	67,558	46,21
Built-Up Land	9,048	6,19	11,315	7,74	11,094	7,59	16,364	11,19
Open Land	15,118	10,34	8,163	5,58	10,685	7,31	1,802	1,23
Vegetation	54,929	37,57	51,411	35,17	46,137	31,56	43,988	30,09
Total	146,192	100	146,192	100	146,192	100	146,192	100
<b>Serpong</b>								
Water Bodies	2,838	1,32	2,925	1,36	2,925	1,36	2,925	1,36
Agricultural Land	1,959	0,91	2,497	1,16	2,497	1,16	1,949	0,91
Built-Up Land	70,462	32,74	71,987	33,45	72,769	33,81	75,315	34,99
Open Land	101,292	47,06	100,888	46,88	100,180	46,55	98,273	45,66
Vegetation	38,667	17,97	36,921	17,16	36,848	17,12	36,756	17,08
Total	215,218	100	215,218	100	215,218	100	215,218	100
<b>Babakan</b>								
Water Bodies	37,053	3,45	37,084	3,46	37,263	3,47	31,361	2,92
Agricultural Land	1,184	0,11	0,992	0,09	1,347	0,13	0,972	0,09
Built-Up Land	759,359	70,79	886,657	82,66	905,972	84,46	923,931	86,13
Open Land	10,228	0,95	74,554	6,95	94,737	8,83	106,328	9,91
Vegetation	264,856	24,69	73,395	6,84	33,361	3,11	10,089	0,94
Total	1,072,681	100	1,072,681	100	1,072,681	100	1,072,681	100
<b>Kedaung</b>								
Water Bodies	25,357	2,34	25,357	2,34	25,357	2,34	25,283	2,33
Agricultural Land	428,306	39,51	419,111	38,66	411,992	38,01	394,692	36,41
Built-Up Land	362,110	33,41	364,884	33,66	369,784	34,11	375,997	34,69
Open Land	211,904	19,55	218,993	20,20	221,513	20,44	234,871	21,67
Vegetation	56,289	5,19	55,620	5,13	55,319	5,10	53,122	4,90
Total	1,083,965	100	1,083,965	100	1,083,965	100	1,083,965	100
<b>Vihara</b>								

Classification	2019		2020		2021		2022	
	Ha	%	Ha	%	Ha	%	Ha	%
Water Bodies	47,958	2,81	47,958	2,81	47,958	2,81	47,422	2,78
Agricultural Land	629,663	36,92	624,042	36,59	621,236	36,43	586,468	34,39
Built-Up Land	672,010	39,41	674,759	39,57	676,186	39,65	682,909	40,04
Open Land	301,719	17,69	301,804	17,70	302,515	17,74	329,780	19,34
Vegetation	54,008	3,17	56,795	3,33	57,462	3,37	58,779	3,45
Total	1705,358	100	1705,358	100	1705,358	100	1705,358	100
Tanjung Burung								
Water Bodies	12,121	18,78	12,121	18,78	12,121	18,78	12,121	18,78
Agricultural Land	10,901	16,89	10,918	16,92	10,496	16,27	9,807	15,20
Built-Up Land	23,477	36,38	24,284	37,63	24,837	38,49	22,877	35,45
Open Land	9,094	14,09	8,656	13,41	9,069	14,06	11,947	18,51
Vegetation	8,934	13,84	8,547	13,25	8,003	12,40	7,774	12,05
<b>Total</b>	<b>64,526</b>	<b>100</b>	<b>64,526</b>	<b>100</b>	<b>64,526</b>	<b>100</b>	<b>64,526</b>	<b>100</b>

The analysis of land cover changes from 2019 to 2022 shows a general trend of increasing built-up land in most locations, except for Tanjung Burung, which is likely influenced by government regulations restricting construction along riverbanks. Agricultural land has decreased in areas such as Kedaung and Vihara, while vegetation has significantly declined, particularly in Babakan. Conversely, open land has increased across nearly all locations, indicating extensive land-clearing activities. These changes suggest a shift in land use from agriculture and vegetation to open or built-up land, potentially disrupting natural ecosystems and altering land use dynamics in the future. These results are in accordance with a study conducted by Basu, R & Das, A (2024) which states that rapid urbanization causes the conversion of agricultural land and vegetation to built-up land, which has a negative impact on ecosystem services and environmental balance. Previous studies have also highlighted that rapid urbanization can exacerbate water quality issues, as observed in Nanjing, China, where urbanization has led to increased water pollution and reduced sustainability of water resources (Ma et al. 2022).

### 3.4 The Effect of Land Cover on Water Quality

The effect of land cover changes on water quality parameters in the Cisadane River Basin can be expressed in the equation below:

$$\text{TDS} = 4,762 - 0,071 X_1 + 0,076 X_2 + 0,143 X_3 - 0,055 X_4 - 0,167 X_5 \quad \dots \text{Eq 3}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression analysis revealed that water bodies, agricultural land, built-up land, open land, and vegetation significantly influence TDS concentrations. An increase in the area of water bodies, open land, and vegetation reduces TDS concentrations by 0.071, 0.055, and 0.167 units, respectively, indicating their role in mitigating dissolved solids. Conversely, agricultural land and built-up land increase TDS concentrations by 0.076 and 0.143 units. These findings align with Sinulingga et al. (2023), who identified a correlation between the prevalence of agricultural land and elevated TDS due to dissolved non-organic fertilizers entering water bodies (Sinulingga et al. 2023). Similarly, Widodo et al. (2019) highlighted the role of vegetation as a natural filter, effectively reducing TDS levels by absorbing harmful dissolved particles (Widodo et al. 2019).

$$\text{TSS} = 3,785 - 0,477 X_1 + 0,036 X_2 + 0,019 X_3 - 0,104 X_4 + 0,270 X_5 \quad \dots \text{Eq 4}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and vegetation (X5). The regression equation for Total Suspended Solids (TSS)

indicates that water bodies, agricultural land, built-up land, open land, and vegetation significantly influence TSS concentrations. An increase in the area of water bodies and open land reduces TSS by 0.477 and 0.104 units, respectively, suggesting that these land covers help minimize suspended solid loads, possibly due to reduced sedimentation and runoff. Conversely, expansions in agricultural land, built-up land, and vegetation increase TSS by 0.036, 0.019 and 0.270 units, respectively. These findings align with Arkan Pratama and Chamid (2021) research, who highlighted that agricultural land contributes to higher TSS due to agricultural waste discharge into rivers (Arkan Pratama and Chamid 2021). This discrepancy underscores the importance of considering local river basin characteristics and human activities when analyzing TSS dynamics, as these factors significantly shape the relationship between land cover and suspended solids in rivers.

$$\text{BOD5} = 0,629 + 0,159 X_1 - 0,175 X_2 - 0,207 X_3 + 0,096 X_4 + 0,377 X_5 \quad \dots \text{Eq 5}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The BOD5 regression equation indicates that water bodies, agricultural land, built-up land, open land and vegetation significantly influence BOD5 values. Expanding water bodies, open land and vegetation areas increase BOD5 by 0.159, 0.096, and 0.377 units, respectively, while agricultural land and built-up land reduce BOD5 by 0.175 and 0.207 units. These findings align with Locke (2024), who noted that runoff from built-up land, by reducing infiltration and accelerating surface flow, limits the decomposition time for organic matter, thereby lowering BOD5. Similarly, agricultural land, often associated with managed runoff, reduces BOD5 levels. However, while absorbing pollutants, vegetation may increase BOD5 when decomposed organic matter enters the water system, highlighting its dual impact on water quality (Locke 2024).

$$\text{COD} = 2,576 - 0,079 X_1 + 0,054 X_2 + 0,003 X_3 + 0,038 X_4 + 0,046 X_5 \quad \dots \text{Eq 6}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression equation for COD demonstrates that water bodies, agricultural land, built-up land, and vegetation significantly influence COD levels. An increase in water body area decreases COD by 0.079 units, while expansions in agricultural land, built-up land, open land and vegetation increase COD by 0.054, 0.003, 0.038 and 0.046 units, respectively. These findings align with Chapra et al. (2021), who observed that increased vegetation and open land could elevate COD levels due to organic matter contributions and runoff. Similarly, agricultural and built-up land were found to contribute to higher COD concentrations, driven by agricultural inputs and household activities that introduce organic pollutants into water systems (Chapra et al., 2021).

$$\text{DO} = 1,294 - 0,098 X_1 + 0,039 X_2 - 0,044 X_3 - 0,004 X_4 + 0,104 X_5 \quad \dots \text{Eq 7}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression equation for DO reveals that increasing the area of water bodies, built-up, and open land decreases DO concentrations by 0.098, 0.044, and 0.004 units, respectively. Conversely, agricultural land and vegetation increase DO levels by 0.039 and 0.014 units, respectively. Built-up land significantly impacts water quality, including DO levels, due to pollutant runoff that reduces oxygen availability. These findings are consistent with studies by Chapra et al. (2021) and Ullah et al. (2024), which highlight that vegetation-rich areas enhance DO through photosynthesis, while runoff from built-up and open land introduces pollutants, leading to lower DO concentrations (Chapra et al. 2021; Ullah et al. 2024).

$$\text{TP} = 3,442 + 0,066 X_1 - 0,364 X_2 - 0,370 X_3 + 0,120 X_4 + 0,778 X_5 \quad \dots \text{Eq 8}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression equation for TP shows that the independent variables water bodies, agricultural land, built-up, open land and vegetation significantly



influence TP concentrations. An increase in the area of water bodies, open land and vegetation raises TP by 0.066, 0.120 and 0.778 units, respectively, while agricultural land and built-up land reduce TP by 0.364 and 0.370 units, respectively. These findings differ from those of Namugize *et al.* (2018), who reported that agricultural and built-up land generally increase TP levels due to runoff from fertilizers and phosphate sediments. The observed differences may result from agricultural practices and drainage systems that reduce direct runoff into water bodies in the study area. Additional factors such as climate, river basin characteristics, the intensity of human activities and management practices may also contribute to these discrepancies (Namugize *et al.* 2018).

$$\text{NO}_3\text{-N} = -0,562 + 0,078 X_1 - 0,165 X_2 - 0,072 X_3 + 0,103 X_4 + 0,152 X_5 \quad \dots \text{Eq 9}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression equation for NO<sub>3</sub>-N shows that the independent variables water bodies, agricultural land, built-up land, open land, and vegetation significantly influence NO<sub>3</sub>-N concentrations. An increase in the area of water bodies, open land and vegetation raises NO<sub>3</sub>-N by 0.078, 0.103 and 0.152 units, respectively, while agricultural land and built-up land reduce NO<sub>3</sub>-N by 0.165 and 0.072 units, respectively. These findings differ from those of Bratek *et al.* (2020), who reported that agricultural and built-up land generally increase NO<sub>3</sub>-N concentrations due to runoff containing fertilizers and other anthropogenic pollutants (Bratek *et al.* 2020). The discrepancies in this study may be attributed to the NO<sub>3</sub>-N levels being below the detection limit and differences in the methodologies applied, thus impacting the equation obtained.

$$\text{NH}_3\text{-N} = -5,303 + 1,012 X_1 - 0,582 X_2 - 0,885 X_3 + 0,869 X_4 + 0,506 X_5 \quad \dots \text{Eq 10}$$

Land cover variables: water bodies (X1), agricultural land (X2), built-up land (X3), open land (X4) and Vegetation (X5). The regression equation for NH<sub>3</sub>-N indicates that the independent variables water bodies, agricultural land, built-up, open land, and

vegetation significantly influence NH<sub>3</sub>-N concentrations. An increase in the area of water bodies, open land, and vegetation raises NH<sub>3</sub>-N levels by 1.012, 0.869, and 0.506 units, respectively. Conversely, agricultural land and built-up land reduced NH<sub>3</sub>-N by 0.582 and 0.885 units. The role of land in raising NH<sub>3</sub>-N highlights its potential to lower water quality. However, Zhang *et al.* (2023) found contrasting results, showing that agricultural and built-up land typically increase NH<sub>3</sub>-N due to fertilizer runoff and organic matter contributions from urban activities. Additionally, vegetation has been shown to mitigate nitrogen pollutants through natural filtration processes (Zhang *et al.* 2023). These differences may arise from the unique characteristics of the Cisadane river basin and variation in measurement methods.

Regression analysis of water quality parameters reveals that land use variables exhibit varying effects on these parameters, with vegetation generally playing a positive role in maintaining water quality. A study by Kang and Kanniah (2022) in the Johor River Basin, Malaysia, showed that conversion of forests to plantations and settlements caused a decrease in forest cover of 45.82% in the last 30 years, which resulted in increased erosion and sedimentation, affecting water quality parameters such as turbidity (TSS) and organic load in waters. Similar results were also found in the Cisadane River Basin, where an increase in built-up land from 212,170 ha in 2019 to 231,522 ha in 2022 in several locations contributed to an increase in TSS values, such as in Genteng, which increased from 10 mg/l in 2019 to 80 mg/l in 2022. While in the Sungai Johor Watershed, pollution was more dominated by an increase in sediment due to erosion, in the Cisadane River Basin the impact of pollution was more visible in the form of an increase in organic matter and a decrease in DO at several points, such as in Tanjung Burung which had a low DO value in 2019 (1.7 mg/l) before increasing to 4.2 mg/l in 2022. This similarity shows that changes in land cover in both river basins contribute to a decrease in water quality, with the main difference in the type of dominant pollutant, which depends on the characteristics of land use in each region.

It aligns with the findings of Tchobanoglous *et al.* (2003), which highlight the critical role of

vegetation and water bodies in pollutant reduction through natural filtration processes and runoff mitigation. On the other hand, agricultural and built-up land often negatively influence water quality, contributing to increased sediment loads, nutrients, and pollutants. These findings corroborate the research of Tong and Chen (2002), who emphasized the significant impact of urbanization and agricultural activities on water quality, particularly through elevated pollutant and nutrient loads.

#### 4. Conclusion

The research highlights that land cover changes in the Cisadane River Basin from 2019-2022 significantly impacted river water quality. Increased built-up land and decreased vegetation contribute to decreased water quality, which is reflected in increased BOD<sub>5</sub> levels and decreased DO in several locations. This decrease in water quality indicates that changes in land cover affect the balance of aquatic ecosystems, especially through increased surface runoff and reduced vegetation function in filtering pollutants. The limitations of this study include the correlational approach, so it cannot explain the direct cause-and-effect relationship between changes in land cover and water quality. In addition, this study did not consider other external factors such as climate change and rainfall patterns, which can also affect water quality in the Cisadane River Basin.

Overall, water quality in the Cisadane River Basin can be categorized as lightly polluted, with higher pollution levels in areas dominated by built-up land. An increase in built-up land of  $\pm 5.5\%$  in the 2019–2022 period contributed to the increase in BOD<sub>5</sub> levels. High BOD<sub>5</sub> concentrations indicate an increase in organic matter load originating from domestic waste and urban activities. Conversely, a decrease in vegetation cover of  $\pm 6.3\%$  at several observation points correlated with a decrease in DO, especially in downstream areas such as Tanjung Burung and Vihara, which can disrupt the balance of the aquatic ecosystem. These results emphasize the need to implement conservation-based spatial planning policies, such as increasing green areas around the watershed and stricter domestic waste

management, to prevent further water quality decline. For further research, it is recommended to develop a more comprehensive approach, such as the use of hydrological models to predict the impact of land cover changes on water quality in the long term. In addition, more detailed field studies with direct monitoring in various seasons can provide a deeper understanding of the factors contributing to water quality decline in the Cisadane River Basin.

#### Data availability statement

The data used in this study are secondary data in the form of Cisadane River water quality data and land cover data. Land cover data was obtained from Google Earth (<https://earth.google.com/>) which is open to the public. Cisadane River water quality data is provided by the Ciliwung Cisadane River Basin Center and the Tangerang Regency Environment and Sanitation Service. This data is available upon request and subject to the approval of the relevant institutions.

#### Funding Agencies

This research was conducted without any external funding. The authors confirm that no financial support was received from any funding agency for this study.

#### Conflict of interests

Every author has stated that there is no conflict of interest to the manuscript's writing or submission.

#### Acknowledgment

The authors would like to express their gratitude to Ciliwung Cisadane River Basin Center and Tangerang Regency Environmental and Sanitation Service for providing access to secondary data on water quality.

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## **The Dual Role of Vetiver Root Extract on Heterotrophic Bacterial Growth in Lake Riparian Zones: Implications for Lake Riparian Zone Water Quality**

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Received: 16 November 2024; Accepted: 26 December 2024; Published: 20 June 2025

**Abstract:** Plant–microbe interactions are fundamental to maintaining ecological balance in aquatic systems. In riparian zones, fluctuating lake water levels create standing water conditions that facilitate exchanges between riparian vegetation and aquatic microbes, particularly heterotrophic bacteria. This study examined the influence of *Vetiveria zizanioides* (Vetiver) root extract on heterotrophic bacterial growth in riparian ecosystems. Vetiver, widely utilized for erosion control and sediment stabilization, was selected as the experimental species. Soil and water samples from two small lakes in Cibinong, Bogor, West Java (Situ Cibuntu and Situ Cibinong), served as sources of bacterial isolates. Vetiver root extract was prepared via aqueous extraction and applied at varying concentrations in 50% tryptone glucose yeast (TGY) liquid medium. Bacterial growth responses were assessed through optical density measurements, alongside environmental parameter evaluations. Results demonstrated that Vetiver root extract modulated bacterial growth in a concentration-dependent manner: a 25% extract concentration significantly promoted heterotrophic bacterial growth, suggesting a role in enhancing nutrient cycling, whereas higher concentrations exhibited inhibitory effects, implying potential applications for microbial population management. These findings highlight vetiver root extract's dual role in stimulating or suppressing microbial activity within riparian ecosystems. This research provides a scientific foundation for developing nature-based solutions (NBS) to promote lake ecosystem stability and environmental sustainability. Further research is needed to assess the long-term effects of Vetiver root extract application on soil health, water quality, plant-microbe interactions, and overall ecosystem sustainability.

**Keywords:** bacterial growth, heterotrophic bacteria, riparian zone, root extract, vetiver plants

DOI: <https://doi.org/10.55981/limnotek.2025.8984>

### **1. Introduction**

Lakes are stagnant aquatic ecosystems occupying basins where inflow exceeds outflow, resulting in dynamic water levels that fluctuate over time (UNEP-IETC/ILEC, 2001). As lake water levels rise, the lakeside area (commonly referred to as the riparian zone) becomes inundated. This transitional area often hosts diverse plant species that serve as ecological buffers between terrestrial and

aquatic ecosystems (Leatemia, et al, 2016). These conditions foster interactions between riparian plants and aquatic microorganisms, such as heterotrophic bacteria carried by the lake water.

Heterotrophic bacteria are microorganisms incapable of synthesizing their food through photosynthesis or chemosynthesis. Instead, they rely on organic material from the environment, including detritus from

decomposing organisms or anthropogenic waste, as energy sources and carbon (Notowinarto and Agustina, 2015). Riparian plants, such as Vetiver grass (*Vetiveria zizanioides*), commonly referred to as Akar Wangi, contribute organic matter that can be metabolized by these microbes, enhancing nutrient cycling and improving soil structure. Vetiver grass (*Vetiveria zizanioides*), with its robust root system extending up to five meters, not only stabilizes soil but also enhances water quality by filtering sediments and nutrients (Jannah H and Safnowandi 2018). Additionally, the organic compounds released through its root exudates support microbial diversity and ecological processes such as nutrient cycling and decomposition of organic matter. Microbial interactions around plant roots also contribute to accelerating the decomposition of organic pollutant compounds, such as pesticides and hydrocarbons, into non-toxic forms (Bolan, 2011).

In the rhizosphere, plants and microbes interact by exchanging root exudates, which may serve as antimicrobial agents or carbon sources for microorganisms. Plants actively shape microbial communities by secreting specific exudate compounds, altering microbial composition and diversity to form beneficial associations (Broeckling, et al., 2008; Cesco, et al., 2012). These exudates' concentration and composition significantly influence the rhizosphere's microbial community structure (Cesco, et al., 2012). Their interactions with riparian plants often include symbiotic relationships where bacteria facilitate nutrient uptake and provide protection against soil-borne pathogens. Additionally, both plants and microbes engage in chemical signal exchanges, which affect gene expression and, consequently, the diversity and abundance of microbial and plant populations (Waters and Bassler, 2005; Zhuang, et al., 2013; Widyati, 2017). The diversity and abundance of microbial populations found around the roots of vetiver, as a result of ecological interactions, may include bacteria capable of detoxifying heavy metals. These microorganisms possess the ability to mitigate the harmful impacts of heavy metals through both intracellular and extracellular mechanisms within their tissues, potentially influencing the efficacy of

bioremediation processes (Triyani & Hafsan, 2021).

This study examines the interaction between Vetiver root compounds and heterotrophic bacteria in riparian zones, particularly in response to fluctuating lake water levels. The aim is to assess the effects of Vetiver root extract on heterotrophic bacterial growth, which is evaluated by measuring cell density using the Optical Density (OD) method. This approach is used to test the hypothesis that biochemical compounds released by Vetiver roots influence bacterial communities. This research contributes to understanding plant-microbe interactions in riparian ecosystems and the ecological implications of root-derived compounds. This research provides a scientific foundation for developing nature-based solutions (NBS) to promote lake ecosystem stability and environmental sustainability. Study of Zhang, et al. (2021) and Arrijani, et al. (2024) found that selecting suitable plant species in riparian zones can enhance microbial communities, improve nutrient cycling, and support ecosystem resilience.

## **2. Materials and Method**

### **2.1. Research Area**

Situ Cibuntu and Situ Cibunong, two small lakes located in Cibinong, Bogor Regency, West Java, Indonesia, were selected as study sites. Situ Cibuntu, situated within the Cibinong National Research and Innovation Agency Complex (06°29'S-106°51'E), covers an area of 2.11 hectares with a maximum depth of 1.20 meters (Sulastri, et al., 2020; Sadi, 2013). Previous studies have indicated that the lake has experienced significant sedimentation due to high suspended solid inputs (Zulti, et al., 2012). Situ Cibunong, located near the Mayor Oking Highway and Cibinong's traditional market (06°28'50"S-106°49'36"E), originally covered 5.77 hectares but has since reduced to 4.5 hectares. The lake suffers from pollution caused by domestic and non-domestic waste (Askhary, 2016), despite being a popular fishing spot.

### **2.2. Water and Soil Sampling**

Surface water samples were collected from three distinct points within the riparian zones of both Situ Cibuntu and Situ Cibunong. Sampling was conducted at three sites in each lake,

selected based on variations in riparian conditions. Water samples were collected at a depth of 50 cm from the lake surface and stored in polyethylene bottles. For water quality analysis, 500 mL of each water sample was collected in polyethylene bottles and either refrigerated or acidified to pH 2 following standard protocols (APHA, 2005). Meanwhile, water samples for microbiological analysis were collected in sterile bottles. Water samples were

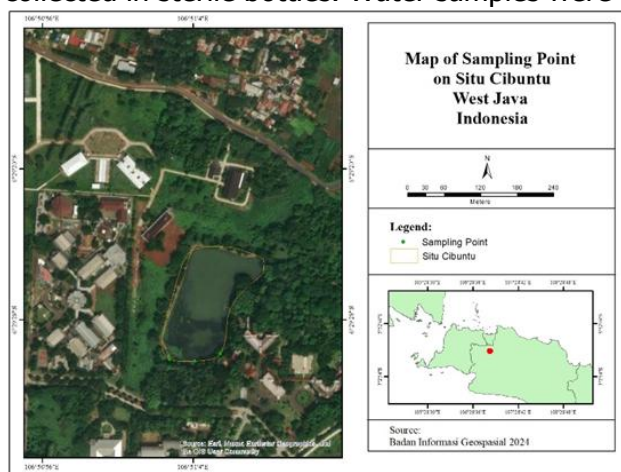


Figure 1. Sampling Location of Situ Cibuntu

### 2.3. Vetiver Root Extraction

Vetiver root extract was prepared using a standard aqueous extraction method. Dried vetiver roots, sourced from Garut, were cleaned and oven-dried at 50°C for 24 hours. The roots were cut into 1 cm segments and added to an Erlenmeyer flask containing 750 mL of distilled water (a 1:15 solid-to-liquid ratio). The mixture was heated to 100°C for 30 minutes and filtered through a multi-step process involving coarse filter paper, 1.5 µm GF/C filter paper, and 0.7 µm GF/F filter paper to remove solid particles and colloidal impurities. The resulting filtrate with no particulate matter was considered the 100% vetiver root extract.

### 2.4. Effect of Vetiver Root Extract on Bacterial Growth

To investigate the impact of vetiver root extract on bacterial growth, 0.6 mL of water samples were inoculated into 6 mL of half-strength TGY medium supplemented with various concentrations of vetiver root extract (0%, 1.5625%, 3.125%, 6.25%, 12.5%, 25%, 50%, and 100%). The inoculated media were incubated at 150 rpm for 48 hours. Microbial growth was evaluated by the turbidimetric

refrigerated and processed for subsequent isolation within 24 hours, as recommended by Sadi (2013). Soil samples were collected using sterilized shovels and gloves. Samples were taken from areas adjacent to the water's edge and slightly drier locations to represent varying soil moisture conditions. The samples were stored in polypropylene plastic containers.

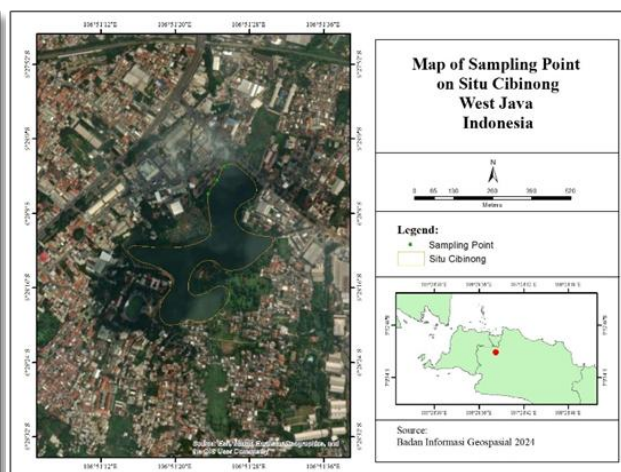


Figure 2. Sampling Location of Situ Cibinong

method, quantified using a UV-Vis spectrophotometer at 600 nm. Optical Density Units (ODU) were used to measure bacterial density. One-way ANOVA, followed by Tukey's HSD test, was employed to analyze the statistical significance of the results using PAST 4.03 software.

### 2.5. Heterotrophic Bacterial Enumeration

Water samples from the three sites were serially diluted at  $10^{-2}$  and  $10^{-4}$  dilutions. Aliquots of 100 µL of each diluted sample were spread plated onto TGY agar plates and incubated at room temperature for 48 hours. Colony-forming units (CFUs) were counted to determine bacterial abundance.

Soil samples (5 g) were added to 45 mL of peptone water and shaken for 1 hour at 150 rpm. Serial dilutions ( $10^{-4}$  and  $10^{-6}$ ) were prepared, and 100 µL aliquots were spread plated onto TGY agar plates. The plates were incubated at room temperature for 48 hours, and CFUs were counted.

### 2.6 Water and Soil Quality Analysis

Water quality parameters were measured using standard analytical methods, including pH, salinity, electrical conductivity, dissolved



oxygen (DO), ammonium, total phosphate, total nitrogen, chemical oxygen demand (COD), nitrate, and nitrite. A portable water quality checker (EZ-9909) was used to determine pH, salinity, and electrical conductivity. DO levels were measured using a Lutron DO meter. Ammonium, total phosphate, total nitrogen, and COD were analyzed using spectrophotometric methods (phenate, persulfate-ascorbic acid, brucine, and closed reflux, respectively) (APHA, 2005). Soil pH was measured using an ATC pH meter. Soil organic carbon content was determined spectrophotometrically at a specific wavelength (APHA, 2005).

### 3. Result and Discussion

#### 3.1. Environmental Condition in Situ Cibuntu and Situ Cibinong

##### *Water Quality*

Table 1 summarize water quality data for Situ Cibuntu and Situ Cibinong. The analysis revealed distinct differences in nutrient composition between the two locations. Situ Cibinong exhibited higher concentrations of ammonium, total nitrogen, and Chemical Oxygen Demand (COD), whereas Situ Cibuntu showed elevated nitrate, nitrite, and organic nitrogen levels.

Nitrogen is a crucial nutrient for plant growth, serving as a key component in protein synthesis and supporting aquatic plant development. In aquatic ecosystems, nitrogen compounds, such as nitrate ( $\text{NO}_3^-$ ), ammonium ( $\text{NH}_4^+$ ), and nitrite ( $\text{NO}_2^-$ ), are naturally derived from the metabolic activities of aquatic organisms and the bacterial decomposition of organic matter (Boyd, 1979; Ibrahim, et al., 2021). These nitrogen forms significantly influence species diversity, abundance, and nutrient availability in aquatic ecosystems for flora and fauna (Horne & Goldman, 1994).

Inorganic nitrogen compounds, particularly nitrate ( $\text{NO}_3^-$ ) and ammonium ( $\text{NH}_4^+$ ), can enhance bacterial growth. Ammonium often originates from industrial or human organic waste entering water bodies, while nitrate is primarily produced through the metabolic processes of aquatic organisms. During these processes, ammonium is oxidized to nitrite, which is subsequently converted to nitrate by waterborne microorganisms (Amalia, et al., 2021). The highest ammonium concentration was recorded at Situ Cibinong ( $2.250 \pm 0.82$  mg/L), while the highest nitrate concentration was observed at Situ Cibuntu ( $0.980 \pm 0.06$  mg/L).

Table 1. Water Quality Data for Situ Cibuntu and Situ Cibinong

Sampling Location	$\text{NO}_3\text{-N}$ Conc. (mg/L)	$\text{NH}_4\text{-N}$ Conc. (mg/L)	$\text{NO}_2\text{-N}$ Conc. (mg/L)	TN Conc. (mg/L)	N Organic Conc. (mg/L)	COD Conc. (mg/L)
Situ Cibuntu	1	0.915	0.849	0.247	2.665	0.654
	2	1.068	0.699	0.548	2.411	0.060
	3	0.966	0.537	0.378	3.057	1.176
	<b>Average</b>	<b>0.980</b>	<b>0.700</b>	<b>0.400</b>	<b>2.711</b>	<b>0.630</b>
	<b>SD</b>	<b>0.06</b>	<b>0.13</b>	<b>0.14</b>	<b>0.27</b>	<b>0.46</b>
Situ Cibinong	1	0.373	3.388	0.085	3.867	0.021
	2	0.484	1.513	0.065	3.369	1.307
	3	0.252	1.848	0.049	2.257	0.108
	<b>Average</b>	<b>0.370</b>	<b>2.250</b>	<b>0.007</b>	<b>3.160</b>	<b>0.480</b>
	<b>SD</b>	<b>0.09</b>	<b>0.82</b>	<b>0.01</b>	<b>0.67</b>	<b>0.59</b>

Conversely, nitrite ( $\text{NO}_2^-$ ), part of the nitrogen cycle, can negatively impact microbial populations. Nitrite contamination from fertilizers, livestock waste, and other organic inputs can inhibit bacterial growth at concentrations exceeding 1 mg/L by

interfering with bacterial respiration. Nitrite binds to cellular components like cytochromes, disrupting electron transfer during respiration (Radiastuti, 2009). Situ Cibuntu exhibited higher nitrite levels ( $0.400 \pm 0.14$  mg/L) compared to Situ Cibinong, potentially

causing slight disturbances to microbial growth.

COD is another critical water quality parameter, representing the oxygen demand required by aerobic microbes to oxidize organic matter into biomass, carbon dioxide, water, and inorganic compounds. High COD values typically indicate elevated organic matter levels, leading to reduced dissolved oxygen availability (Rahmawati, et al., 2013). The COD concentration at Situ Cibunong ( $39.167 \pm 16.16$  mg/L) was significantly higher than at Situ Cibuntu, likely due to household wastewater and market waste contributing additional organic material to the water.

The data in Table 2 represent the measurements conducted during the sampling process. The observed differences in water temperature between Situ Cibunong and

Situ Cibuntu reflect the environmental conditions at the sampling time. Water samples from Situ Cibuntu were collected earlier than those from Situ Cibunong. Additionally, the presence of dense vegetation and shaded areas surrounding Situ Cibuntu likely contributed to its lower water temperature than Situ Cibunong.

The pH values of the water samples from both Situ Cibuntu and Situ Cibunong were within the optimum range for microbial growth, thereby avoiding harmful effects on microorganisms. Specifically, the optimal pH range for bacterial growth is between 6.5 and 7.5, while the tolerable range extends from 4 to 9 (Hamdiyati, 2011). Furthermore, the measured values of Electrical Conductivity (EC) and salinity in both water bodies were relatively low, indicating minimal impact on bacterial growth.

Table 2. Water Quality Data from Situ Cibuntu and Situ Cibunong.

Sampling Location	Temperature (°C)	pH	Electrical Conductivity (μS/cm)	Total Dissolved Solid (ppm)	Salinity (%)	Dissolved Oxygen (ppm)	
Situ Cibuntu	1	26.40	6.81	144.7	72	0	4.40
	2	26.70	6.84	144.0	71	0	4.53
	3	26.70	6.80	144.0	72	0	4.60
	<b>Average</b>	<b>26.60</b>	<b>6.82</b>	<b>144.23</b>	<b>71.67</b>	<b>0.00</b>	<b>4.51</b>
	<b>SD</b>	<b>0.14</b>	<b>0.02</b>	<b>0.33</b>	<b>0.47</b>	<b>0.00</b>	<b>0.08</b>
Situ Cibunong	1	29.67	6.97	203.0	101	0.01	3.83
	2	29.90	7.21	156.0	78	0	5.60
	3	29.53	7.95	155.7	77	0	8.33
	<b>Average</b>	<b>29.70</b>	<b>7.38</b>	<b>171.57</b>	<b>85.33</b>	<b>0.00</b>	<b>5.92</b>
	<b>SD</b>	<b>0.15</b>	<b>0.42</b>	<b>22.23</b>	<b>11.09</b>	<b>0.00</b>	<b>1.85</b>

Dissolved oxygen (DO) is another critical factor for microbial survival, as aerobic microorganisms require oxygen to oxidize nutrients for metabolic processes. However, some microbial groups are anaerobic, with oxygen potentially toxic to their growth (Hamdiyati, 2011). The study results indicate that the dissolved oxygen concentrations in both Situ Cibuntu and Situ Cibunong were sufficient to support microbial activity and sustain their life processes.

#### *Soil Condition of Situ Cibuntu and Situ Cibunong*

The soil conditions of Situ Cibuntu and Situ Cibunong are summarized in Table 3. The organic carbon content in the soil sample from

Situ Cibunong was higher than that observed in Situ Cibuntu. Organic carbon is a vital energy source for microorganisms, supporting their growth and development (Waryanti, et al., 2013; Pitrianingsih, et al., 2014). Despite this, the bacterial count in the soil samples from Situ Cibunong was lower than in Situ Cibuntu. This discrepancy may be attributed to factors beyond organic carbon availability, such as toxic substances, competition with other microorganisms, and unfavorable environmental conditions, including temperature, pH, and humidity.

Environmental conditions, particularly soil pH, which was acidic in both samples, appear

less conducive to microbial growth. Prior research indicates that highly acidic conditions can disrupt cellular metabolism and hinder microbial development (Larosa, et al., 2013). However, the impact of acidity varies among

microbial species. In cases where pH levels fall outside the tolerance range of certain bacteria, cellular damage can occur, leading to inhibited growth (Larosa, et al., 2013).

Table 3. Soil Quality from Situ Cibuntu and Situ Cibunong.

Sampling Location		Temperature (°C)	pH	Moisture Content (%)	C-Organic Levels (%b/b)
Situ Cibuntu	1	28.50	5.07	3.23	8.36
	2	28.00	4.63	3.34	7.32
	3	28.30	4.60	3.41	8.01
	<b>Average</b>	<b>28.27</b>	<b>4.77</b>	<b>3.33</b>	<b>7.90</b>
	<b>SD</b>	<b>0.21</b>	<b>0.21</b>	<b>0.07</b>	<b>0.43</b>
Situ Cibunong	1	28.60	5.50	3.18	72.32
	2	28.50	5.43	6.14	162.49
	3	28.50	5.77	2.46	6.25
	<b>Average</b>	<b>28.53</b>	<b>5.57</b>	<b>3.93</b>	<b>80.35</b>
	<b>SD</b>	<b>0.05</b>	<b>0.15</b>	<b>1.59</b>	<b>64.04</b>

### 3.2 Bacterial Population Across Study Sites

The analysis of bacterial populations revealed that water and soil samples from Situ Cibuntu consistently exhibited higher Total Plate Count (TPC) values compared to those from Situ Cibunong (Table 4). In water samples, the TPC for Situ Cibuntu was  $(7.5 \times 10^4 \pm 8.0$

$\times 10^4)$  CFU/mL, whereas in Situ Cibunong, the TPC was  $(2.5 \times 10^4 \pm 3.2 \times 10^4)$  CFU/mL. Similarly, in soil samples, the bacterial count in Situ Cibuntu was significantly higher at  $(170 \times 10^6 \pm 220 \times 10^6)$  CFU/mL compared to  $(7.2 \times 10^6 \pm 1.1 \times 10^6)$  CFU/mL in Situ Cibunong.

Table 4. Number of Bacteria in Water and Soil Samples in Situ Cibuntu and Situ Cibunong.

Sampling Point		Water TPC (CFU/mL) Value	Soil TPC (CFU/mL) Value
Situ Cibuntu	1	$3.7 \times 10^4$	$1.1 \times 10^7$
	2	$1.9 \times 10^5$	$6.8 \times 10^6$
	3	$5.8 \times 10^2$	$4.8 \times 10^8$
	<b>Average</b>	<b><math>7.5 \times 10^4</math></b>	<b><math>170 \times 10^6</math></b>
	<b>SD</b>	<b><math>8.0 \times 10^4</math></b>	<b><math>220 \times 10^6</math></b>
Situ Cibunong	1	$7.0 \times 10^4$	$6.1 \times 10^6$
	2	$4.0 \times 10^3$	$8.3 \times 10^6$
	3	$7.0 \times 10^2$	-
	<b>Average</b>	<b><math>2.5 \times 10^4</math></b>	<b><math>7.2 \times 10^6</math></b>
	<b>SD</b>	<b><math>3.2 \times 10^4</math></b>	<b><math>1.1 \times 10^6</math></b>

These findings indicate that the soil in Situ Cibuntu harbors a substantially more abundant bacterial population than that of Situ Cibunong. Variations in bacterial populations between the sites may be attributed to differences in environmental conditions, the chemical composition of water and soil, and

anthropogenic activities. Situ Cibuntu, characterized by lush forest vegetation, experiences relatively cooler and more stable daytime water temperatures, which may facilitate microbial growth. Conversely, the absence of vegetation in Situ Cibunong likely results in greater diurnal temperature

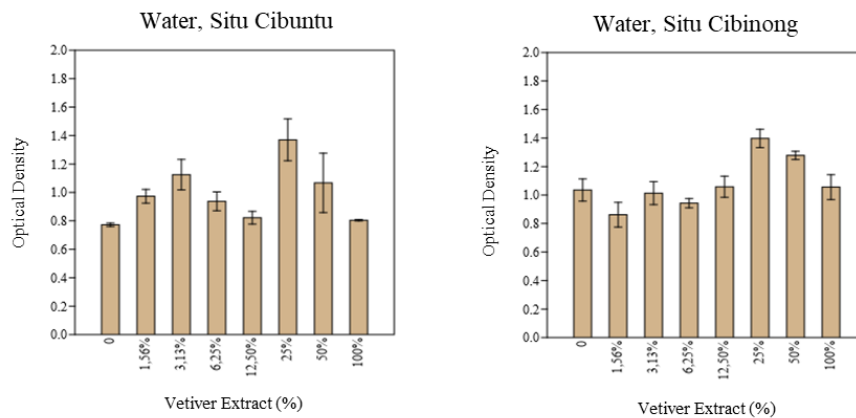
fluctuations, potentially hindering microbial development.

Vetiver root exudates play a critical role in supporting microbial activity by serving as carbon sources, while also influencing microbial community composition and driving riparian ecosystem processes such as nitrogen cycling and organic matter decomposition.

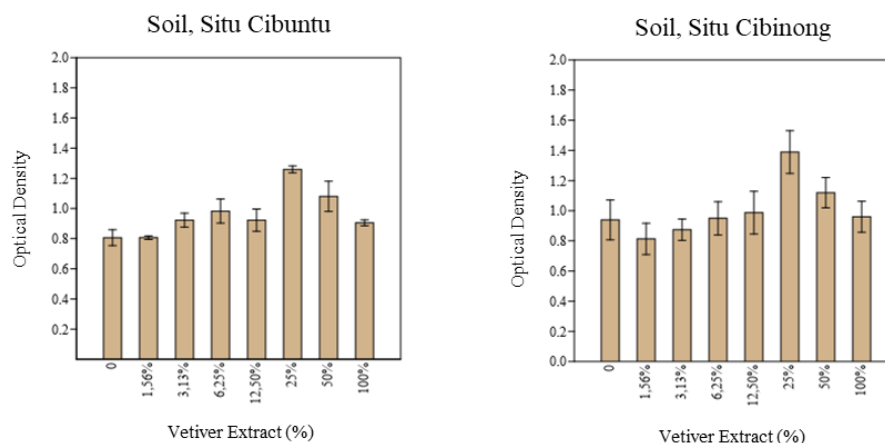
### 3.3 The Impact of Vetiver Root Extract on Bacterial Growth

The effect of vetiver root extract on the growth of heterotrophic bacteria in water and soil samples from the riverside areas of Situ Cibuntu and Situ Cibinong was investigated through a series of experiments. These

experiments utilized culture media supplemented with varying concentrations of vetiver root extract, ranging from 0% to 100%. Bacterial density was assessed by measuring the optical density (OD) of the cultures following a 48-hour incubation period. The turbidity of liquid media, which correlates with bacterial population density, was quantified using a UV-Vis spectrophotometer at 600 nm. The absorbance value, which is proportional to the OD, reflects the abundance of bacterial cells in the solution (Seniati & Irham, 2019). Comparisons of bacterial density in water and soil samples from the two study locations are presented in Figure 1 and Figure 2.



**Figure 3.** Bacterial density of the water sample.



**Figure 4.** Bacterial density of the soil sample.

Bacterial density exhibited a decreasing trend as the concentration of vetiver root extract increased, with notable differences observed across the tested concentrations (Figure 3 and Figure 4). At extract concentrations between 0% and 25%, a

significant increase in bacterial density was observed in both water and soil samples collected from Situ Cibuntu and Situ Cibinong. However, bacterial density began to decline at concentrations approaching 100%. The 25% extract concentration demonstrated the highest



bacterial growth, significantly exceeding that observed at other concentrations in both aquatic and riparian soil bacteria (Table 7). The findings indicate that at lower concentrations (0-25%), the vetiver root extract likely contains organic compounds that act as readily available nutrients or growth stimulants for bacteria. In contrast, the extract exhibits antibacterial properties at higher concentrations (50-100%), thereby inhibiting bacterial growth. This dual response highlights the necessity for further investigation to identify the specific bioactive compounds present in vetiver root extract and elucidate their mechanisms of action on microbial communities.

Our results align with the study conducted by Narayanasamydamodaran, et al. (2025) which found Vetiver root exudates selectively

shape the rhizosphere microbial community, favoring certain bacterial taxa that contribute to nutrient cycling and plant health. The abundance of beneficial groups like Proteobacteria and Actinobacteria, which are involved in nitrogen and phosphorus cycling, increases in the presence of specific exudate.

The release and diffusion of root exudates are affected by soil water content (Zhang, et al., 2023). Under optimal moisture, root exudates are more effectively distributed which in turn supports higher microbial activity. However, drought conditions can limit exudate diffusion and shift the balance of rhizosphere processes. This condition, in turn, makes water availability a more critical factor than exudation alone in sustaining microbial enzyme activities and nutrient cycling.

Table 7. Tukey's Pairwise Test

Comparison	P-Tukey's			
	Water, S. Cibuntu	Water, S. Cibinong	Soil, S. Cibuntu	Soil, S. Cibinong
0% - 25%	0,01534 *	0,03442 *	0,001122*	
1,56% - 25%		0,00118 *	0,00113*	0,04389*
1,56% - 50%		0,01209 *		
3,125% - 25%		0,02263 *	0,01652*	
6,25% - 25%		0,00572 *		
12,50% - 25%	0,02940 *		0,01678*	
25% - 100%	0,02332 *		0,01095*	

#### 4. Conclusion

Significant variations in bacterial populations were observed between Situ Cibuntu and Situ Cibinong, emphasizing the influence of environmental factors, nutrient availability, and water quality parameters. Situ Cibinong exhibited higher bacterial densities, which were attributed to favorable environmental conditions characterized by elevated ammonium and organic carbon levels. In contrast, Situ Cibuntu showed greater bacterial abundance in both water and soil samples, potentially driven by higher concentrations of nitrate, organic nitrogen, and chemical oxygen demand (COD). Plant exudates act as a source of organic compounds in the riparian zone.

This study highlights the dual effects of vetiver root extract on bacterial growth, demonstrating that lower concentrations (0–

25%) promote bacterial density, likely due to the presence of readily available nutrients, while higher concentrations (50–100%) inhibit bacterial proliferation through the action of antimicrobial compounds. This dynamic relationship is key to nutrient cycling, plant health, and ecosystem function in both natural and engineered environments, which in turn directly influence the structure and function of rhizosphere microbial communities. Water availability affects root exudation, thereby modulating microbial communities in the rhizosphere. These findings highlight the complex interplay between environmental conditions and vetiver root extract on bacterial communities. Further research is needed to identify specific compounds in vetiver root extract and their effects on bacteria in diverse aquatic ecosystems.

### Data availability statement

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request. Access requests will be considered to ensure compliance with ethical and legal guidelines.

### Funding Agencies

This study was funded by the Priority Lake and Water Resources Management Program in the 2022 fiscal year, managed by the Earth Sciences Research Organization of BRIN, under the grant number B-22/III/PR.03/1/2022. The funding agency had no role in the study's design, data collection, analysis, or interpretation of results, nor in the decision to publish this manuscript.

### Conflict of interests

The authors declare no conflicts of interest related to this study. All efforts have been made to ensure that the research was conducted objectively and free from any commercial or financial relationships that could be construed as a potential conflict of interest.

### Acknowledgment

The authors extend their gratitude to Fitri Yuliani for her valuable contributions to sampling works. Special thanks are also extended to the Research Center for Limnology and Water Resources BRIN, for providing technical support and facilities for this research. This study was supported by Rumah Program ORKM BRIN, which provided financial assistance for the successful completion of the project.

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p-ISSN: 0854-8390

e-ISSN: 2549-8029

**LIMNOTEK**

**Perairan Darat Tropis di Indonesia,**

***transforming into the Journal of Limnology and Water Resources***

**Volume 30, Number 1, June 2025**

**Published by:**

National Research and Innovation Agency (BRIN)

Indonesian Limnology Society (MLI)



BRIN

