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Advancing Freshwater Ecosystem Science Through Monitoring, Innovation, and Ecological Research

LIMNOTEK continues to focus on establishing an integrated understanding of the interface between natural water processes, inland aquatic ecosystems, and human interactions.

In this 2026 volume 1 issue, we are pleased to present six research articles covering diverse aspects of limnology and water resources. The studies explore ecological processes and biodiversity in freshwater ecosystems, including predator–prey interactions in zooplankton communities, habitat characteristics influencing fish diversity, and the vertical distribution of chlorophyll-a in relation to environmental conditions and water column stratification. Advances in water quality management and monitoring are highlighted through assessments of chemical oxygen demand removal using nature-based and adsorption approaches, as well as evaluations of data quality in real-time river water quality monitoring systems. In addition, a methodological contribution demonstrates the application of UV-Vis spectrophotometry for quantitative prediction of *Spirulina platensis* biomass. Together, these studies emphasize the importance of integrating ecological understanding, environmental monitoring, and innovative approaches to support sustainable water resources management.

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

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Life-History Responses of *Daphnia* to Catfish Kairomones: Does *Ipomoea aquatica* Function as an Effective Refuge?

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Abstract

Zooplankton, particularly *Daphnia* sp., play a key role in aquatic food webs and exhibit strong life-history plasticity in response to predation risk mediated by chemical cues (kairomones). Aquatic macrophytes such as water spinach (*Ipomoea aquatica*) are often assumed to provide refuge that may mitigate these effects. This study examined the influence of catfish (*Clarias* sp.) kairomones and the potential moderating role of *I. aquatica* on reproduction and survival of *Daphnia* sp. using an individual-based bioassay with a randomized block design. Four treatments were applied: control, *I. aquatica*, kairomones, and kairomones + *I. aquatica*, observed in 15 days. Parameters measured included neonate number, age at first and second reproduction (AFR, ASR), Second Clutch/First Clutch ratio (C2/C1 ratio), and mortality. Kairomone exposure significantly reduced first-clutch offspring production and delayed AFR, and induced compensatory reproduction in the second clutch, reflected by the highest C2/C1 ratios (K = 2.30; H = 2.11). Mortality was also highest in kairomone treatments (>42%). The presence of *I. aquatica* did not significantly mitigate kairomone effects and was associated with increased mortality relative to the control. These results indicate that chemical predator cues dominate over physical shelter in shaping *Daphnia* sp. life-history strategies, with important implications for zooplankton dynamics in vegetated freshwater ecosystems.

1. Introduction

Zooplankton, including *Daphnia* sp. are a key component in aquatic ecosystems and serve as a link between primary producers (phytoplankton) and higher-level consumers (fish) (Andersen & Hessen, 2005; Ogorelec et al., 2021). The predation pressure they experience triggers the development of various defense responses, which can include changes in behavior, morphology, and life cycle (Diel et al., 2020). These responses are specific to the type of predator (Octorina et al., 2022; Baludo et al., 2024) and can be induced by chemical signals (kairomones) released by predators (Rabus et al., 2013). One important life cycle response is a change in reproduction timing and offspring number (Diel, 2020). For example, exposure to fish kairomones can cause *Daphnia* sp. to reproduce earlier with more offspring as an emergency strategy (Stibor, 1992).

However, this defensive response often comes with an “ecological cost,” such as reduced survival rates.

On the other hand, aquatic macrophytes such as water spinach (*Ipomoea aquatica*) play an important ecological role, one of which is as a provider of habitat and shelter for aquatic organisms, including zooplankton (El-Hady & Khalifa, 2015; Thomaz, 2017). The presence of macrophytes can influence community structure by providing refugia from predation pressure (Dos Santos et al., 2020).

Chemically, some aquatic plants such as *I. aquatica* can produce chemical compounds or allelochemicals that can affect the self-defense of *Daphnia* sp. when exposed to predator threats (kairomones) (Diller, 2023). Catfish (*Clarias* sp) like other fish carnivore release kairomone that can induce prey aquatic ecosystem (Owsley, 2017).



Ipomoea aquatica (water spinach) as a common floating macrophyte in tropical Asian freshwater ecosystems, where it provides structural complexity and increases habitat heterogeneity that can serve as shelter for zooplankton against visual predators (Thomaz, 2017; Dos Santos et al., 2020). Additionally, macrophytes including *I. aquatica* play important ecological roles in nutrient filtration, maintaining water quality, and supporting overall aquatic biodiversity (Wetzel, 2001; De et al., 2019). However, a key limitation that must be acknowledged is that the presence of floating macrophytes can alter microhabitat conditions through mechanisms such as reduced light penetration, decreased water circulation, and potential fluctuations in dissolved oxygen, which may create physiological stress for organisms inhabiting the water column (De et al., 2019). Furthermore, some aquatic plants are known to release allelochemical compounds that can influence the defensive responses of zooplankton such as *Daphnia* when exposed to predator kairomones, adding another layer of complexity to plant-zooplankton interactions (Diller et al., 2023). This inherent duality—offering structural protection while potentially altering water quality and releasing bioactive compounds—makes *I. aquatica* an ideal test subject for investigating whether physical refugia can moderate chemical signals from fish predators (Burks, 2002; Diel et al., 2020). Understanding this interaction is particularly important for tropical limnetic ecosystems, where research on antipredator responses of zooplankton remains limited compared to subtropical regions (Burks, 2002).

However, it is unclear whether the physical protection provided by macrophytes can eliminate or moderate the physiological responses and life cycle changes induced by kairomones. With the presence of shelter, does *Daphnia* sp. no longer need to employ the costly “live fast, die young” strategy? This study aims to examine: (1) the effect of catfish kairomones on the reproduction and life cycle of *Daphnia* sp. and (2) the role of *I. aquatica* as a shelter in moderating the impact of these kairomones.

2. Materials and Method

2.1. Time and Location of Study

This research was conducted from July to October 2025 at the Bioecology Laboratory, Faculty of Agriculture, Universitas Muhammadiyah Sukabumi, Indonesia. The laboratory is equipped with controlled environmental facilities for aquatic organism culture and experimental bioassays (USEPA, 1987). All experimental procedures, including culture maintenance, treatment preparation, and data collection, were carried out under standardized laboratory conditions to ensure consistency and reproducibility of results.

2.2. Experimental Design

The study employed an experimental method using a randomized block design (RBD) to evaluate the effects of catfish kairomones and the presence of water spinach (*Ipomoea aquatica*) on the life cycle parameters of *Daphnia* sp. Four treatment conditions were established (Table 1) with three replications each.

A total of 120 synchronized *Daphnia* sp. individuals (<24 hours old) were used in this experiment, with 10 individuals allocated to each replicate (30 individuals per treatment). Each experimental unit consisted of a single *Daphnia* sp. individual maintained in 250 mL of the respective treatment medium (Figure 1), following the protocol established by Octorina et al. (2022). This individual-based approach allowed for precise tracking of reproductive events and survival rates throughout the observation period in 15 days.

Table 1. Experimental treatment conditions used to evaluate the effects of catfish kairomones and *Ipomoea aquatica* on the life cycle of *Daphnia* sp.

Symbol	Treatment
K	Fish conditioned water (Kairomone)
H	Fish conditioned water (Kairomone) + <i>I. aquatica</i>
A	Aerated Well water + <i>I. aquatica</i>
C	Aerated Well water (Control)

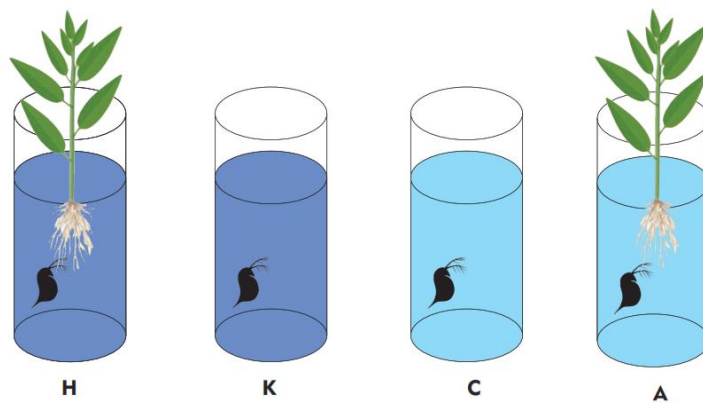


Figure 1. The four experimental treatments: (H) combination of *I. aquatica* and kairomone-conditioned water; (K) kairomone-conditioned water only; (C) control group containing neither *I. aquatica* nor kairomones; and (A) *I. aquatica* only.

2.3. Preparation of Kairomone Conditioned Water

Kairomone-conditioned water was prepared following the method described by Pietrzak et al. (2017). Juvenile catfish (*Clarias sp.*) measuring 3–5 cm in total length were starved for 24 hours prior to kairomone production to eliminate confounding effects from fecal material (Figure 2). The fish were then placed in 1-day-old aerated well water at a density of 5 individuals per liter for 24 hours at room temperature (25°C). After the conditioning period, the water was gently filtered through a 0.45 µm membrane filter to remove particulate matter, including bacteria and any remaining solid waste, while retaining dissolved chemical cues (kairomones). The filtered kairomone water was stored at 4°C and used within one week of preparation to ensure kairomone stability and activity (Pietrzak et al., 2017).

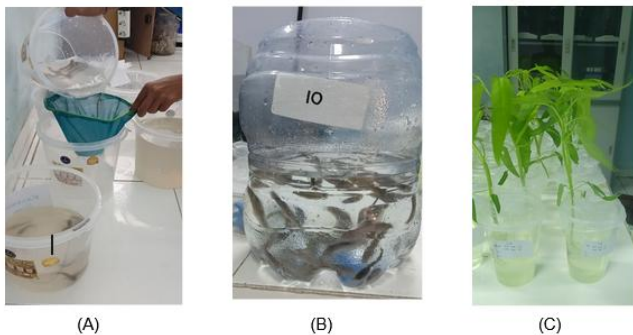


Figure 2. (A) Experimental preparation process, (B) including filtration of fish-conditioned water to obtain dissolved kairomones, conditioning of juvenile catfish (*Clarias sp.*), and (C) preparation of *Ipomoea aquatica* for treatment application.

2.4. Preparation of Water Spinach

Fresh water spinach (*Ipomoea aquatica*) plants were collected from clean, uncontaminated freshwater sources in Sukabumi, West Java. Plants were thoroughly rinsed with distilled water to remove epiphytes, sediment, and any associated organisms. Healthy specimens of uniform size (approximately 15 cm in height) were selected and acclimated in aerated well water for 48 hours prior to experimental use. For treatments requiring macrophytes (A and H), one individual plant was placed in each experimental container, with the root system fully submerged in the medium and leaves emerging above the water surface, simulating natural floating macrophyte conditions.

2.5. Feed Protocol

Experimental units received standardized feeding with *Chlorella sp* at a density 195,840 cells mL⁻¹ (Figure 3). Algal density was determined using a Sedgwick Rafter Counting Cell (SRC) under a compound microscope (400× magnification) and adjusted daily to maintain consistent food availability throughout the experiment. Feeding was performed immediately following water

renewal and offspring removal to ensure stable nutritional conditions.



Figure 3. Cultivation and preparation of *Chlorella sp.* was used as a standardized food source for *Daphnia sp.* during the experimental period.

2.6. Data Collection

Data collection was performed daily for 15 consecutive days, with observations conducted at the same time each day (09:00–11:00 AM) to maintain consistency. The reproductive parameters recorded included the number of neonates in the first clutch (C1) and second clutch (C2), the age at first reproduction (AFR) and age at second reproduction (ASR) defined as the day when the first and second clutches were released, and the C2/C1 ratio calculated as an indicator of reproductive energy allocation strategy. Mortality rate was recorded daily by observing each *Daphnia sp.* with an individual considered dead when no movement or heartbeat was observed over a 30-second period, and cumulative mortality was calculated as the percentage of initial individuals that died during the experimental period.

Collected data were compiled in Microsoft Excel and analyzed using R statistical software (version 4.2.1), employing a linear mixed-effects model (LMER) with experimental replicates treated as random factors, followed by independent sample t-tests for pairwise comparisons between treatments and chi-square tests for mortality data analysis, with statistical significance set at $\alpha = 0.05$ for all analyses and results presented as mean \pm standard deviation (SD).

3. Results

3.1. Impact on the Number of Neonates

The kairomone treatment (K and H) showed a clear suppression of *Daphnia sp.* reproduction in the first clutch. The number of C1 neonates in K and H was significantly lower than in the control. In the second clutch, despite an increase, the K and H groups remained lower than the control. The water spinach treatment (A) actually showed a decrease in the number of neonates from C1 to C2 (Table 2 and Figure 8).

Table 2. Mean number of neonates produced in the first (C1) and second (C2) clutches under different treatments

Treatment	Clutch 1 (individuals)	Clutch 2 (individuals)
Control	13.59 ± 5.05 (n=41)	19.98 ± 4.50 (n=41)
<i>I. aquatica</i>	14.28 ± 2.29 (n=40)	14.97 ± 2.94 (n=36)
<i>I. aquatica</i> + Kairomone	9.92 ± 6.23 (n=25)	17.88 ± 6.36 (n=24)
Kairomone	9.85 ± 6.08 (n=26)**	17.92 ± 3.62 (n=24)

*Significance levels: ** $p < 0.001$, $p < 0.01$, $p < 0.05$, $ns = not\ significant$.

3.2. Effect on Reproductive Age

Kairomones consistently slowed down reproductive development in the tested organisms. The values of AFR and ASR observed in treatments K and H were significantly higher than those recorded in the control and treatment A, indicating delayed reproductive maturity under kairomone exposure (Figure 6). This result suggests that the presence of predator-related chemical cues may alter energy allocation and physiological processes associated with reproduction.

In contrast, the treatment containing only water spinach did not show a statistically significant difference compared with the control treatment (Table 3). This finding indicates that water spinach alone had little influence on reproductive timing in the absence of kairomones. Overall, the results demonstrate that kairomones play a more dominant role in affecting reproductive age than the presence of aquatic vegetation.

3.3. C2/C1 Ratio and Mortality

The C2/C1 ratio is an indicator of reproductive energy allocation strategy calculated from the ratio of the number of neonates in the second clutch (C2) to the first clutch (C1). This parameter reflects how organisms adjust their reproductive investment under different environmental conditions and stress levels. The highest C2/C1 ratio was found in treatments K (2.3) and H (2.11) (Table 4), indicating a compensatory reproductive strategy under stressful conditions. The highest mortality rates also occurred in treatments K (45.24%) and H (42.86%), which were significantly higher than the control (2.38%) and treatment A (14.29%) (Figure 4). These

findings suggest that increased reproductive allocation may occur simultaneously with elevated physiological stress and mortality risk.

The C2/C1 ratio indicates how *Daphnia* sp. allocates its reproductive energy over time under treatment stress (Table 4). Control (Ratio = 1.9): A ratio of nearly 2 indicates a normal and sustainable reproductive strategy. *Daphnia* sp. in safe conditions can increase their reproductive investment in the second generation, which is a common and healthy reproductive pattern (Stibor 1992). This pattern suggests that individuals have sufficient energy reserves and experience minimal environmental pressure during reproduction.

I. aquatica treatment (Ratio = 1.07): A ratio close to 1 indicates that reproduction hardly increased from C1 to C2. It can be argued that the presence of the physical structure of duckweed, although not a direct threat, may create slightly stressful microhabitat conditions or resource competition (e.g., for space or light for algal food growth), so that the energy available for increased reproduction is not significant. As a result, reproductive performance remained relatively stable between the two clutch periods.

Kairomone and Kairomone + *I. aquatica* (Ratio = 2.3 and 2.11): These are very crucial findings. Both treatments show the highest ratios, even higher than the control. This pattern indicates that *Daphnia* sp. may respond to predator-related chemical cues by increasing reproductive investment in later clutches as an adaptive survival strategy. The elevated reproductive ratio under high-stress conditions may represent an attempt to maximize offspring production before mortality occurs.

Table 3. Mean reproductive age of *Daphnia* sp. in the first reproductive event (AFR) and second reproductive event (ASR) under different treatments

Treatment	AFR	ASR
Control (C)	6.10 ± 0.58 (n=41)	8.05 ± 0.44 (n=41)
<i>I. aquatica</i> (A)	5.95 ± 0.22 (n=40)	7.94 ± 0.23 (n=36)
<i>I. aquatica</i> + Kairomone(H)	7.04 ± 0.84 (n=25)	9.42 ± 1.32 (n=24)
Kairomone (K)	7.35 ± 0.94 (n=26)**	9.00 ± 0.85 (n=24)

*Values are presented as mean ± SD. Significance levels: ** $p < 0.001$, $p < 0.01$, $p < 0.05$; $ns = not\ significant$.

Table 4. C2/C1 ratio of *Daphnia* sp. under different treatments

Treatment	C2/C1 Ratio
Control (C)	1.9 ± 1.7
<i>I. aquatica</i> (A)	1.07 ± 0.29
<i>I. aquatica</i> + Kairomone(H)	2.11 ± 1.02
Kairomone (K)	2.3 ± 0.91

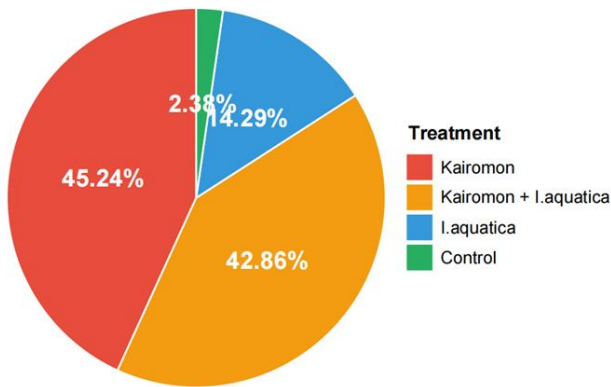


Figure 4. Percentage mortality of *Daphnia* sp. under different experimental treatments, including kairomone exposure, *Ipomoea aquatica*, combined treatment, and control conditions.

Mortality rate (Figure 4) provides direct evidence of the “cost” or negative consequences of kairomone exposure. Control (2.38%) indicates very low mortality, suggesting ideal maintenance conditions and the absence of significant stressors. *I. aquatica* (14.29%) shows an

increase in mortality compared to the control, reinforcing the previous argument that the presence of water spinach causes some kind of pressure, possibly through competition or changes in environmental conditions (e.g., O₂ fluctuations), which increases stress and causes death in some individuals (Diller 2023). Kairomone and Kairomone + *I. aquatica* (45.24% and 42.86%) showed very high mortality rates, and the near-equal mortality rates in both treatments are the most compelling evidence of the negative impact of kairomone. Mortality occurred on different days from the first to the last days for each treatment.

4. Discussion

The results of this study indicate that catfish kairomones trigger changes in the life cycle strategy of *Daphnia* sp. The pattern of reproductive suppression in C1 followed by increased compensation in C2 (high C2/C1 ratio) (Figure 5) is consistent with the “Delayed Reproduction” as stress-induced compensatory reproductive strategy reported by Stibor (1992). In his study, *Daphnia* sp. exposed to fish kairomones also showed a decrease in the age of first reproduction and an increase in the number of offspring in the first generation, an emergency response to predation pressure.

However, this strategy does not come without a cost. The high mortality rate in the kairomone treatment reflects the substantial ecological cost of induced defense. This concept of trade-off is the basis of many inducible defense responses, in which resources allocated to one function (e.g., early reproduction) sacrifice other functions (e.g., body maintenance and long-term survival) (Diel et al., 2020).

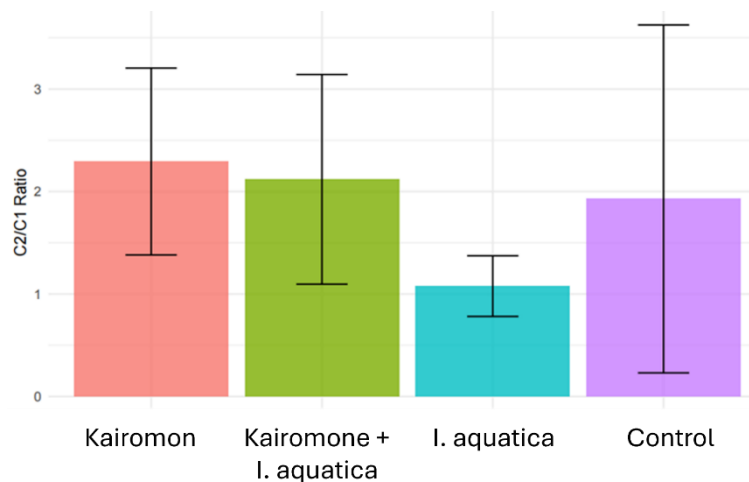


Figure 5. Comparison of C2/C1 neonate ratios of *Daphnia* sp. among control, *Ipomoea aquatica*, kairomone, and combined treatments.

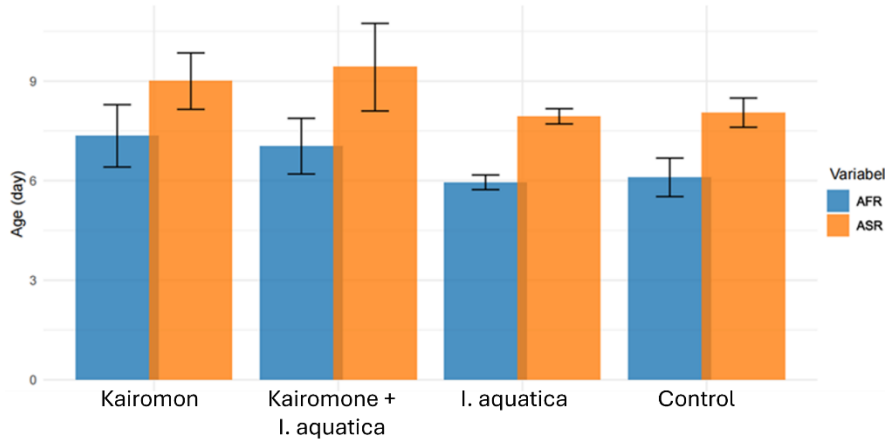


Figure 6. Average reproductive age in two variables, age at first reproduction (AFR) and age at second reproduction (ASR).

While our results showed that the roots of a single *I. aquatica* plant did not provide an effective physical refuge against kairomones, it remains to be seen whether submerged macrophytes with more complex morphological structures, or simply a higher density of roots, might offer the anticipated protection. The absence of consistent significant differences between the K and H treatments in all measured parameters (number of neonates, AFR, ASR, C2/C1 ratio, and

mortality) suggests that chemical signals (kairomones) have a stronger and more direct influence on *Daphnia* sp. physiology than the presence of a simple physical refugia. This reinforces the findings of Van De Meutter et al. (2004) that the response of *Daphnia* sp. to kairomones is highly specific and can influence habitat selection, but in this context, physical refuge cannot “deactivate” the physiological response that has been triggered.



Figure 7. Morphological arrangement of *Ipomoea aquatica* in experimental units, illustrating the limited structural complexity of its submerged root system as potential refuge for *Daphnia* sp.

As a floating macrophyte, *I. aquatica* exhibits limited structural complexity (Figure 7). Its root system simply hangs in the water column, offering only two-dimensional protection rather than an adequate three-dimensional shelter for *Daphnia* sp. This simple architecture might be insufficient against dissolved chemical cues from predators. The effectiveness of macrophyte protection is highly dependent on the density and complexity of the architecture (Burk et.al., 2002).

However, other macrophytes, such as *Ceratophyllum* and *Myriophyllum*, possess complex leaf structures and dense branches. Whether these more structurally complex macrophytes can offer an effective refuge for daphnids warrants further investigation.

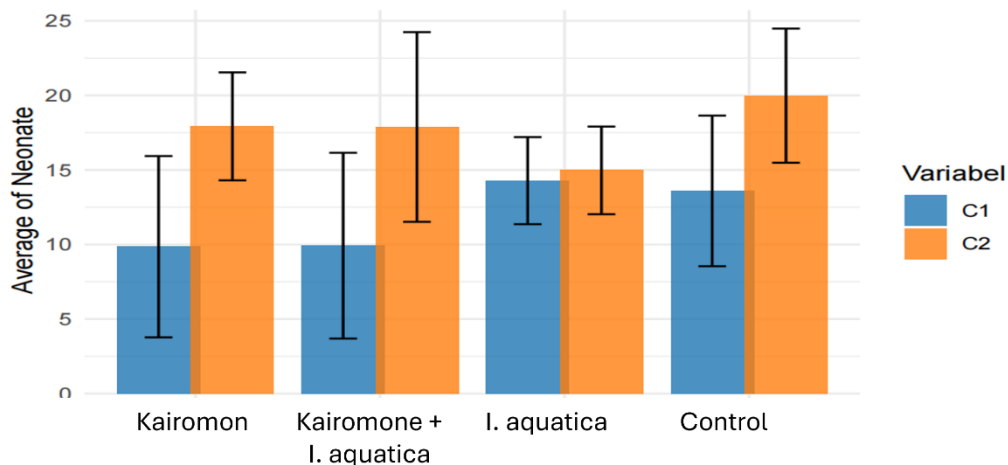


Figure 8. Comparison of average offspring number of *Daphnia* sp. among kairomone, kairomone and *Ipomoea aquatica*, *Ipomoea aquatica*, and control.

The function of water spinach as a habitat and shelter provider (Thomaz, 2017) appears to be limited to protection from visual predators that rely on physical contact. Against chemical stressors that spread throughout the water column (Rajchard, 2013), the presence of physical structures does not provide protection. The fact that *I. aquatica* treatment showed an increase in mortality (14.29%) compared to the control (2.38%) indicates that the presence of macrophytes can create microhabitat pressures, such as oxygen fluctuations or space competition, which are detrimental to zooplankton. The ecological implications are far-reaching: in ecosystem restoration efforts, planting aquatic vegetation without considering ecological balance can disrupt *Daphnia* sp. populations as a keystone species in the food web. This disruption has the potential to trigger chain reactions that threaten aquatic biodiversity, especially when interacting with anthropogenic stressors such as eutrophication and climate change. Therefore, ecosystem management requires a holistic approach that considers the complex interactions between habitat structure, population dynamics, and environmental pressures to maintain the balance and sustainability of aquatic biodiversity.

5. Conclusion

Catfish kairomones induce compensatory reproductive strategies in *Daphnia* sp. characterized by delayed early reproduction, increased later investment, and high mortality. The physical presence of *Ipomoea aquatica* does not mitigate these effects, indicating that chemical predator cues outweigh shelter availability in shaping *Daphnia* sp. life-history responses. These findings highlight the importance of chemical interactions in structuring zooplankton dynamics in freshwater ecosystems. However, the study has limitations: it used only one macrophyte and one

predator species, applied a single fixed kairomone concentration, was conducted under simplified laboratory conditions, and focused only on short-term responses over two reproductive clutches, without examining transgenerational or molecular effects. Consequently, further research under more ecologically realistic conditions is needed to confirm the generalizability of these findings.

6. Data Availability Statement

The data supporting the findings of this study are available from the corresponding author upon reasonable request.

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8. Conflict of Interests

The authors declare they have no competing interests.

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10. Authors' Contribution

MH: Conceptualization (equal), Methodology (equal), Formal analysis (equal), Investigation, Data curation, Writing – original draft (equal), Writing – review and editing (equal). **MK:** Supervision, Writing – original draft (equal), Writing – review and editing (equal). **NTMP:** Supervision (Lead), Writing – original draft (equal), Writing – review and editing (equal). **PO:** Conceptualization (equal), Methodology (equal), Formal analysis (equal), Project administration (lead), Writing – original draft (equal), Writing – review and editing (equal).

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Comparison of Chemical Oxygen Demand Removal Using Zeolite Adsorption and Floating Treatment Wetlands in Polluted River Water

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Abstract

The Citarum River in Bandung, West Java, provides drinking water, irrigation, and industrial resources for the local community. However, the recent increase in textile industry activity has reduced water quality, with textile waste contributing significantly to organic pollution in tropical inland waters. Addressing these environmental challenges requires treatment technologies to protect public health and the integrity of the region's ecosystem. This study aims to evaluate the adsorption performance of natural zeolite using the tea bag method (laboratory scale) and investigate its integration with a floating treatment wetland (FTW) system planted with *Canna indica* (mini-pilot scale) to reduce chemical oxygen demand (COD) under tropical environmental conditions. Batch experiments were conducted using real textile wastewater with an initial COD concentration of approximately 752 mg/L to reflect actual pollution levels. The tea bag adsorption system showed a maximum COD reduction of 43%, while the integrated FTW system achieved a higher COD reduction of 48% after 24 days of operation. The enhanced performance of the FTW system relative to the tea bag system results from the combined effects of zeolite adsorption, microbial biodegradation, and improved oxygen transfer facilitated by the *Canna indica* root system. Experimental findings indicate that the integrated FTW system offers a sustainable, cost-effective, and nature-based approach for improving water quality in tropical inland waters affected by industrial pollution. Nevertheless, additional optimization is required to achieve compliance with regulatory discharge standards.

1. Introduction

Rivers are essential freshwater resources that support human activities and aquatic ecosystems. However, increasing anthropogenic pressures have led to a decline in river water quality, particularly in rapidly industrializing regions (Ahsanul et al., 2024). Various anthropogenic activities along riverbanks can significantly degrade water quality and accelerate its decline (Anh et al., 2023; Khairuddin et al., 2019). One primary source of pollution from human activity is the textile industry. This is what happened to the Citarum River located in Bandung City, West Java Province, Indonesia (Ahsanul et al., 2024). The Citarum River has been significantly impacted by the textile industry, with thousands of textile factories lining its banks (Oktaviyani et al., 2023).

Industrial textile wastewater is a significant source of organic pollution, containing a complex mixture of chemicals, dyes, salts, detergents, softeners, acids, bases, finishing agents, and other synthetic substances that are difficult to remove. Due to these complex substances, textile wastewater often has high Chemical Oxygen Demand (COD) levels (Suyata, 2008), making treatment before discharge challenging and contributing to suspended solids and reduced water quality (Abdissa & Beyecha, 2021). Untreated textile effluents severely affect the flora and fauna in nearby freshwater bodies. This situation is of particular concern, as many textile industries still lack effective wastewater treatment prior to discharge (Khandare et al., 2013). Textile effluents, when discharged directly into freshwater bodies, deplete



dissolved oxygen, negatively impacting aquatic life and human health (Haydar et al., 2011). Effective COD treatment methods are therefore essential.

Adsorption is generally considered an up-and-coming method for removing pollutants from wastewater, due to its low cost, high efficiency, and stability. Activated carbon as an adsorbent is commonly used to remove toxic pollutants from textile wastewater (Almadani, 2023), but it is also costly (Mahmoud et al., 2020). In line with this, many efforts have been made to develop effective local activated carbon from cassava peel, orange peel, bagasse fly ash, bamboo, and avocado seed (Dehghani et al., 2018; Niazi et al., 2018; Rizzo et al., 2019; Fito et al., 2019; Choong et al., 2020). However, these adsorbents present major limitations in treatment performance, preparation time, cost, regeneration difficulty, and adsorption capacity. Further investigation of adsorbents is warranted, as many are constrained by insufficient integration with biological treatment systems, highlighting the demand for more reliable and scalable alternatives. In response to these limitations and the lack of industrial-scale validation, recent research has shifted the focus from activated carbon to zeolites to identify more effective adsorbents. Zeolite and clay adsorbents exhibit higher adsorption efficiency than activated carbon, thanks to their large surface area and unique properties (Titchou et al., 2020).

Prior to zeolite being integrated into the FTWs, preliminary adsorption tests were conducted under controlled conditions to evaluate the adsorption performance. The tea bag method, applied on a laboratory scale, was used to measure the adsorbent. Although this method is not commonly used, it offers a simple, controlled approach to assess adsorption efficiency, the attainment rate of maximum capacity, and desorption potential. In the context of FTWs, persistent emergent plant most commonly Indian shot (*Canna indica*) is frequently utilized. *Canna indica* has been identified as an optimal species for application in FTWs due to its ability to tolerate a wide range of soil types and pH conditions (Bhutiani et al., 2019). Beyond its adaptability, *Canna indica* contributes to the aeration of the root zone and nutrient removal in wetland microcosms, thereby enhancing microbial activity and accelerating the degradation of organic matter (Zhang et al., 2007). Furthermore, this species increases filtration efficiency and substrate porosity through its extensive root system, thereby facilitating the uptake and sequestration of essential nutrients in plant tissues via diverse rhizosphere processes (Fraser et al., 2004), (Ramesh et al., 2017). Additionally, *Canna indica* delivers oxygen to the rhizosphere, promoting the proliferation of aerobic bacteria responsible for contaminant degradation (Yang et al., 2007; Zhu et al., 2017) and exudes root-derived compounds that serve as substrates for biofilm development (Jamwal et al., 2021). While numerous studies have investigated zeolite and *Canna indica* separately for treating industrial wastewater,

focusing primarily on specific pollutants such as dyes or heavy metals (Zulti et al., 2025). Still, no research has combined the adsorption mechanism of zeolite with the FTW system using *Canna indica*.

To address the research gap, this study systematically evaluated COD removal performance using two methods in a staged approach. In the initial stage, zeolite performance was evaluated using a tea bag system. The second stage continued by integrating zeolite with *Canna indica* in the FTWs. This sequential method can demonstrate the differences between the two methods and explore the mechanistic interactions of adsorption and microbial degradation under tropical environmental conditions. To the best of our knowledge, no previous study has systematically compared preliminary zeolite adsorption performance using the tea bag method with its subsequent integration into FTWs for COD removal under tropical conditions. Therefore, the objectives of this study are: (1) to evaluate the efficiency of natural zeolite in COD removal using a tea bag adsorption system; (2) to compare COD removal performance between the tea bag system and the integrated FTW system planted with *Canna indica*; and (3) to evaluate the potential application of the integrated system as a natural-based supplementary treatment to enhance organic pollution mitigation in tropical inland waters.

2. Materials and Method

2.1 Textile Wastewater Characteristics

This research was conducted at the Testing Laboratory at the Limnology and Water Resources Research Centre, BRIN, Bogor, Indonesia, from March to July 2025. The materials used included zeolite powder, textile wastewater, and COD analyzers. Textile wastewater samples were collected from PT.X, a textile factory located in Bandung, West Java, Indonesia. The wastewater originated from the final production stage and represented the combined effluent from the entire textile process. Preliminary testing for waste characterization showed COD levels of 752 mg/L in the final production wastewater. Preliminary colour testing showed 359.1 PCU in final production waste, indicating the presence of residual dyes and organic compounds typical of textile effluents. In this test, the parameters tested were the COD parameters in the final production textile wastewater. Before use in the experiment, the final textile production wastewater was diluted to 50%. After dilution, the COD concentration was 596 mg/L; this value was used in all experiments.

The natural zeolite used in this study was obtained from Cikembar, Sukabumi, Indonesia, and was prepared to a particle size of 100 mesh prior to use. The *Canna indica* used in the FTWs was approximately 3 months old, with uniform initial heights of 40-50 cm. In the FTW experiment, *Canna indica* was first acclimatized for 50 days under controlled conditions to allow the plants to adapt to the media and environment, maximizing root integration into FTWs. All experimental treatments were

repeated three times to ensure data reliability and reproducibility (triplication).

2.2. Zeolite Preparation and Tea Bag System

Natural zeolite powder wrapped in a tea bag, with a zeolite dosage of 105 g and 100 mesh particle size. These adsorption experiments were conducted in a batch system, in which the zeolite-filled tea bag was immersed in 1 L of 50% diluted final production textile wastewater in a glass beaker, with a COD concentration of 596 mg/L. During this lab-scale experiment, there was no water flow, allowing adsorption to occur under static conditions. The contact time was varied over 18 days, with sampling conducted at 0, 3, 6, 9, 12, 15, and 18 days to observe COD removal trends and determine the adsorption equilibrium time.

2.3. FTW Experimental Set-up

The FTWs included four sequential treatment configurations in a batch system, as shown in Figure 1. Each treatment was performed in triplicate. The configurations were: (i) tank K (control), with textile wastewater and an aerator but no zeolite or *Canna indica*; (ii) tank ZT, with zeolite and *Canna indica* physically separated from the zeolite, as shown in Figure 2a; (iii) tank ZsT, with zeolite integrated within the *Canna indica* root zone, as shown in Figure 2b; and (iv) tank T (plant only), with *Canna indica* but no zeolite.

Each tank had a capacity of 95 L and was filled with 50% diluted final-production textile wastewater, resulting in an initial COD concentration of 596.5 mg/L. The FTWs operated in batch mode without continuous inflow or outflow. Samples were collected at 0, 3, 6, 9, 12, 15, 18, 21, and 24 days to evaluate COD removal over time.

The natural zeolite used in the ZT and ZsT tanks was sourced from Cikembar, Sukabumi, with a particle size of 100 mesh and packaged in 105 g tea bags. For each 95 L tank, 950 g of zeolite was used in both ZT and ZsT tanks. In the ZT tank, zeolite tea bags were placed separately from the plant roots to allow independent adsorption. In the ZsT tank, zeolite tea bags were placed adjacent to the root zone to enable direct interaction among adsorption, root oxygenation, and microbial activity in the rhizosphere. In the FTWs, supporting parameters such as DO, temperature, pH, and salinity were also measured to control system conditions during testing.

The number of *Canna indica* plants varies by treatment: the ZT tank contains 6 plants, while the ZsT and T tanks each contain 9 plants. This aims to evaluate the potential dominance of zeolite adsorption over plant phytoremediation. Meanwhile, in the ZsT tank, the interactions between the adsorbent and plant roots during rhizosphere formation and microbial activity for pollutant degradation will be examined. This setup is expected to provide a clear distinction between adsorption mechanisms and plant mechanisms.

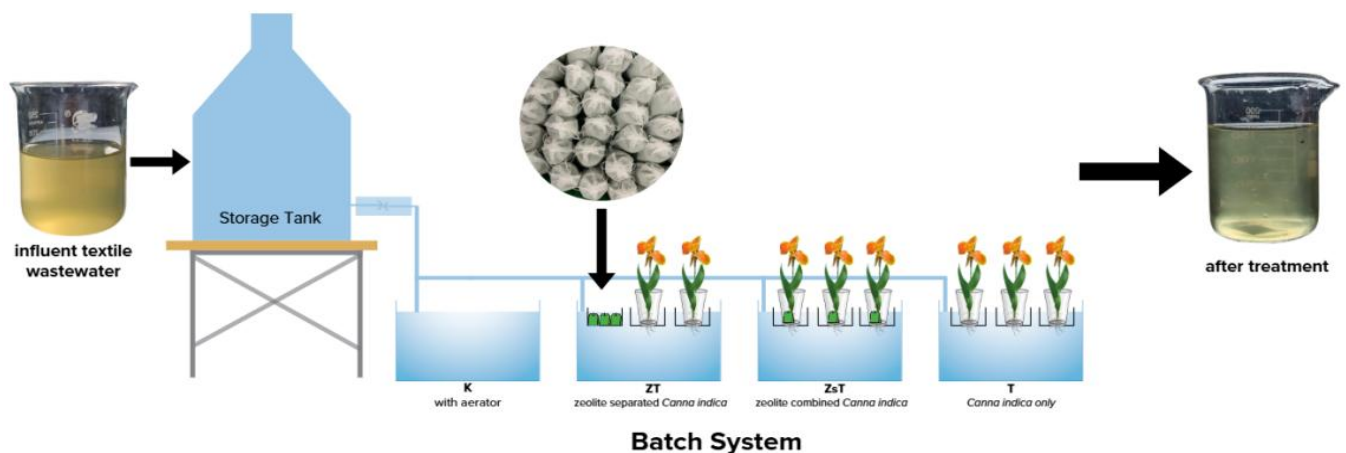


Figure 1. Graphical abstract of the experimental batch system process: from influent textile wastewater storage to the four sequential FTW treatment configurations—control (K), separated zeolite-*Canna indica* (ZT), combined zeolite-*Canna indica* (ZsT), and plants only (T)—resulting in the post-treatment effluent.

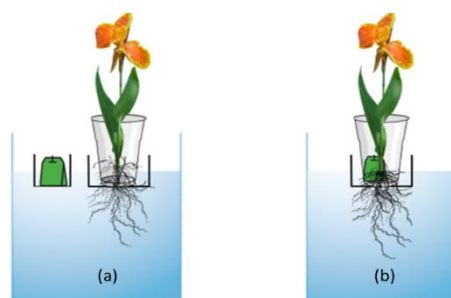


Figure 2. Structural comparison of the FTW design configurations: (a) ZT system with zeolite separated from the roots, and (b) ZsT system with zeolite integrated directly with the *Canna indica* roots.

2.4. Analytical Procedure

COD measurements were performed in the laboratory according to the Standard Method using the dichromate reflux method (APHA, 2017). All measurements were conducted in triplicate, and the average values were reported. The COD removal efficiency value after treatment can be calculated using Equation 1.

$$\%R = \frac{C_i - C_t}{C_i} \times 100\% \quad (1)$$

C_i is the initial COD concentration (mg/L), C_t is the COD concentration at time t (mg/L), and $\%R$ represents the COD removal efficiency.

2.5. Data Analysis

Test results were analysed using descriptive statistics, including mean values and SD, to evaluate trends in COD removal efficiency, adsorption capacity, and changes in tropical environmental parameters (pH, temperature, dissolved oxygen, and salinity). Differences among FTWs variations (K, ZT, ZsT, and T) were assessed using ANOVA. To compare the COD removal performance between the

tea bag and FTW systems, an independent t-test was used.

3. Results and Discussion

3.1. COD Reduction Using Zeolite in the Tea Bag System

The effect of contact time on COD removal from textile wastewater using the tea bag method with zeolite is shown in Figure 2. The COD concentration decreased with increasing contact time until the sixth day. The COD concentration decreased from 596.5 mg/L to 345.75 mg/L, resulting in a removal efficiency of 33%. However, the concentration increased again on the 9th day, after which the removal efficiency decreased. The graph shows that the zeolite is most effective until the 9th day. This phenomenon was statistically significant ($p < 0.001$), indicating that zeolite has a limited adsorption capacity (Castro et al., 2021). Although efficiency increased again on day 18 (43%), this unstable trend underscores the need for more sustainable system integration. In addition, the absence of oxygen or anaerobic conditions in the tea bag system results in zeolite saturation due to the lack of natural biological regeneration (Skouteris et al., 2020).

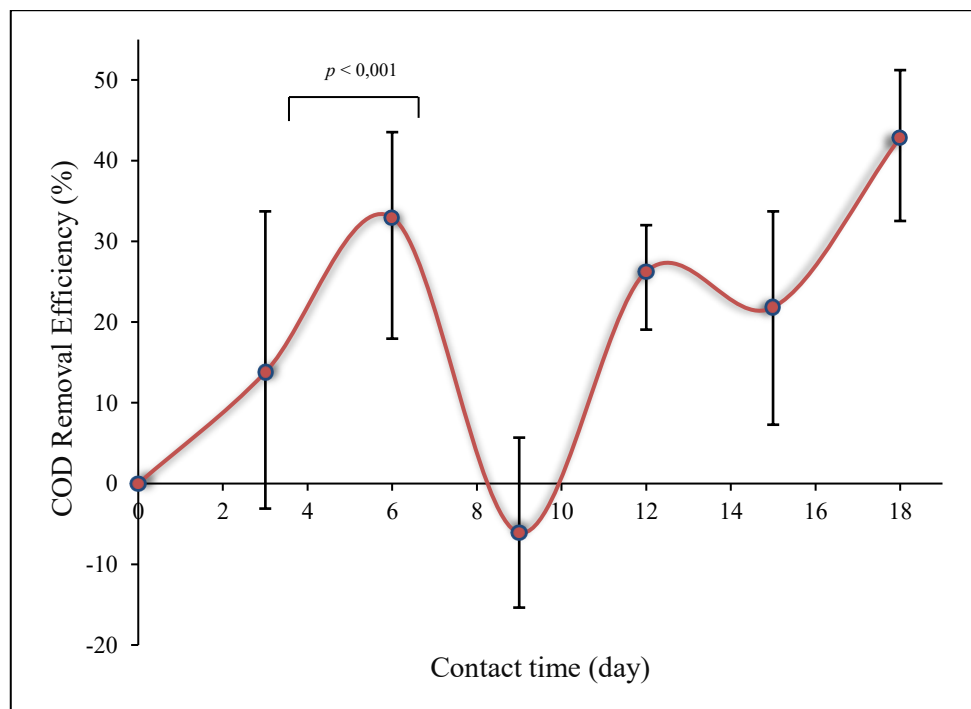


Figure 3. COD reduction trend over contact time and removal efficiency

Although dissolved oxygen was not directly measured in the tea bag system, the absence of aeration suggests that the system may have operated under oxygen-limited conditions. Therefore, the tea bag system was combined with an integrated FTW system for *Canna indica* plants, and an aerator was added to the K tank. This is consistent with the literature (Zhang et al., 2026), which indicates that aeration can increase system efficiency and improve organic biodegradation and treatment effectiveness. Another variation in the FTW system provides dissolved

oxygen to plants through photosynthesis in leaves and through the diffusion of atmospheric oxygen (Stefanakis & Tsihrantzis, 2012).

3.2. COD Reduction in Zeolite–*Canna indica* Integrated System

This is the result of textile wastewater treatment using an integrated FTWs with zeolite and *Canna indica* plants. Figure 3 shows the system's degradation rate as a function of COD concentration and contact time. The

initial COD concentration of 596.50 mg/L became 437 mg/L in tank K, 242 mg/L in tank ZT, 286 mg/L in tank ZsT, and 322 mg/L in tank T significant interaction ($p = 0.167$), indicating consistent trends across treatments. Over the 24-day testing period, FTWs produced varying COD reduction efficiencies across treatments. The ZT tank achieved the highest removal efficiency RE at 48% on day 24, followed by the ZsT tank at 37%, the T tank at 30%, and the K tank at 16%.

Two-Way ANOVA confirmed that COD reduction was significantly affected by contact time and treatment type ($p < 0.001$), with no

degradation. The mechanism in this tank begins with zeolite adsorption, which serves as the primary COD removal mechanism, while *Canna indica* rhizofiltration regenerates the system over a longer timescale. The zeolite surface provides a surface for microorganisms to form a biofilm. Separation from the roots prevents nutrient competition, allowing the zeolite to form a stable biofilm for COD biodegradation (Ho et al., 2024). However, in the ZsT tank, competition occurs between organic compounds and plant roots for nutrients (Yugi Prasetyo et al., 2024).

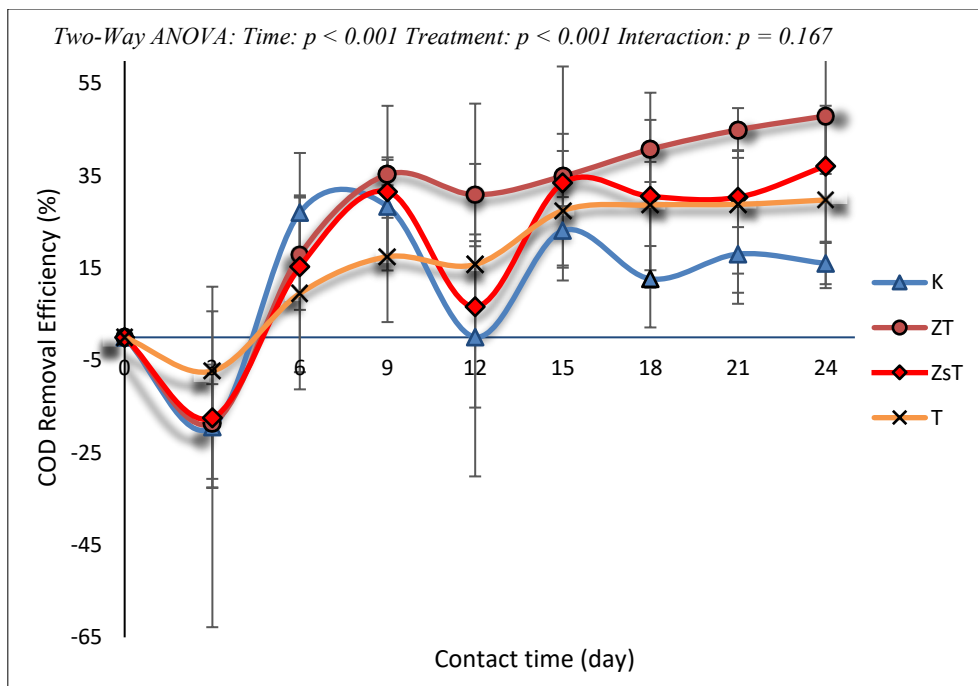


Figure 4. COD removal efficiency trend over contact time.

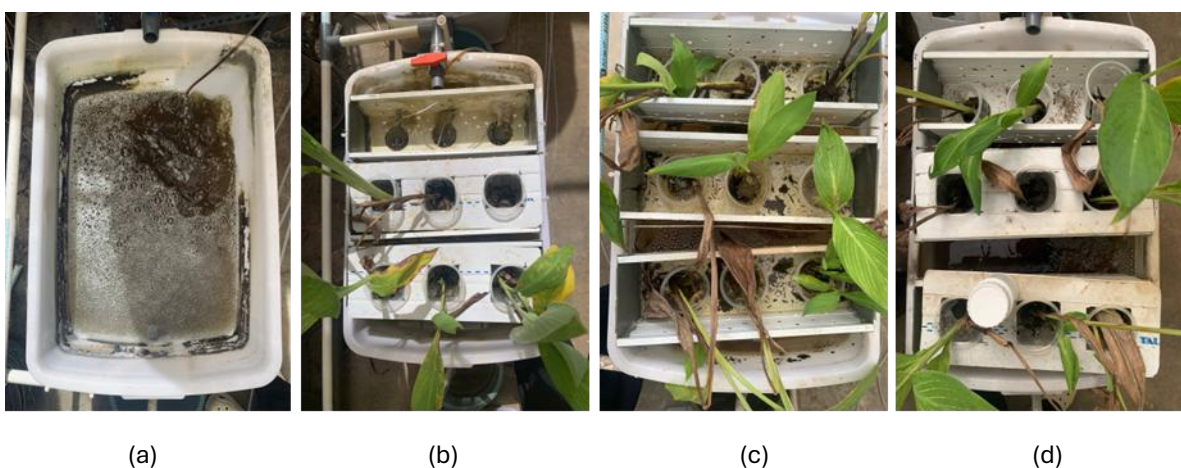


Figure 5. Top-view images of the four sequential treatment configurations during the batch experiment: (a) K tank, (b) ZT tank, (c) ZsT tank, and (d) T tank.

Zeolite binds nutrients such as NH_4^+ and reduces their availability to *Canna indica* roots. Biofilms develop on the zeolite surface and in the root environment,

spreading their formation. This also triggers a decrease in their dynamic concentration. This scattered formation inhibits optimal COD biodegradation, emphasizing the

importance of separating the zeolite-root zone to minimize biofilm dispersion (Malakar et al., 2023). The temporal variation of COD removal efficiency observed in all treatment configurations is presented in Figure 4. The role of *Canna indica* plants in the ZT and ZsT tanks is evident in their roots. *Canna indica* provides an area for microorganisms to grow, reduces pollutants in wetland systems, and creates highly aerobic conditions (Sharma et al., 2014). Visual observations of the four treatment configurations at the end of the batch experiment are shown in Figure 5.

Table 1. Comparison of removal efficiencies (% Removal at (t15) and highest RE) and statistical significance between the tea bag substrate and the FTW (ZT tank) systems.

System	% Removal (t15)	Highest RE	Statistical Comparison
Tea bag	-6%	43%	$p = 0,046$
FTW (ZT Tank)	31%	48%	

Table 1 above shows the results of textile wastewater treatment using the tea bag method (zeolite adsorption) and the integrated FTW system with *Canna indica*. An independent t-test with unequal variances was used to compare the tea bag method and the FTWs (ZT Tank) at t=15. The FTW system demonstrated a significantly higher removal efficiency (31.97%) than the tea bag method (-5.28%) ($p = 0.046$), which suggests superior and more consistent treatment performance. Given that testing of zeolite tea bags and FTW was conducted simultaneously, their removal efficiency values could be compared on the 15th day of testing. On the 15th day, zeolite tea bags reduced COD by -6%, while FTW (ZT Tank) achieved a removal efficiency of up to 48%. This indicates a difference between zeolite in the tea bag method and zeolite integrated with *Canna indica* in the FTW system. The zeolite adsorption process plays a role in reducing organic matter, in line with research conducted by (Ojstrsek & Fakin, 2011). The highest RE values achieved by each system were 43% and 48% for teabags and FTW (ZT Tank), respectively. Several processes contribute to reducing organic matter, including biodegradation by microorganisms in the media and root zone, as well as assimilation by plants (Trifando et al., 2022). Waste treatment in the FTW system integrated with zeolite adsorbents also depends on plant roots and microbes. According to J. Zhang et al. (2014), under both aerobic and anaerobic conditions, organic matter is removed by bacteria and other microbes. Submerged plant roots provide a rhizosphere that supports the degradation rate of organic compounds by microbes. Plant activity enhances synergy with zeolite adsorption during aerobic COD degradation. Additionally, high oxygen transfer in the system aids the nitrification process and organic waste removal (Liang et al., 2017; Hdidou et al., 2022). Plants act as areas that provide rhizomes, which serve as containers for microorganisms to grow. Radial Oxygen Loss (ROL) facilitates the transfer of oxygen to the rhizosphere, thereby enhancing nutrient absorption. The root system plays a vital role in transferring oxygen from

3.3. Comparative Analysis and Mechanism Interpretation

Based on processing results using a tea bag system with an integrated FTW system, the data show that both can reduce COD pollutants in wastewater with varying levels of efficiency. The comparison data are shown in Table 1, using data from the FTW system for one of the treatment variations, namely ZT.

the leaves to the roots. Oxygen is obtained from the air through photosynthesis, then transported through the aerenchyma in plants to the roots and the rhizosphere (Trifando et al., 2022).

In general, plants used in wetlands in tropical and intertropical regions are macrophytes characteristic of natural wetlands, such as *Phragmites australis* and species of *Typha*, *Scirpus*, and *Cyperus* (Sandoval et al., 2019). However, ornamental vegetation is a promising alternative due to its aesthetic and commercial value, among which the *Canna* species stands out (Sandoval et al., 2019; Tejeda et al., 2022). *Canna indica* plants in the FTWs play a role in supplying oxygen through the intercellular space, known as aerenchyma. Research (Li et al., 2013) explains that aerenchyma functions to channel oxygen from the air to the leaves, stems, and roots/rhizosphere. Many microorganisms utilize dissolved oxygen in the rhizosphere to degrade organic matter. Photosynthesis also plays a role in supplying oxygen from the leaves to the roots. Increased dissolved oxygen makes the system more aerobic, which in turn increases the rate of degradation by microorganisms both in the root zone and on the adsorbent. Sufficient oxygen supports the decomposition of organic matter and the metabolism of microorganisms (Trifando et al., 2022). Research (Li et al., 2013) also shows that microorganisms break down organic matter into simpler forms that plants can then utilize as nutrients. Microorganisms can decompose organic matter because they have specialized enzymes during metabolism that break it down into simpler forms. Microorganisms decompose organic matter to produce energy, synthesize cellular components, respire, and move (Hammer & Mark J. Hammer Jr., 2014).

3.4. Application Potential in Indonesian Tropical Waters

As a consideration for potential application in Indonesian tropical waters, one example of COD levels in the Citarum River, in Bandung, West Java, was taken

specifically in a tributary, namely the Cibodas River (Fig. 6). This concentration falls within the range of previous studies, which reported values of 114-610 mg/L (Sumantri & Rahmani, 2020).

Data collection was conducted at several monitoring stations along the Citarum River watershed, as presented in Figure 6a. Among these stations, point 3D was selected for COD analysis and detailed assessment due to its proximity to industrial activities. Figure 6b provides a high-resolution satellite view of the sampling location, illustrating the surrounding industrial facilities and the

river's visual condition, thereby providing additional field-scale context for interpreting water quality conditions. This visual evidence (b) complements the spatial distribution map shown in (a) and supports selecting site 3D as a representative location for COD assessment.

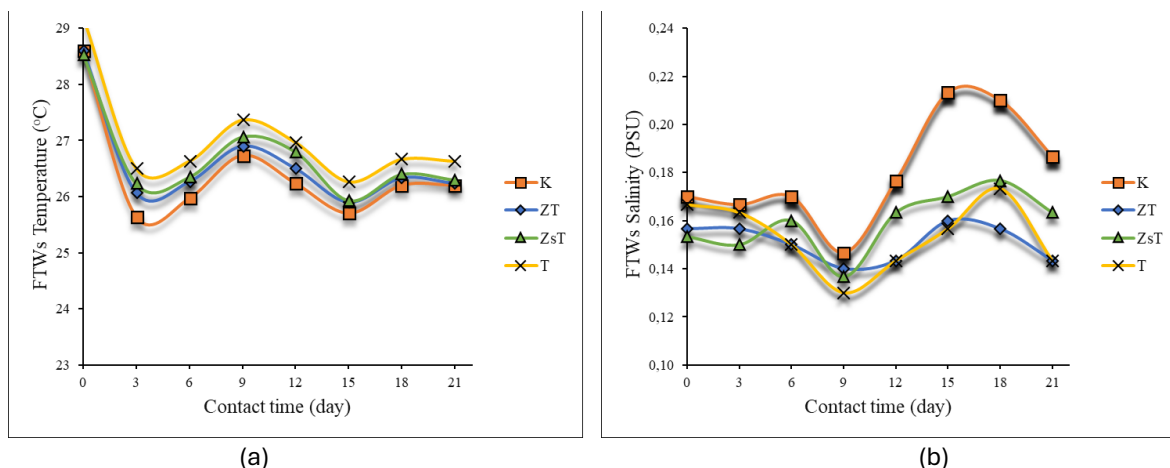
The concentration in the Cibodas River, 397 mg/L, was close to the textile waste concentration used in this test, with an average of 475 mg/L per tank. As an innovative treatment method, the COD reduction achieved by the FTW system demonstrates potential for application in additional in-situ treatment.



Figure 6. (a) Location of sampling sites (3A–3D) along the Cimannde River and surrounding land-use characteristics. (b) High-resolution satellite image showing the exact location of sampling site 3D selected for detailed field observation and water quality assessment.

The FTW system reduced COD concentrations to 242 mg/L, achieving a 48% removal efficiency. Although this value does not meet the water quality standard of 150 mg/L (PermenLHK No. 14 Tahun 2019 tentang Baku Mutu Air Limbah Industri Tekstil), it represents a significant reduction in the organic load. For polluted river waters such as the Cibodas tributary, COD reduction is an important first step in restoring dissolved oxygen levels and improving ecological conditions. The tributary estuary also has low hydraulic conductivity, which supports prolonged contact between the polluted water and the roots of FTW plants (such as *Canna indica*), increasing COD rhizofiltration efficiency before pollution spreads to the main river (Yang et al., 2022).

Therefore, the FTW system can be positioned as a treatment technology, implemented in strategic locations such as tributary estuaries, industrial discharge points, or low-flow river sections to reduce pollution before it is transported further downstream. This shows that the results of this FTW system test have the potential to be applied on a field scale in Indonesian waters with tropical characteristics. This wetland treatment is promising, robust, and compact in tropical climates (Trein et al., 2019) compared to non-tropical regions (Molle et al., 2005). Figure 7 shows graphs of pH, temperature, DO, and salinity as an overview of the climate during the FTW testing.



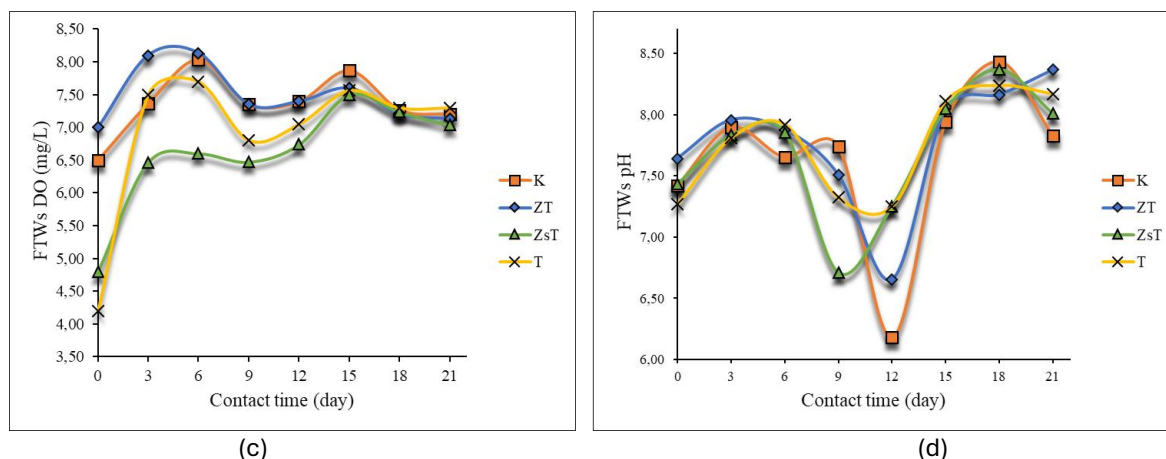


Figure 7. Water quality fluctuations as a function of contact time in the batch FTW systems: (a) temperature, (b) salinity, (c) dissolved oxygen (DO), and (d) pH profile.

The temperature in the ZT tank showed no significant day-to-day differences, decreasing only on the third day of data collection from 29 °C to 26 °C. Consistent with the increase in RE observed at $t = 9$ in the ZT tank (Figure 3), the temperature increased from 26 °C to 27 °C. High temperatures enhance enzymatic activity and accelerate the biodegradation of organic matter (Wang et al., 2021). DO level remained relatively stable at 7-8 mg/L. This condition aligns with the sustained COD removal observed in the FTWs, particularly in the ZT tank, where DO levels were relatively stable compared to the other tanks (ZsT, T, K). This supports the ZT tank's superiority over other tank variations in COD degradation. Adequate DO supports microbial degradation and prevents anaerobic conditions that can lead to COD desorption, as observed in the tea bag system. Consistent DO levels indicate effective oxygen transfer and are essential for ongoing biodegradation (Murti et al., 2021). pH ranged from 7 to 8, which is optimal for microbial activity and zeolite adsorption. Previous studies have shown that biodegradation efficiency increases significantly at pH values above 7.0 (Ado et al., 2025). This aligns with findings at pH on $t = 12$. Decreasing the pH to pH = 6 resulted in a decrease in RE in all tank variations. Acidic pH can inhibit microbial-assisted COD degradation because the microbial community involved in organic matter degradation is more active under neutral to slightly alkaline conditions (Rohim et al., 2015). Plants and microorganisms in wetlands require a pH between 6 and 8 to support the biochemical processes that transform and degrade contaminants (Monteagudo-Hernández et al., 2024). The water salinity levels were relatively low, ranging from 0.14 to 0.17 ppt, indicating a biological balance between microbes and plants in the system, with both actively participating in degradation. Increasing salinity is expected to reduce the efficiency of organic matter removal (Frank et al., 2017). This is evidenced by the finding that increasing salinity levels in the ZsT tank at $t=18$ were accompanied

by a decrease in RE COD (Figure 5). Under high salinity conditions, microbes experience metabolic disruption due to cell shrinkage, resulting in a decreased rate of pollutant degradation (Zhang et al., 2023). Temperature, pH, DO, and salinity are factors that need to be considered if this system is to be applied on a field scale, in situ treatment of tropical climate waters.

Given the environmental characteristics of the study area, the FTW system shows potential for future application and scale-up in direct river water treatment. The tropical climate provides favorable conditions that enhance plant growth and stimulate microbial activity, both of which are essential for effective pollutant degradation. High sunlight exposure maintains warm water temperatures, which in turn support synergistic processes such as adsorption, biofilm formation, and rhizofiltration. It is also important to emphasize that the textile wastewater used in this study originated directly from an industrial outlet rather than being synthetic wastewater, which likely increased the treatment challenge due to its more complex and variable composition.

Although this study focused on textile wastewater treatment, the system could also treat other types of wastewaters. The treatment mechanisms occurring in FTWs, such as zeolite adsorption and rhizofiltration, demonstrate potential applications for wastewater treatment that also contain organic matter. Previous studies have reported successful applications of FTW systems for municipal wastewater treatment (Faulwetter et al., 2011), aquaculture wastewater treatment (Rehman et al., 2019), and agricultural runoff management (Spangler et al., 2019), all of which contain organic matter, supporting the broader relevance of this approach. However, the efficiency of an integrated FTW system for other types of wastewaters will depend on the wastewater's characteristics, such as salinity, nutrient content, acidity level, and other specific characteristics (Oliveira et al., 2021). Therefore, FTWs is best

positioned as a treatment or post-treatment option for wastewater containing organics, particularly in tropical regions, as it supports the biological processes within the system.

4. Conclusion

This study demonstrates the adsorption capacity of zeolite on tea bags and its integration with an FTW using *Canna indica* to reduce COD levels in textile wastewater. The highest RE value in the tea bag system was 43% via direct adsorption, and in the FTW system, it was 48% in the ZT treatment tank, with a final COD concentration of 242 mg/L. However, the final treatment results do not meet industrial wastewater quality standards. Therefore, further optimization of the FTW system's wastewater treatment is needed. Despite these limitations, the findings indicate that zeolite-integrated FTW has potential for application in tropical river environments, where adsorption, rhizofiltration, and microbial processes contribute to pollutant degradation. For field-scale applications, optimizing zeolite dosage, adjusting contact time, and increasing wastewater flow rate are required. Future studies should also evaluate other textile wastewater parameters, such as color, TSS, and ammonium, to ensure a comprehensive assessment of treatment performance. Overall, this study provides initial evidence on the feasibility of zeolite-integrated FTW and sheds light on the development of effective, nature-based, and scalable wastewater treatment solutions.

5. Data Availability Statement

The data included and used in this study are not confidential and are available upon request.

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7. Conflict of Interest

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

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9. Author Contribution

FZ contributed to the conceptualization of the study, manuscript outline, and review of the manuscript. RN contributed to data analysis, graphical interpretation, and review of the manuscript. CC conducted experimental work, performed data analysis, prepared figures, and drafted the manuscript. All authors read and approved of the final manuscript.

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Assessing Data Quality in Real-Time River Water Quality Monitoring Using Multi-Parameter Indicators: A Case Study of Ciliwung and Cisadane, Indonesia

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Abstract

Water pollution has become a complex environmental challenge in rapidly urbanizing Southeast Asia. Indonesia has deployed over 347 online river water quality monitoring stations (ORWQM) in several watersheds to provide real-time data for pollution control and informed ecological decision-making. However, problems have arisen with some of the longstanding stations, including equipment degradation, data loss, sensor malfunctions, and inconsistencies, raising concerns about the reliability of these datasets. This study aims to address these issues by using a hybrid Total Data Quality Management (TDQM), Online Data Quality Evaluation Model (ODQEM) framework to evaluate and improve the data quality at four stations (Manggarai, Kelapa Dua, Pasar Baru, and Empang Dam) representing the upstream and downstream areas of the Ciliwung and Cisadane Rivers. The workflow consists of four steps: (i) defining the relevant data quality dimensions; (ii) measuring data quality using completeness index, range validity, accuracy, uniqueness, and stability index; (iii) analyzing cross-parameter and cross-station patterns to identify potential failure modes and governance gaps; and (iv) translating findings into targeted calibration, cleaning, and maintenance actions. The results show strong structural integrity; completeness indicators reach 100%, and uniqueness exceeds 99.9%, indicating robust acquisition and temporal consistency. However, functional reliability varies widely across parameters and station locations. Completeness indicators for non-zero values show systematic zero values for the DO probe (1.02%) and nitrate (0.19%) at Empang Dam Station; TDS (9.57%) at Pasar Baru Station; ammonia (60.02%) at Kelapa Dua Station. Overall stability is high for physical probes (temperature, pH), but low for chemical/ion probes at some stations (salinity 1.47%–6.62%; TDS 5.49%–10.92%; ammonia 30.20%–71.40%). Ion sensors also show higher risks, including low validity or accuracy for nitrate at Kelapa Dua Station (61.67%). These findings indicate that the dataset appears complete and valid, but still contains substantial functional uncertainty due to several zero-value parameters and unstable sensor behavior. Because the evaluation is limited to four stations, these findings should be interpreted as an in-depth case study rather than a complete representation of the ORWQM network in Indonesia. The proposed TDQM–ODQEM Data Governance Model offers a replicable method for improving online and real-time environmental monitoring, helping bridge the gap between technological performance and policy design in sustainable river management.

1. Introduction

River pollution, caused by various pollutants, significantly impacts aquatic ecosystem degradation,

reduces water quality, and poses a threat to human health (Lin et al., 2022). One preventative measure to control pollution is the implementation of continuous



monitoring and strict supervision to maintain the river water quality status. The online river water quality monitoring (ORWQM) system consists of monitoring stations installed along riverbanks. This system is equipped with multiparameter sensors that continuously measure and transmit water quality data in real-time to a central database via the internet. As of 2024, 347 ORWQM stations have been established across several watersheds (DAS) in Indonesia (KLHK, 2024). This monitoring plays a crucial role in water resource management, providing insight into river water conditions and tracking changes over time (Soares et al., 2020). Furthermore, long-term water quality monitoring is crucial for detecting ecological changes in river ecosystems and ensuring compliance with water quality standards (Meyer et al., 2019). The real-time data obtained from this monitoring system is invaluable for developing innovative studies that capture dynamic temporal variations, including water quality prediction, assessment, and environmental management (Zhang & Thorburn, 2022). This information provides a crucial foundation for developing policies and planning strategies for water quality management at the national, provincial, and local levels of government (Adu-Manu et al., 2020). Therefore, the main challenge is not only ensuring the continuous transmission of data but also establishing trust in the ORWQM data, analytically, and in its suitability for regulatory interpretation, early warning, and evidence-based environmental management. This sharper focus addresses the practical need to assess whether the available data in the system is sufficiently reliable to support decision-making in environmental policy.

A common problem in real-time water quality monitoring is data loss, often caused by equipment failure, limited network coverage, or data corruption (Zhang & Thorburn, 2022). As Internet of Things (IoT)-enabled water quality monitoring expands, the likelihood of system and network failures also increases significantly (Zhang et al., 2021). These failures can lead to lost data, duplicate records, or outliers, which reduce data quality and affect the integrity of analysis results. Data quality is defined as the extent to which data is suitable for its intended use in operations, decision-making, and planning (Mansouri et al., 2023). Inaccurate sensor data can cause misinterpretation of environmental conditions, resulting in inappropriate policy responses or inefficient resource allocation (Teh et al., 2020). Furthermore, poor data quality directly impacts the effectiveness of organizational processes. It results in misinformation, affects reporting accuracy, increases operational risks, and often requires costly data reprocessing or cleansing efforts from a strategic perspective (Wijayanti et al., 2018; Bowo et al., 2019). Data quality is a key factor in evidence-based decision-making, especially in fields such as environmental protection, urban water management, and public health. High-quality data supports accurate modeling, risk

forecasting, and automation systems in innovative environmental initiatives. However, maintaining this quality in an IoT environment demands robust system architecture, real-time validation protocols, standardized data formats, and coordination among stakeholders (Karkouch et al., 2016; Pezoulas et al., 2019). In Indonesian river systems, particularly in urban areas like Jakarta and Tangerang, pollution levels remain high due to increasing discharges of domestic waste, industrial waste, and urban runoff (Sulthonuddin et al., 2019). Therefore, high-quality monitoring data is essential for sustainable watershed (DAS) management and mitigation programs. However, systematic research on the quality of multidimensional ORWQM data in Indonesia remains limited, especially studies that distinguish between aspects such as structural data integrity (including completeness and timestamp uniqueness) and the reliability of operational sensors (including stability and parameter anomaly detection) in IoT-based monitoring systems. Most previous studies have focused more on the technical aspects of water quality monitoring or analysis itself (Meyer et al., 2019; Soares et al., 2020), but have not explicitly addressed the integrity and reliability of the data generated by the system. Although the literature on water quality monitoring systems is extensive, most focus on hydro chemical interpretation, pollution source analysis, and sensor placement strategies, rather than on evaluating monitoring data quality as an asset supporting decision-making (Chapman, 1996; Behmel et al., 2016; Meyer et al., 2019; Soares et al., 2020). Many IoT-based environmental monitoring studies highlight sensing architecture and communication systems but provide less of a multidimensional framework to assess whether the generated time-series data remains reliable during long-term operation (Karkouch et al., 2016; Adu-Manu et al., 2020; Teh et al., 2020). As a result, operational data issues such as stagnation, instability in certain parameters, and records that appear complete but are unusable are often treated as technical disruptions rather than methodological and governance problems that affect policy credibility.

Data quality is crucial for the effective use of information in policy formulation, managerial decision-making, and responding rapidly to pollution and changing environmental conditions. Furthermore, the knowledge gap is evident in the lack of studies employing a comprehensive conceptual framework, such as Total Data Quality Management (TDQM), that enables comprehensive data assessment across multiple data quality dimensions (Batini & Scannapieco, 2016). However, TDQM itself lacks an operational mechanism for assessing sensor performance in real time. In dynamic environments such as tropical rivers.

This gap is particularly pronounced in Indonesia, where ORWQM systems have strategic policy relevance, but published studies systematically assessing ORWQM data quality using a multidimensional framework are

limited. What is needed is not only reporting descriptive sensor performance, but also identifying which data quality dimensions are structurally strong, which are operationally weak, and how these weaknesses should be considered in maintenance priorities and institutional governance.

To close this gap, this paper integrates TDQM with the Online Data Quality Evaluation Model (ODQEM), which was initially formulated for continuous IoT sensor networks. ODQEM provides parameter-specific temporal metrics such as the Stability Index (SI) and Range Validity (RV), which quantify sensor reliability under fluctuating field conditions. This hybrid approach enables multi-level evaluation, utilizing TDQM for strategic data governance and ODQEM for real-time operational assessment.

Therefore, the objectives of this study are to evaluate the quality of online river water quality monitoring (ORWQM) data from selected stations in the Ciliwung and Cisadane Rivers using an integrated TDQM–ODQEM framework to identify key weaknesses affecting data reliability and to support improvements in institutional data management and maintenance planning.

2. Methodology

2.1. Data Sources and Station Locations

The data used in this study were river water quality monitoring records collected between 2016 and 2020. The year 2016 marked the beginning of the construction of the four stations (Manggarai, Kelapa Dua, Empang Dam, and Pasar Baru), and 2020 marked the end of the database storage on the server of the BJ Habibie Science and Technology Area (KST BJ Habibie), Serpong, South Tangerang City, Banten Province. Subsequently, from 2021 onward, the database was transferred to a server at the Ministry of Environment and Forestry (KLHK). The Kelapa Dua and Manggarai stations selected represent the upstream and downstream segments of the Ciliwung River in the DKI Jakarta area, while Empang Dam and Pasar Baru stations represent the upstream and downstream segments of the Cisadane River in the Bogor and Tangerang areas. Figure 1 illustrates a map of the monitoring stations and data center facilities, while Table 1 provides the addresses and geographic coordinates of the four station locations.

An advanced data logger automatically manages the process of storing real-time, online water quality monitoring data. This device operates using two sampling intervals: the Early Warning System (EWS) interval and the periodic interval. Water quality measurements were carried out using a WQC-type multi-probe sensor with 11 parameters; in this study, only 10 were analyzed: temperature, DO, ammonia, conductivity, pH, TDS, turbidity, salinity, nitrate, and ORP. This sensor is equipped with a handheld to display the measurement results directly. Of the two data collection intervals, only data from the periodic measurements are stored in internal memory and transmitted to a central database via SMS or internet data services. The central server

receives and stores monitoring data in a structured relational database system, which enables data retrieval through a web-based application interface designed for environmental data analysis and reporting.

In operational practice, calibration and maintenance are carried out following the technical guideline procedures from each vendor. This includes cleaning the probes (removing biofouling), calibrating standard solutions (such as pH buffer and conductivity standards), inspecting cables and connectors, checking telemetry, inspecting power subsystems, and recalibrating or resetting sensors after disturbances. Since complete records of calibration and historical maintenance are not always available in digital format for all stations and time periods, this data is not analyzed as a separate dataset. However, it very likely influence on data quality indicators is explicitly considered during result interpretation.

2.2. Integrated TDQM-ODQEM Framework

Data stored in a structured relational database system, then, raw data, including station identification, date, time, and multiparameter water quality observations, were downloaded from the database for analysis. Before conducting TDQM-ODQEM analysis, raw data is first processed through a transparent quality control workflow to ensure reproducibility. The preprocessing steps include: (i) extracting and organizing data based on station and parameter; (ii) standardizing the date-time field into a single timestamp format; (iii) sorting and identifying duplicate timestamps; (iv) detection of missing values (without imputation for completeness calculations); (v) detection of zero values using a formula check for all parameters; (vi) range-based outlier detection using established physical/sensor validity limits (Table 2); and (vii) harmonizing parameter names and units across stations.

To assess the quality of the data collected, this study utilized the Total Data Quality Management (TDQM) framework introduced by Wang (1998). TDQM is a proven methodology that offers a systematic way to manage and evaluate data quality through an ongoing improvement cycle (see Figure 2), consisting of four repeating steps: define, measure, analyze, and improve (Wang, 1998). The TDQM framework highlights that data should be regarded as a strategic asset, requiring proactive quality assurance measures throughout its lifecycle (Mohamed et al., 2009). The first step, Define, aims to identify user needs and establish relevant data quality dimensions such as accuracy, completeness, validity, and uniqueness. During this phase, quality standards and business rules are set as the basis for measurement. Next, the Measure stage is conducted to evaluate how well the available data meets the established standards using measurable metrics, such as the ratio of valid values, detection of extreme values, or comparison with reference data. After measurement, the analyze phase is used to find the root causes of poor data quality, including technical, procedural, and operational issues. This step helps identify where and why errors or discrepancies happen.

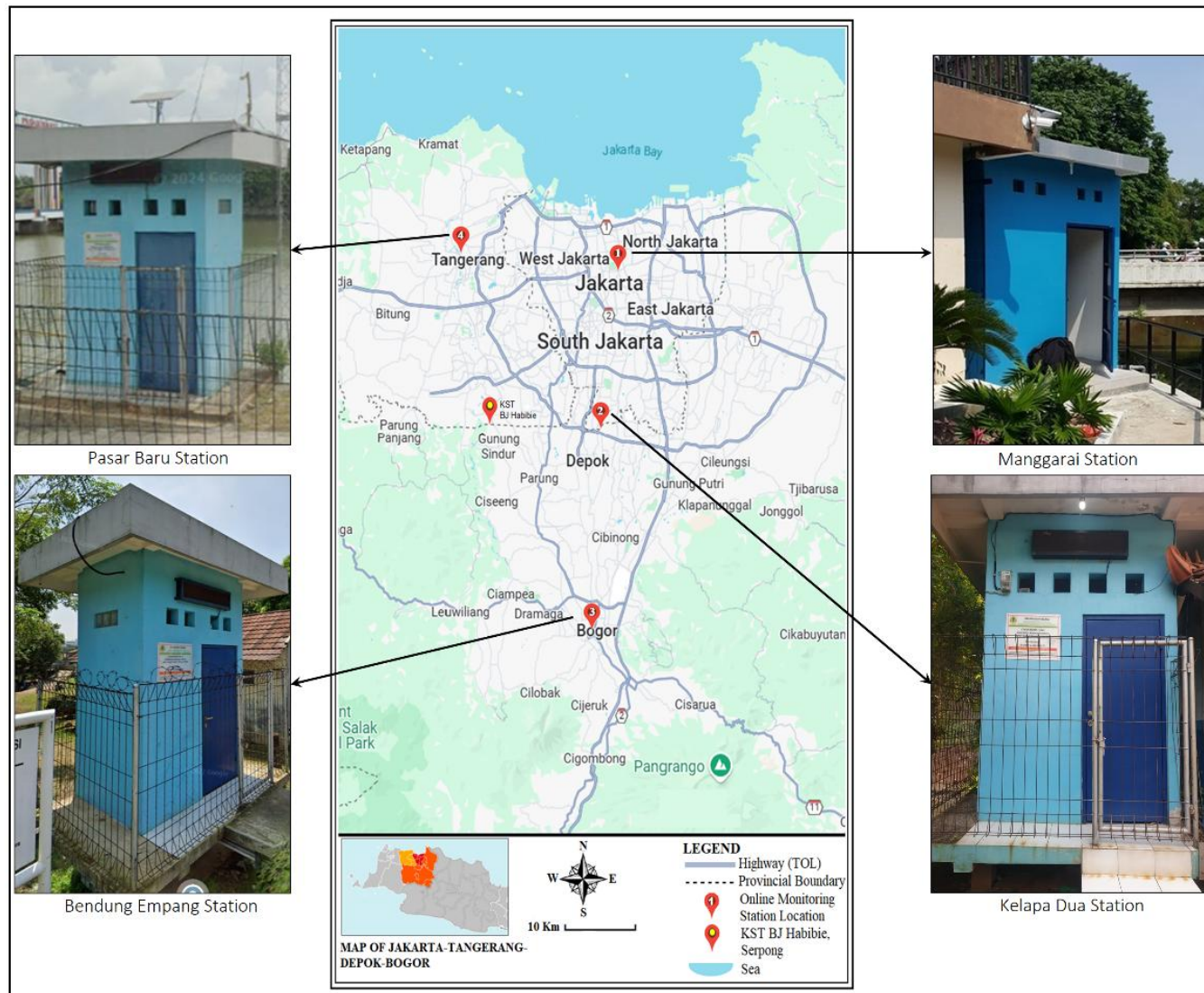


Figure 1. Location map of four monitoring stations and data centers at KST BJ Habiebie, Serpong, South Tangerang City, Banten Province, Indonesia

Table 1. Location of four online monitoring stations on the Ciliwung River and Cisadane River

No.	Station Name	River	Address	Coordinate	
				Latitude	Longitudes
1.	Manggarai Station	Ciliwung River	Jl. Tambak, RT.07/ RW.04, Pegangsaan Subdistrict, Menteng District, Central Jakarta, DKI Jakarta	-6.20784	106.84850
2.	Kelapa Dua Station	Ciliwung River	Arus Hill, Srengseng Sawah, Jagakarsa, South Jakarta, DKI Jakarta.	-6.35264	106.83564
3.	Empang Dam Station	Cisadane River	Cisadane Mountain, Paledang, Bogor City, West Java Province	-6.60777	106.79288
4.	Pasar Baru Dam Station	Cisadane River	Jl. KS Tubun, Koang Jaya, Tangerang City, Banten Province	-6.16079	106.62772

Lastly, the Improve phase involves implementing corrective actions like sensor calibration, operator training, automated validation systems, or enhanced data integration. After improvements are implemented, the process loops back to the Define stage to update data quality standards and strategies based on previous lessons learned, creating a continuous cycle of data quality management. This framework has been widely adopted in Internet of Things (IoT)-based systems, including online water quality monitoring, due to its capacity to systematically and measurably uphold data integrity and reliability.

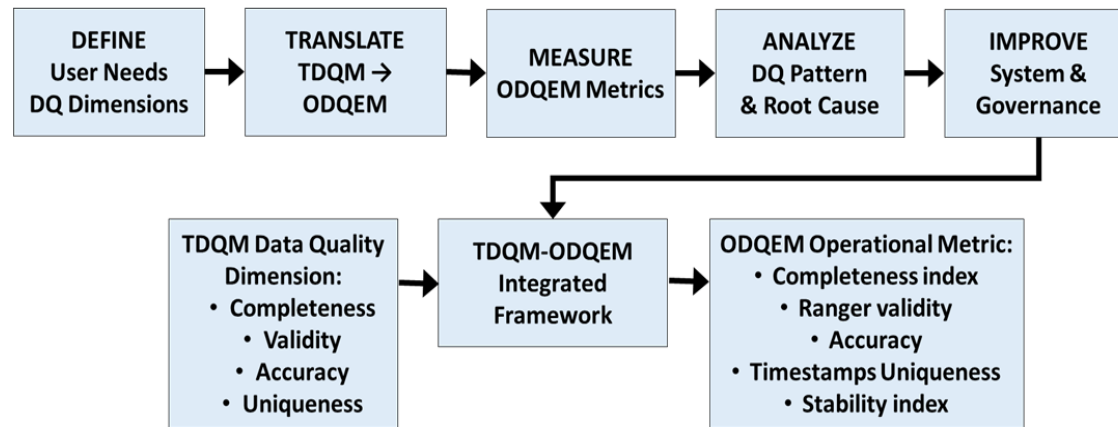


Figure 2. Four main components of the TDQM cycle
(Source: Wang, 1998; Klein and Lehner, 2009)

This diagram shows a hybrid data quality approach that combines the conceptual cycle of TDQM with the operational indicators of the Online Data Quality Evaluation Model (ODQEM). The process starts with defining user needs and the main data quality dimensions of completeness, validity, accuracy, and uniqueness (Cahyono et al., 2020), which form the basis for assessing real-time monitoring data. To evaluate the data quality collected from four stations, this study uses a combined method of Total Data Quality Management (TDQM) and the proposed Online Data Quality Evaluation Model (ODQEM). TDQM provides a conceptual framework based on common data quality dimensions, including completeness, validity, accuracy, and uniqueness (Wang & Strong, 1996; Wang, 1998). This framework is then tailored to the context of sensor-based online water quality monitoring, using ODQEM technical indicators such as the Completeness Index (CI), Range Validity (RV), Stability Index (SI), and Uniqueness (duplicate check). This integration allows data quality assessment not only from a technical standpoint (sensors) but also in line with global data quality management standards. Below is an explanation of each ODQEM indicator.

a. Completeness index (CI)

The Completeness Index (CI) measures the proportion of available data relative to the total observation period. In environmental datasets, completeness is vital because missing data can greatly affect analysis results (Batini & Scannapieco, 2016). In an online water quality monitoring system, completeness indicates whether data are recorded consistently without missing values caused by communication issues, power outages, or sensor failures. The CI formula is expressed as:

$$\text{Completeness Index} = \frac{\text{Number of recorded (non - null) values}}{\text{Number of value that should have been recorded}} \times 100\% \quad (1)$$

A high CI (>95%) indicates a well-functioning system, while a moderate CI (70–90%) indicates some incomplete data, possibly due to sensor failure or extreme river conditions. Completeness assessment is one of the first steps in data quality (Behmel et al., 2016; Li et al., 2018).

b. Range Validity (RV)

The range validity (RV) indicator evaluates whether sensor data is within acceptable limits, considering both the physical range and water quality standards (Table 2). This is crucial for filtering out unreasonable or out-of-specification readings that may result from sensor errors or data transmission issues (Teh et al., 2020). Sensor errors, drift, or extreme hydrological conditions often cause values to exceed these limits. The RV formula can be expressed as:

$$\text{Range Validity} = \frac{\text{Number of data within standard range}}{\text{Number of data}} \times 100\% \quad (2)$$

In this study, the Validity Range (VR) interpretation approach is operationally classified into three levels: high (≥90%), moderate (70–<90%), and low (<70%). These thresholds are used as research-based evaluation criteria to indicate the proportion of observations within a reasonable range. This interpretation aligns with quality control principles in sustainable water quality monitoring, where unreasonable values are typically addressed through rough range checks and further investigated for possible sensor damage, deviations, or calibration issues (Bushnell & Worthington; 2016 U.S. EPA, 2018).

Range validity is widely used to assess the quality of environmental sensor data (Ramirez et al., 2011). It is a basic and standard quality control (QC) technique that confirms sensor readings stay within specified upper and lower bounds.

c. Stability Index (SI)

The Stability Index quantifies the proportion of data that remains unchanged over time, specifically values that do not vary over ≥3 consecutive measurements (Qartod, 2018). Flatlining can occur if the sensor is covered with sediment (fouling), loses calibration, or sustains technical damage (Bushnell et al., 2015). This indicator has been used to evaluate the quality of environmental sensor data (Zhang et al., 2022). Measuring the percentage of flatline data:

$$SI = \frac{\text{Number of flatline segments}}{\text{Total segments}} \times 100\% \quad (3)$$

In this study, the Stability Index (SI) is operationally defined as follows: high (>90%), moderate (70–90%), low (50–70%), and very low (<50%). These limits are used as research-based assessment criteria, not as universal standards. This interpretation aligns with the principles of continuous water quality QA/QC, where decreasing signal stability may indicate increasing anomalies and requires further verification through field inspection, comparison with discrete/manual samples, sensor maintenance, fouling checks, deviation correction, and recalibration (Qartod, 2018).

d. Accuracy

Accuracy was assessed as the percentage of sensor readings within a predetermined acceptable range based on standards or regulations (e.g., river water quality standards), and was calculated while the formula (Redman, 2005) used is as follows:

$$\text{Accuracy} = \frac{\text{(Number of valid (in range))}}{\text{Total values}} \times 100\% \quad (4)$$

Data accuracy was assessed by comparing sensor reading them with laboratory data (when available) or masked tests using imputation techniques. The metric

used was the Mean Absolute Error (MAE) (Zhang & Thorburn, 2022). MAE measures the average absolute difference between sensor values and laboratory values, regardless of whether the difference is positive or negative.

$$MAE = \frac{1}{n} \sum |Y_{sensor} - Y_{lab}| \tag{5}$$

Where

n = number of data or observations

Y_{sensor} = actual value (sensor value)

Y_{lab} = predicted value or measurement result (lab value)

| ... | = absolute value

e. Uniqueness

Uniqueness indicators are used to ensure data uniqueness, thereby maintaining the integrity of time series analysis and forecasting models (Qin et al., 2016). In online river water quality monitoring systems, uniqueness primarily concerns timestamps since each measurement must be unique. If two datasets share the same timestamp, duplication occurs, which reduces data quality. The uniqueness index is calculated by dividing the number of unique data points by the total number of data points (Ehrlinger & Wöb, 2022).

$$UQ = \frac{\text{Number of unique values}}{\text{Total values}} \times 100\% \tag{6}$$

The dataset evaluated includes 15 key attributes listed in Table 3, which make up the main measurement records from four stations, totaling 66,505 entries. Each record has 15 columns, including the station ID, date, time, and 12 water quality parameters.

Table 2. Acceptable range for each water quality parameter

No.	Parameter	Range
1	BOD	0.1 - 60 mg/L
2	COD	0.1 - 500 mg/L
3	Temperature	0° C - 50° C
4	DO (Dissolved Oxygen)	0 - 15 mg/L; or 0 - 200%
5	pH	0 - 14 units
6	Nitrate	0 - 50 mg/L
7	Nitrite	0 – 10 mg/L*
8	TSS	0 - 500 mg/L
9	TDS / conductivity / salinity	0 - 100,000 μS/cm or 0 - 100 mS/cm
10	Turbidity	0 - 1000 NTU
11	Ammonia	0 - 100 mg/L as N / 0 – 10 mg/L **
12	Depth (pressure)	0 - 10 m or more

(Source: Regulation of the Minister of Environment and Forestry No. 2 of 2022;

*Regulation of the Minister of Health of the Republic of Indonesia No. 492 of 2010;

** Regulation of the Minister of Environment and Forestry No. 68 of 2016)

Table 3. Description of river water quality monitoring data attributes

No.	Attribute Name	Data Type	Description
1	IDStation	Char(10)	Monitoring station identifier
2	Date	Date	Date of measurement
3	Time	Time	Time of measurement
4	Temperature	Double (5.2)	Water temperature (°C)
5	EC	Double (5.2)	Electrical Conductivity (µS/cm)
6	TDS	Double (5.2)	Total Dissolved Solids (mg/L)
7	Salinity	Double (5.2)	Water salinity (ppt)
8	DO	Double (5.2)	Dissolved Oxygen (mg/L)
9	pH	Double (5.2)	Acidity/alkalinity level
10	Turbidity	Double (5.2)	Water turbidity (NTU)
11	Depth	Double (5.2)	Water depth at the monitoring point (m)
12	SwSG	Double (5.2)	Seawater Specific Gravity
13	Ammonia	Double (5.2)	Ammonia concentration (mg/L)
14	Nitrate	Double (5.2)	Nitrate concentration (mg/L)
15	ORP	Double (5.2)	Oxidation-Reduction Potential (mV)

(Source: BPPT, 2015)

3. Results and Discussion

3.1. Validity Range Assessment

The results show that most physical parameters, including temperature, conductivity, TDS, salinity, and turbidity, remained within the valid range across all monitoring stations (90%-100%). This pattern suggests that the sensors operated within their specified limits and did not produce values outside the expected environmental ranges. This performance supports the ODQEM assumption about parameter robustness. It also aligns with the observations of Karkouch et al. (2016), who emphasized the inherent stability of physical IoT water quality sensors compared to biochemical probes.

In contrast, nutrient parameters exhibited significant variability. Nitrate validity was very low at Kelapa Dua (61.67%) and moderately low at Manggarai (84.30%) and Pasar Baru (82.56%), indicating possible ion-selective electrode drift, environmental interference, or intermittent fouling. Low nitrate validity is often caused by long-term sensor drift, measurement surface fouling, and ionic cross-interference, especially in nutrient sensors used in dynamic water environments (Snyder et al., 2018; Daniel et al., 2020).

Dissolved oxygen (DO) validity at the Empang Dam also decreased (90.40%), suggesting membrane fouling, disrupted optical paths, or unstable power supply (Samuelsson et al., 2018). These differences underscore the importance of the TDQM definition and analysis phase in ensuring that acceptable ranges are based on realistic environmental conditions and maintained through regular recalibration. Figure 3 displays the average validation ranges from the four stations.

3.2. Accuracy Evaluation

Accuracy values for physical parameters remained consistently high across all stations, approaching 100%. This confirms that raw measurements are generally consistent with reference values or expected ranges, and that the calibration procedures used at the installation remain effective. These findings align with U.S. EPA (2018) guidelines, which indicate that physical sensors typically exhibit low intrinsic error when supported by reliable power and continuous communication links. Inaccuracies can occur due to sensor calibration deviations, contamination, or configuration errors (Wei et al., 2019).

Chemical parameters showed notably more variability. The accuracy of nitrate in Kelapa Dua (61.67%) and ammonia in Manggarai (74.67%) may be due to measurement noise, signal drift, or occasional sensor interference (Luna et al., 2020). ORP accuracy was the lowest, especially in Manggarai (27.26%), suggesting possible biofilm buildup on the probe surface, which is known to affect oxidation-reduction potentials (Saboe, 2022). Within the TDQM framework, these deviations suggest that the Measure and analyze phases should include more frequent assessment cycles for high-risk parameters, especially those related to nutrients and the redox sensor (ORP). Figure 4 displays the average accuracy across the four stations.

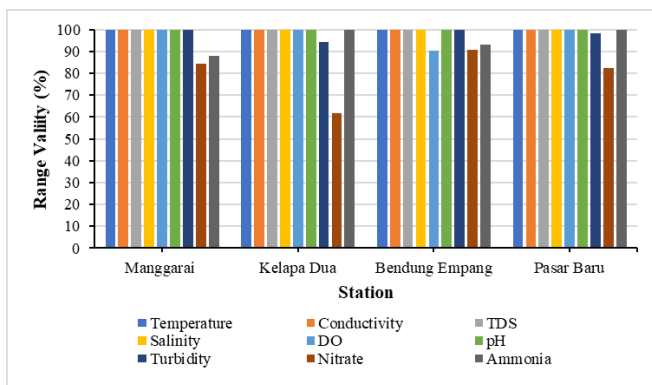


Figure 3. Validation range at four stations

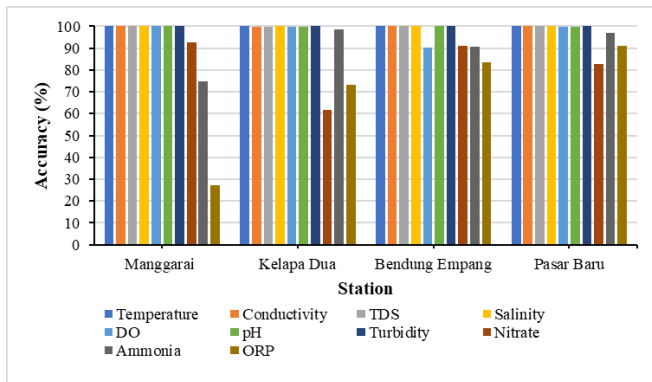


Figure 4. Average dimension accuracy at the four stations

Since manual data were unavailable for the 2016-2020 period, monitoring data from the Manggarai and Kelapa Dua stations for 2023-2024 were used and compared with manual measurement data. The manual data were obtained from the Ministry of Environment and Forestry's report, which included six sampling data points taken in December 2023, June 2024, July 2024, and August 2024. The following are the results of the analysis using the Mean Absolute Error (MAE) statistical approach for the Manggarai and Kelapa Dua stations. As shown in Tables 4 and 5.

MAE analysis at Manggarai Station revealed that the DO and ammonia parameters showed large and unstable deviations, along with extremely negative MAE, indicating sensor drift, power failure, or masking failure under poor

conditions. This matches the findings of Lucas et al. (2025), who explained that chemical sensors in online monitoring systems are highly prone to fouling, membrane degradation, and fluctuations in river current, which can cause significant errors. In contrast, pH, temperature, and TDS exhibited high and consistent MAE (>88%), indicating better measurement stability, which agrees with the literature stating that physical sensors are more stable than electrochemical sensors (Karkouch et al., 2016). The TDS parameter experienced moderate fluctuations due to variability in sediment loads, which is common in urban river flows.

At Kelapa Dua Station, MAE instability was more noticeable, with almost all DO and ammonia parameters showing large negative MAEs. These could indicate periods of complete measurement failure or system shutdown, consistent with what is observed when dissolved oxygen and ammonium sensors completely fail or produce constant errors (Tena et al., 2020; Yang et al., 2021). Although pH and temperature remained relatively stable, a single instance of negative MAE values at both stations suggests a systemic failure affecting the entire sensor array. Overall, the MAE patterns at these two stations demonstrate that chemical sensor accuracy heavily relies on environmental stability, routine maintenance, and power conditions, as highlighted by Mahmud et al. (2020).

Table 4. MAE Analysis at Manggarai Station

Sample data date	Test	DO	pH	Ammonia	Temperature	TDS
28-Dec-23	MAE (%)	-17.31	97.56	-76.72	99.86	71.86
3-Jun-24	MAE (%)	-20.69	98.40	-61.81	91.97	46.96
June 13, 2024	MAE (%)	-65.81	89.18	94.29	94.89	91.46
30-Jul-24	MAE (%)	-75.40	97.90	-94.03	95.93	99.21
5-Aug-24	MAE (%)	68.61	98.58	-91.30	92.38	44.92
15-Aug-24	MAE (%)	32.84	98.87	-87.10	88.36	85.93

Table 5. MAE Analysis at Kelapa Dua Station

Sample Data Date	Test	DO	pH	Ammonia	Temperature	TDS
December 31, 2023	MAE (%)	-51.64	92.96	74.35	98.84	66.21
June 3, 2024	MAE (%)	-93.57	97.02	-93.06	92.83	95.69
June 11, 2024	MAE (%)	-92.58	90.00	17.65	98.58	60.52
5-Aug-2024	MAE (%)	-100.00	-100.00	-100.00	-100.00	-100.00
August 15, 2024	MAE (%)	-90.96	90.01	-91.90	82.49	46.23

3.3. Completeness and Zero-Value Analysis

The Completeness Index values, as shown in Figure 5, across all stations and parameters stayed at 100%, indicating no missing timestamps and dependable data transmission. From a TDQM perspective, this indicates effective data collection, communication, and storage,

especially during the Measurement phase. However, the zero-completeness analysis tells a different story when evaluating the validity of the collected data. While all timestamps were recorded, some parameters exhibited a high rate of unreasonable zeros, which ODQEM classifies as a systematic anomaly.

The Empang Dam in Table 6 showed complete data with extremely low values for DO (1.02%) and nitrate (0.19%), while Pasar Baru experienced a sharp decline in TDS (9.57%), despite having perfect timestamp completeness. Such zero or constant readings are common indicators of sensor failure, including constant output mode or total failure, which is often linked to a dead sensor or malfunctioning probe (Yang et al., 2021). Kelapa Dua exhibited significant missing data for ammonia, with only 60.02% of values being non-zero, further highlighting the vulnerability of the ion sensors.

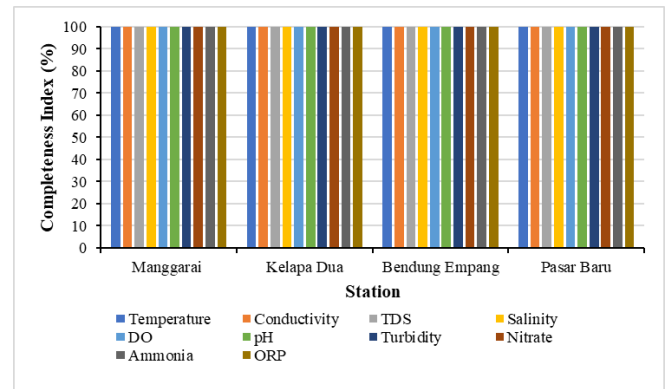


Figure 5. Completeness index at the four stations.

Table 6. Completeness with non-zero values at the four stations

Station	Temp.	EC	TDS	Salinity	DO	pH	Turbidity	Nitrate	NH ₃	ORP
Manggarai	99.30	99.27	75.22	47.86	83.46	99.31	96.77	76.38	85.14	98.95
Kelapa Dua	99.14	98.83	89.27	42.82	83.50	99.16	79.31	92.34	60.02	99.14
Empang Dam	95.92	99.76	98.36	30.93	1.02	97.8	99.74	0.19	46.15	39.38
Pasar Baru	99.06	50.66	9.57	90.58	99.23	59.1	63.15	53.70	98.23	90.4
Average	98.35	87.13	68.11	53.05	66.8	88.84	84.75	55.65	72.38	81.97

The results show that although the time-stamp-based Completeness Index reached 100% at all stations, the proportion of non-zero values indicates that some parameters are only nominally complete and not analytically informative. The average non-zero completeness was very low for salinity (53.05%), nitrate (55.65%), DO (66.80%), and TDS (68.11%), with the most severe degradation observed at Dam Empang for DO (1.02%) and nitrate (0.19%) and at Pasar Baru for TDS (9.57%). These results imply that a dataset may appear structurally complete but still contain prolonged periods of inactivity and repeated zero values, which weaken the interpretation of river water dynamics. Therefore, non-zero completeness should be treated not merely as a supplementary descriptive metric, but as a key operational indicator for assessing whether the transmitted data remains meaningful for environmental analysis and decision-making support.

These findings demonstrate that completeness alone does not fully capture true data quality because incomplete data sets can significantly weaken the integrity of analytical results and the reliability of decision-making (Batini et al., 2009). Therefore, ensuring a high level of completeness is critical in supporting robust and actionable environmental intelligence (Pipino et al., 2002). TDQM analysis requires a more in-depth investigation beyond surface-level completeness, while the zero-valued ODQEM metric offers an important measure for identifying subtle sensor errors. This multi-layered assessment highlights that nominal completeness does not guarantee the completeness of information, emphasizing the need for a TDQM

Improvement phase to address sensor maintenance, recalibration schedules, and system-level corrections.

3.4. Timestamp Uniqueness and Temporal Integrity

The uniqueness score exceeded 99.9% across all stations, as seen in Figure 6, indicating that the system does not produce duplicate timestamps and that the data stream maintains its temporal integrity. This demonstrates effective interaction between the sensor's Real-Time Clock (RTC), the gateway, and the server synchronization protocol. The high level of uniqueness meets the ODQEM requirements for structural data reliability and aligns with the ISO/IEC 30141 standard (IEC, 2024), which ensures that various IoT devices, systems, and platforms can connect, communicate, and exchange data.

In TDQM, temporal uniqueness relates to the Definition phase (where rules for timestamp integrity are established) and the Measurement phase (which tracks timestamp anomalies as a quality indicator). The observed stability demonstrates effective time synchronization governance and a low risk of redundant or duplicated data that could distort analysis results. Therefore, implementing systematic deduplication strategies, such as constraint-based validation, timestamp hashing, and version control, is crucial for maintaining high-quality data streams (Kaur et al., 2018).

3.5. Stability Index

At all four stations, as shown in Table 7, the Stability Index values show that Manggarai has strong stability in physical parameters, including temperature (89.81%), conductivity (86.53%), pH (91.69%), turbidity (91.24%),

and ORP (92.44%). However, nutrient and ion parameters display different patterns. The stability of nitrate (65.97%) and ammonia (71.40%) indicates moderate temporal consistency, while TDS and salinity are very unstable (5.55% and 5.56%). The instability of these ion parameters may be caused by variable sedimentation, sudden changes in dissolved solids from rainwater runoff, or sensor problems such as scaling or membrane clogging.

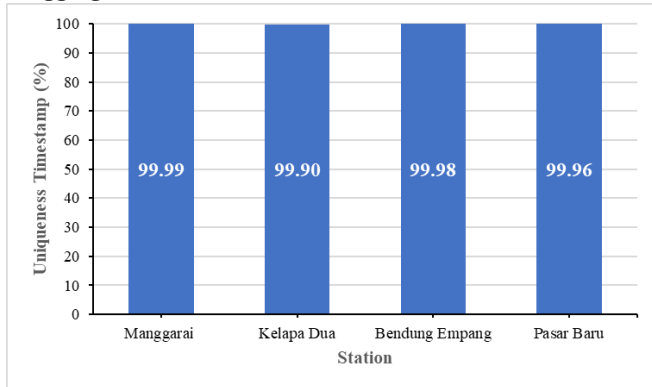


Figure 6. Average timestamp uniqueness across the four stations.

Kelapa Dua showed a strong stability pattern in physical parameters, especially conductivity (88.80%), DO (80.99%), pH (92.74%), turbidity (78.78%), and ORP (90.26%). Nitrate stability was relatively high (89.43%), but ammonia was volatile at 35.21%, possibly due to equipment malfunction or significant temporal variability in ammonia levels at this site. This aligns with urban runoff patterns, where ammonia spikes caused by domestic wastewater input can occur irregularly.

The Empang Dam showed the highest physical stability among the stations, with temperature at 95.23%, pH at 97.21%, turbidity at 93.09%, and ORP at 90.59%. DO stability was also quite high at 94.34%. However, nutrient stability remained low, with nitrate at 49.91% and ammonia at 41.78%. This pattern of high physical stability but low chemical stability matches previous findings in the Completeness and Validity of Range section, where the Empang Dam posed significant challenges for

nutrient sensors. The high sediment load and periodic turbulence in this river segment probably worsen these challenges.

Pasar Baru showed the lowest overall stability, especially for conductivity (76.32%), turbidity (47.05%), nitrate (80.47%), and ammonia (30.20%). TDS (5.49%) and salinity (1.47%) also had very low stability, indicating significant environmental variability and possible sensor degradation. Despite these issues, the DO stability of 84.42% and pH stability of 89.01% suggest some core sensors remain operational.

The Stability Index results clearly show differences in sensor performance across stations, and provide a clearer basis for empirical prioritization across parameters and indicators. These results indicate that the most critical weaknesses were found in salinity (4.30%) and TDS (7.89%), followed by ammonia (44.65%), while pH (92.66%), ORP (90.06%), temperature (88.58%), and conductivity (85.02%) remained relatively stable across all stations. This emphasizes that physical parameters generally stay reliable. In contrast, chemical parameters tend to be unstable over time. This aligns with global patterns seen in WQMS systems, where ion sensors like nitrate and ammonia consistently perform no better than physical sensors (Rinn et al., 2025). During the TDQM cycle, such variability requires targeted improvements, especially recalibration, anti-fouling measures, and redundancy for critical parameters. Physical parameters generally remain more stable across stations, while chemical and ionic parameters are more susceptible to drift, fouling, scale formation, and flat-line behavior. Unstable variables dominated by zero values require more stringent recalibration intervals and targeted cleaning and anti-fouling measures. Meanwhile, variables with high validity and strong temporal stability can remain under routine quality control procedures. This analysis provides a more robust empirical foundation for the discussion by going beyond generalizations about sensor instability. The findings specifically identify parameters experiencing consistent failures and their impact on data reliability across multiple indicators.

Table 7. Average stability index at the four stations

Station	Temp.	EC	TDS	Salinity	DO	pH	Turbidity	Nitrate	NH ₃	ORP
Manggarai	89.81	86.53	5.55	5.56	59.36	91.69	91.24	65.97	71.40	92.44
Kelapa Dua	88.38	88.80	9.61	6.62	80.99	92.74	78.78	89.43	35.21	90.26
Empang Dam	95.23	88.43	10.92	3.55	94.34	97.21	93.09	49.91	41.78	90.59
Pasar Baru	80.91	76.32	5.49	1.47	84.42	89.01	47.05	80.47	30.2	86.94
AVG	88.58	85.02	7.89	4.3	79.78	92.66	77.54	71.44	44.65	90.06

3.6. Integrated Interpretation of TDQM-ODQEM Results

The hybrid evaluation showed that while structural data quality indicators such as uniqueness and validity were generally strong, operational indicators, particularly

stability and zero-value analysis, revealed chronic weaknesses in chemical sensor performance. Average non-zero completeness values were low for salinity (53.05%), nitrate (55.65%), DO (66.80%), and TDS (68.11%), while stability was very poor for salinity (4.30%),

TDS (7.89%), and ammonia (44.65%). In other words, a dataset may appear good because its records are complete and well-organized, but it is generally unreliable for practical use if many readings are unstable or repeatedly recorded as zero. This finding demonstrates why data quality should be assessed not only by whether the data are present and well-structured, but also by whether the measurements are consistent and can be used to interpret actual river conditions and support timely decision-making. A further implication is that indicator integration should be interpreted hierarchically rather than descriptively. In this dataset, uniqueness is a fundamental structural requirement that has largely been met, while completeness and non-zero stability serve as decisive filtering criteria for operational utility. Range validity and accuracy remain important, but both should only be interpreted after ensuring that the time series is not dominated by persistent zeros, flat lines, or repetitive data. Practically, a parameter should be classified as high risk when it performs poorly in at least two operational dimensions, particularly completeness and non-zero stability. This explains why salinity, TDS, and ammonia require more urgent corrective action than temperature, pH, or ORP. Therefore, completeness and non-zero stability are treated as primary operational indicators, as they more directly determine whether the time series still contains meaningful environmental information over time. Validity and accuracy are then interpreted as supporting indicators, but only after the data has undergone operational filtering. Within this hierarchy, parameters exhibiting poor performance across more than one operational dimension are classified as higher-priority targets for corrective action, while parameters with strong structural quality and stable temporal behavior are considered more reliable for routine interpretation. This approach does not assign formal mathematical weights but rather provides a clear, practical prioritization framework for assessing the usefulness of data.

The integrated TDQM–ODQEM model suggests that improving ORWQM data quality requires not only sensor-level interventions but also institutional-level improvements in calibration governance, maintenance standardization, and interagency coordination. In Indonesia, water pollution control efforts are carried out through prevention, mitigation, and restoration of water quality, as stipulated in Government Regulation (GP) of the Republic of Indonesia No. 22 of 2021 concerning the implementation of environmental protection and management. GR Number 82 of 2001 concerning Water Quality Management and Water Pollution Control. This GR specifically regulates water quality management and water pollution control, including river water quality monitoring. Although it does not explicitly mention online river water quality monitoring, this regulation serves as the basis for developing monitoring systems, including technology-based systems that monitor water quality parameters in real time. Further institutional

responsibilities are regulated by the Ministry of Environment and Forestry's organizational regulations, including Regulation of the Minister of Environment and Forestry No. 15 of 2021 concerning organizational structure and functions. These legal instruments imply that data quality assurance, validation procedures, and monitoring follow-up should not be treated as ad hoc technical tasks but as formal institutional responsibilities related to environmental monitoring, reporting, and control functions. Therefore, the governance recommendations proposed in this study, such as calibration standardization, maintenance scheduling, and cross-agency quality control, are based not only on observed sensor anomalies but also on the existing regulatory architecture of the Indonesian environmental monitoring system. Furthermore, the identified instability and frequent zero values should not only be interpreted as limitations requiring further research, but also as a basis for practical handling of existing datasets. A practical workflow is to first flag prolonged zero values and flatlines, second apply robust filtering to isolate short anomalous spikes, third incorporate unstable chemical parameters into daily or weekly summaries, and fourth retain only validated segments for interpretation against regulatory thresholds. This is particularly relevant for stations such as Empang Dam and Pasar Baru, where certain parameters exhibit significant degradation while others remain operationally informative. Thus, this study not only identifies quality issues but also highlights the potential for further analysis. Therefore, future research should explore multidimensional strategies for improving data quality, encompassing not only technological improvements but also institutional coordination, stakeholder engagement, socioeconomic factors, and environmental dynamics (Zhang & Thorburn, 2022).

4. Conclusion

This study approach uses an integrated framework of Total Data Quality Management (TDQM) and the Online Data Quality Evaluation Model (ODQEM) to identify priority issues for improvement to enhance the reliability of real-time monitoring data. The aims of this study were to evaluate the quality of data generated by four online river water quality monitoring (ORWQM) stations located in the downstream and upstream areas of the Ciliwung and Cisadane Rivers. Results indicate that the ORWQM system performs strongly on the structural data quality dimension, with validity, accuracy, and uniqueness generally ranging from 99% to 100%, and timestamp uniqueness consistently exceeding 99.9%. However, the evaluation also revealed persistent operational weaknesses. Completeness levels based on non-zero values varied significantly across parameters, with average scores of 68.11% for TDS, 53.05% for salinity, 66.80% for DO, and 55.65% for nitrate. The Stability Index indicates that physical parameters remain relatively stable, including temperature (88.58%), EC (85.02%), pH (92.66%), and ORP (90.06%), while chemical and ionic

parameters are much less stable, particularly TDS (7.89%), salinity (4.30%), and ammonia (44.65%). These findings suggest that nominally complete and structurally valid data streams remain operationally unreliable for decision-making when impacted by flat data, anomalous zero values, or unstable chemical sensor behavior.

The novelty of this study lies in integrating TDQM with ODQEM, a strategic data governance framework with a real-time operational diagnostic framework, to evaluate data quality in Indonesia's ORWQM system. This integration is a substantive contribution because it goes beyond conventional assessments that focus solely on missing data or basic sensor outputs, and instead provides a multidimensional interpretation that links data structure, sensor behavior, and governance implications. Practically, this study provides a replicable evaluation model to identify which parameters can be managed institutionally and which require targeted technical interventions, particularly for ion sensors. More broadly, these findings suggest that the success of an ORWQM program should not be judged solely by the presence of real-time infrastructure or high-volume data streams, but rather by whether the resulting data are reliable enough to support pollution control, regulatory enforcement, and timely environmental responses. In this regard, this study has broader relevance for watershed management in Indonesia and other developing country contexts, where digital monitoring systems are evolving faster than the institutional capacity needed to ensure data quality.

At the same time, this study has several limitations: it only analyzed four stations, relied primarily on historical records from 2016–2020, and had limited laboratory reference data for direct accuracy validation, which was only available for specific parameters and periods at two stations in Manggarai and Kelapa Dua in 2023–2024. Therefore, future research should pursue three priority areas. First, we tested this framework across a range of stations, river types, and seasonal conditions to evaluate its robustness and applicability. Second, we integrated more systematic laboratory validation and maintenance logs to better distinguish between sensor drift, fouling, and true environmental variation. Third, we extended this framework to predictive and decision-support applications, including an anomaly alert system and a management dashboard that translates data quality diagnostics into operational actions. Overall, this study addresses a key analytical gap by demonstrating that the reliability of ORWQM depends not only on sensor placement but also on integrated data quality governance that supports transparent, evidence-based river pollution control. Without robust data quality governance, real-time monitoring remains merely an infrastructure promise, and with such governance, ORWQM can serve as a credible foundation for adaptive and accountable river management.

5. Data Availability Statement

The data used in this study are primary data (2016–2020), consisting of online water quality monitoring of the Ciliwung and Cisadane Rivers at four stations. The data is stored in the BPPT's Onlimo data center database. The data are available upon request and subject to approval by the relevant agencies.

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7. Conflict of Interest

Each author has declared that there is no conflict of interest in the writing or submission of this manuscript.

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9. Author Contribution

SY conducted investigations, formal analysis, literature review, and manuscript preparation. **DNM**, **DS**, and **BK** were involved in the conceptualization and review of the manuscript. All authors read and approved the final manuscript.

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Quantitative Prediction of *Spirulina platensis* Biomass Using UV-Vis Spectrophotometry

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Abstract

Spirulina platensis has become a promising feedstock for the synthesis of several industrially important biomolecules, including proteins, lipids, and carotenoids. However, significant technological obstacles pertaining to optimization, growth monitoring, and biomolecule extraction in *Spirulina* remain despite advancements in industrial-scale microalgae production and biomolecule harvesting. Standard techniques used for microalgal biomass and biomolecules monitoring include FTIR spectroscopy, colorimetric techniques, and manual cell counting. However, these techniques have drawbacks, particularly processing time and handling errors. This paper seeks to establish an operational equation that effectively relates measured absorbance (or optical density, OD) to the dry weight of *Spirulina platensis* microalgae using a UV-Vis spectrophotometer. The wavelengths of 680, 750, and 565 nm were selected based on the absorption spectrum of chlorophyll-a, as well as the wavelengths at which absorbance does not reach its peak. The best results were obtained at a wavelength of 680 nm with the equation $y = 1.2759x + 0.1512$, with an R^2 value of 0.9914. This technique allows for more accurate measurement of *Spirulina platensis* dry weight and total biomass.

1. Introduction

Indonesia is a biodiversity hotspot of considerable magnitude, harboring an extensive array of microalgae species. According to Guiry and Guiry (2026), the country of Indonesia is home to 2,060 species of algae, encompassing both microalgae and macroalgae. These species exhibit high adaptability and flourish across a wide range of ecosystems, including freshwater and seawater environments. Microalgae, which span the spectrum of eukaryotic and prokaryotic organisms and measure between 2 and 50 mm, exhibit remarkable capabilities in photosynthesis (Baweja & Sahoo, 2015; Elisabeth et al., 2021). Microalgae play a critical role in Indonesian inland water ecosystems (rivers, lakes, wetlands) by serving as the foundation of the food chain as primary producers, and oxygenating water through photosynthesis. With a species count exceeding 2,060, they function as vital bioindicators, facilitating the monitoring of water quality, heavy metal bioremediation, and nutrient cycling in diverse habitats (Goher et al.,

2016; Rinanti et al., 2022; Mahlangu et al., 2024). Apart from functioning as the primary producer in aquatic ecosystems, the microalga's substantial biomass represents a valuable reservoir of carbon compounds. These compounds have significant applications in the production of biofuels, health supplements, and beauty products. In biotechnological and nutraceutical applications, several common microalgal species are employed. These include *Spirulina platensis/maxima* (*Arthrospira*), *Chlorella vulgaris*, *Dunaliella salina*, and *Haematococcus pluvialis* (Elisabeth et al., 2021; Tiwari et al., 2026).

Spirulina platensis is a microscopic, filamentous cyanobacterium rich in protein. Promoted as a "superfood," it contributes to high energy and contains special pigments called phycocyanin that can be used as dyes. These microalgae can be cultivated in various ways, either outdoors in open tanks or indoors in photobioreactors (Soni et al., 2017). The careful management of microalgal cultures is paramount within



the aquaculture sector to maintain a reliable and high-quality food source. Timely adjustments to process conditions, including light intensity, pH, temperature, and nutrient levels are crucial for preventing costly downtime and production failures (Nyakundi & Cleophas, 2021). Therefore, accurately measuring *Spirulina*'s biomass is critical for ecological, commercial, and research purposes (Schagerl et al., 2022). Similar results were highlighted by the existing literature on microalgal biomass and/or estimation (Podevin et al., 2017; Liu et al., 2019).

Researchers have applied several techniques in this regard; however, each has its own sets of merits and demerits as outlined below:

- Cell counting by hemocytometry is a process that necessitates the expertise of a professional scientist. This method involves the manual enumeration of cells within a hemocytometer under the guidance of a microscope. Although widely utilized, hemocytometer cell counting is restricted to small-scale operations due to its labor-intensive and time-consuming methodology (Malletzidou et al., 2024). Additionally, counting personnel's divergent perspectives may introduce inconsistencies in measurement (Heriberto et al., 2018). This phenomenon is particularly evident in cultures exhibiting cellular clumps (Caprio, 2020).
- To measure cellular components such as proteins, lipids, carbohydrates, chlorophyll, and carotenoids, researchers typically employ established biochemical assays. These standard methods include Soxhlet extraction for lipids, the phenol-sulfuric method for total carbohydrates, and Kjeldahl digestion or colorimetric tests (like Lowry and Bradford assays) for proteins (Richmond & Hu, 2013). A major drawback of these protocols is that they require laborious sample preparation and result in the destruction of the analyte.
- FTIR spectroscopy provides an avenue to chemically characterize microalgae like *Chromochloris zofingiensis*, *Acutodesmus obliquus*, *Chlorella sorokiniana* (Bartošová & Blinová, 2015), and *Scenedesmus sp.* (Sudhakar & Premalatha, 2015). Conventionally, this method spatially records the distribution of macro-constituents through characteristic wavenumber peaks. However, subsequent investigations have attempted to move beyond qualitative detection by attempting to quantitatively measure the biomolecules present.

Attaining commercial viability necessitates the development of a streamlined and effective framework. Such a system must prioritize user accessibility, operational speed, stability, and selectivity while minimizing the frequency of required calibrations. The goal is to monitor cell growth in a simple, fast, stable, and selective manner to achieve commercial viability.

Evaluating optical density (OD) is a crucial method for successfully managing microalgae production, allowing

operators to monitor population density and culture health in real time (Nielsen & Hansen, 2019; Nagabhushan, 2023). Measuring OD is useful for determining biomass concentration (Salgueiro et al., 2018; Dziosa & Makowska, 2016). Researchers frequently use spectrophotometers to estimate microalgal growth through OD tracking. This non-invasive, indirect method calculates biomass by directly linking OD values to cell density. Because the process easily integrates into automated systems, it is a highly practical tool for monitoring and controlling microalgae cultures. This study examined the ability to estimate *Spirulina platensis* biomass versus optical density using a UV-Vis spectrophotometer at various wavelengths, including 565, 680, and 750 nm. Researchers rely on specific light wavelengths to evaluate *Spirulina platensis* cultures. Specifically, biomass is often quantified using 565 nm wavelength (Santos-Ballardo et al., 2015). To determine the amount of *Spirulina* present, the 680 nm wavelength measures chlorophyll-a absorption, as the two are directly proportional (Hotos et al., 2020). Finally, because pigments do not absorb light at 750 nm, this wavelength is used to assess apparent turbidity through light scattering (Eddiwan et al., 2023).

2. Materials and Methods

2.1. Materials

Fresh, active *Spirulina platensis* culture was acquired from the membrane laboratory collection at BRIN-Limnology and Water Resources Research Center. The strain was axenically maintained under controlled laboratory conditions by inoculating 500 mL of a commercially purchased cyanobacterial starter culture into a 5 L transparent PTE plastic bottle, inoculated to 4 L of modified Zarrouk medium (Chrismadha et al., 2022). *Arthrospira platensis* was cultivated at $27.0 \pm 0.5^\circ\text{C}$ temperature and pH 9.4 under constant aeration of ~ 3 L/min. The culture was illuminated continuously with an 18W full-spectrum LED array (Philips 18W, Holland) providing 2,000–3,000 lux and a photoperiod of 24 h light: 0 h dark. Following three weeks of growth, the wet biomass was harvested from the clear PTE container. *S. platensis* culture was observed microscopically using a 40x objective lens (Olympus CKX53 Inverted Microscope). Observations were made on the first and last days of the experiment.

2.2. Methods

The wet culture was air-dried until a dry culture with a constant weight was obtained. Because standard reference materials were unsuitable for creating a calibration curve between *S. platensis* biomass and spectral response, we used a modified serial dilution technique based on standard addition (Malletzidou et al., 2024). The dried *S. platensis* culture was weighed at 0.01 grams to 0.4 grams, as described in Table 1, and was employed as the stock solution to create ten dilution levels that served as the calibration curve's calibration

standards. Using a volumetric flask, these dilution levels were created by combining volumes of the dried *S. platensis* culture with distilled water up to 50 mL. All dilution levels were performed in three replicates.

A Shimadzu UV-1800 UV-visible spectrophotometer (Shimadzu, Japan) was used to conduct spectrophotometric measurements. To determine the wavelength to be chosen in the O.D.-cell density regression model, the absorption spectrum of *Spirulina platensis* in the 400-800 nm range was first examined to identify the absorption peaks. According to the APHA method, the best wavelengths to measure O.D. in microalgae cell density calculations are 750, 664, 647,

and 630 nm (APHA AWWA, 2017). Since 750 nm is a wavelength that is significantly longer than the absorbance peak of the chlorophylls in every species we looked at, we chose it for every species. We selected 680 nm and 565 nm for *Spirulina platensis* to target the maximum absorbance of chlorophyll-a and the combined accessory pigments (phycobilin and carotenoids) unique to cyanobacteria, respectively. Distilled water blank was used for baseline comparison. Data analysis was conducted in Microsoft Excel to determine the slope, coefficient of determination (R^2), and standard deviation for both wavelengths.

Table 1. Modified serial dilution of *S. platensis* for preparing standard calibration curve

Calibration Standard	Dried <i>S. platensis</i> Culture (g)	Distilled Water (mL)
0	0.01	50
1	0.02	50
2	0.03	50
3	0.05	50
4	0.075	50
5	0.1	50
6	0.125	50
7	0.15	50
8	0.2	50
9	0.4	50

3. Results And Discussion

Spirulina platensis has been viewed under the 40x lenses of a microscope before and after cultivation, and *Spirulina* species were identified based on their spiral structure morphology. The images from 40x magnification of *Spirulina platensis* cultured in modified Zarrouk medium are shown in Figure 1.

Microscopic analysis showed a high-density *Spirulina* culture with a single strain before and after cultivation. The *Spirulina* cells exhibited uniformity both prior to and following cultivation. Additionally, the absence of bacteria or other foreign organisms in the culture was noted. This finding suggests that the *Spirulina* culture utilized in this study was free from contamination (Barth et al., 2025; Taṽ & Dolinar, 2025).



(a)



(b)

Figure 1. Microscopic image under 40X magnification of *Spirulina platensis* culture before (a) and after cultivation (b) in modified Zarrouk medium.

The result of the measurements is tabulated in Table 2. Table 2 presents the correlation results between microalgal biomass (measured as dry weight) and OD at 565 nm wavelengths. The data for 565 nm (in the green/yellow range) were chosen because this wavelength is commonly found to provide the optimum balance for detecting biomass while minimizing error

from pigment variability (Wacogne et al., 2024). Besides that, the 565 nm wavelength is primarily due to the scattering of light by the cell volume, not the variable pigment concentration inside the cells, and can provide better sensitivity in certain turbidity ranges (Myers et al., 2013; Nanni, 2023).

Table 2. Obtained absorbance values at 565 nm for the prepared *Spirulina* serial dilutions.

Cell Density (gram/Liter)	Optical Density (OD ₅₆₅)	Standard Deviation (SD)
0.2520	0.287	0.00289
0.5000	0.488	0.00321
0.7540	0.779	0.00755
1.0040	0.885	0.00723
1.5060	1.210	0.00351
2.0000	1.372	0.00971
2.5060	1.551	0.00252
3.0120	1.666	0.00436
4.0000	1.922	0.00058

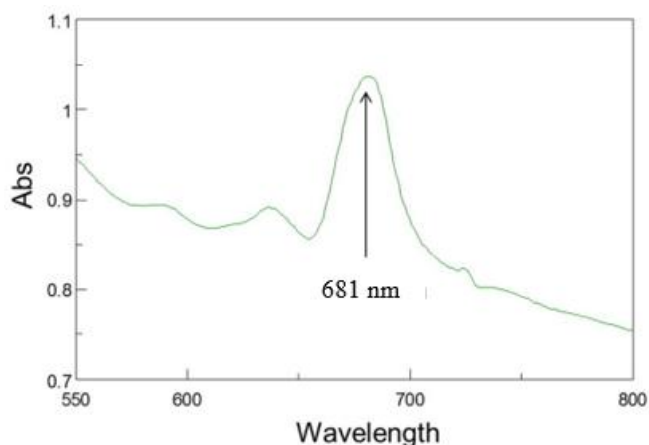


Figure 2. Pattern of light absorbance for a solution with *Spirulina platensis* screened between 400 and 800 nm.

Figure 2 shows the absorption spectrum of the *Spirulina platensis* microalgae culture in the 400–800 nm range, which exhibits a prominent peak at approximately 680 nm. These results indicate that the absorption peak corresponds to the composition of cell pigments and represents the maximum absorbance of chlorophyll a.

Photosynthetic organisms, such as *Spirulina platensis*, have accessory pigments in their chloroplasts that are essential for photosynthesis because they capture light. Important accessory pigments include chlorophyll (chlorophyll a and c for cyanobacteria such as *S. platensis*), carotenoids, and phycobilins (Hotos et al., 2020; Dziosa & Makowska, 2016). In addition to capturing light, these pigments give organisms their specific colors.

The absorbance of all dilution levels at all wavelengths (565; 680; and 750 nm) were used to create a standard curve and determine the experimental parameters, including the detection and upper limits and the linear curve. Figure 3 presents the optical density measurement results.

Figure 3 shows that the optical density (OD₇₅₀) reading begins to reach saturation at a dry weight of *Spirulina* greater than 2 g/L. This is evident from the absorbance value, which approaches a linear line. Therefore, it is not advised to measure samples with an absorbance greater than 3.0 for accurate quantitative measurements since they may be more prone to error (Mori et al., 2021; Phansi et al., 2022). Therefore, data above 2 g/L is excluded from the standard curve.

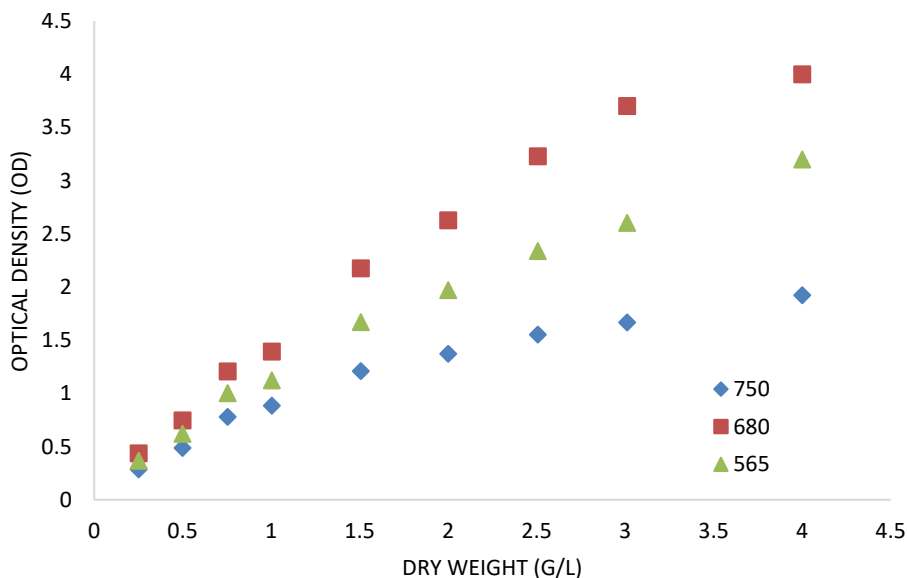


Figure 3. Measured optical density at various dry weight concentrations.

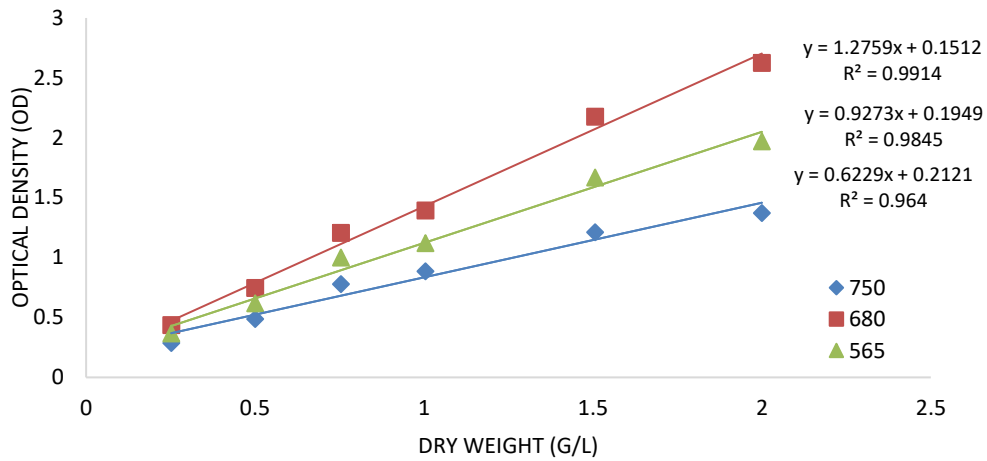


Figure 4. Standard curve for biomass of *Spirulina* using spectrophotometer at 565 nm, 680 nm and 750 nm.

Figure 4 shows that there is a change in absorbance value for every change in *Spirulina*'s dry weight at all positive wavelengths. This indicates that a change in *Spirulina*'s weight will correspond to a change in absorbance value. The correlation coefficient (R^2) varies between 0.96 and 0.99 (close to 1). The high R^2 value indicates that the linear model is a good representation of the variation of the two variables. The highest R^2 value was found when *Spirulina* was observed at a wavelength of 680 nm. Therefore, from the result of this study, cell weight measurements can be more accurately performed using a 680 nm wavelength spectrophotometer.

The results of this study are similar to those of a previous study that used Photopette® (Yap et al., 2018). Another study by Hotos et al (2020) mentioned using a wavelength of 680 nm to estimate cell density in *Nephroselmis* (green microalgae) and *Amphidium* (dinoflagellates). In several studies, the 680 nm wavelength was used to measure cell concentration while being sensitive to pigment content (chlorophyll) (Pahija & Hui, 2019; Katam et al., 2022). Chlorophyll demonstrates a direct proportionality to growth (cell weight) under conditions unaffected by external factors, such as light stress, temperature changes, or the nutrient content of the medium (Hotos et al., 2020; Fakhri et al., 2021; Eddiwan et al., 2023).

The utilization of the OD 680 wavelength can be contemplated in the assessment of cell weight, contingent upon the substantiation that these factors do not contribute to the chlorophyll content in algae cells (Grif et al., 2011; AlFadhly et al., 2022; Eddiwan et al., 2023). However, its application is constrained to the period of logarithmic or exponential growth (Ferreira & Sant, 2017; Nielsen & Hansen, 2019). Following the entry of cells into the stationary phase, there is an accumulation of metabolites, leading to a decrease in chlorophyll content despite an increase in cell weight (Aparicio et al., 2022).

The limit of detection (LOD) for this UV-Vis spectrophotometer measurement is derived from the equation below, accounting for both the variance in blank readings and empirical data:

$$LOD = \frac{3 \times SD_{\text{blank}}}{\text{slope}_{\text{standard curve}}}$$

It is imperative to ascertain the LOD of a UV-Vis spectrophotometer when undertaking calculations pertaining to algae growth, as this parameter delineates the minimum cell concentration that can be reliably discerned from the blank (i.e., the medium). Absent a clearly delineated LOD, it becomes unfeasible to differentiate between the onset of genuine growth and baseline noise, resulting in imprecise growth rates (Agberien & Örmeci, 2020; Malhotra & Örmeci, 2023).

The blank measurements with seven repetitions had a standard deviation of 0.001. The limit of detection for dry mass using a spectrophotometer and the aforementioned equation was therefore as follows.

for 565 nm:

$$LOD = \frac{3 \times 0.001 \text{ OD}}{(0.9273 \text{ OD/gram per Liter})}$$

$$LOD = 0.003 \text{ gram per Liter}$$

for 680 nm:

$$LOD = \frac{3 \times 0.001 \text{ OD}}{(1.2759 \text{ OD/gram per Liter})}$$

$$LOD = 0.002 \text{ gram per Liter}$$

for 750 nm:

$$LOD = \frac{3 \times 0.001 \text{ OD}}{(0.6229 \text{ OD/gram per Liter})}$$

$$LOD = 0.005 \text{ gram per Liter}$$

Our research results show that the 680 nm wavelength produces the smallest LOD value compared to the 565 nm and 750 nm wavelengths. This value indicates that sample measurements at the 680 nm wavelength are more sensitive and capable of measuring smaller cell weights (Taleuzzaman, 2018). The lowest cell weight measurable at 680 nm wavelength is 0.002 grams

per liter. If the cell sample weight is below this value, the measurement cannot be trusted.

The case below perfectly demonstrates the application of this for *Spirulina platensis* biomass calculation. The measured absorbance of the untested sample at 565 nm is 0.818. Using the linear regression analysis formula $y = 0.9273x + 0.1949$ and rearranging it to $x = (y - 0.1949) / 0.9273$, the calculated biomass is 0.641 grams per Liter. The measured biomass of the sample clocks at 0.641 grams per Liter. Similar calculation approach is used for 680 and 750 nm. In instance of known total culture volume (e.g., six 120 Liter culture barrels), the total expected dry biomass for the *Spirulina* harvest can be calculated. For example, if the total volume is 720 Liters (6 x 120 Liters), multiplying by 0.641 g/L yields a total dry weight of 461 grams. The wet biomass of the harvested microalgae can be calculated using the typical 90% moisture content of algal cells, which is ~10 times greater than the dry mass.

Although Optical Density (OD) is a widely used, fast, and cost-effective method for monitoring microalgae growth, this method has weaknesses in predicting dry weight accurately. Specifically, changes in environmental conditions (e.g., nutrient limitation, high emission light, or aging) can cause cells to change their size, shape, and internal structure (Hotos et al., 2020; Malhotra & Ormeci, 2023). Additionally, microalgae growth can reach a stationary phase, which also complicates accurate prediction (Aparicio et al., 2022). Consequently, there is an imperative for an alternative method that can be integrated with OD to predict the weight of microalgae biomass with speed, precision, and reliability, obviating the necessity for frequent calibration regulations.

4. Conclusion

A simple linear model describes the close relationship between cell density and absorbance values as a function of the diversity of spectrophotometer types and wavelengths used. A spectrophotometer with an optical density (OD) of 680 nm is a practical device for estimating the cell density of cyanobacteria, such as *Spirulina platensis*. The dry mass and anticipated total biomass of a *Spirulina* harvest can be measured in a matter of seconds using the technique outlined above. The experiment is inexpensive and simple to carry out. However, due to the shortcomings of this approach, more study is needed to find a different technique that can be combined with OD to quickly, accurately, and reliably forecast the weight of microalgae biomass, eliminating the need for regular calibration regulations.

5. Conflict of Interest

Each author has declared that there is no conflict of interest in the writing or submission of this manuscript.

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7. Author Contribution

All authors have made significant contributions to this work. **FSL** conceptualized and designed the study, conducted data analysis, and drafted the manuscript. **AZ** provided critical revisions and experiments. **AS** assisted in the interpretation of results and contributed to the final manuscript revision. All authors reviewed and approved the final version of the manuscript and agreed to be accountable for all aspects of the work.

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Assessment of Habitat Characteristics Influencing Fish Diversity in the Ijo River, Central Java, Indonesia

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Abstract

Freshwater fish populations are highly sensitive to habitat variation, yet information on how habitat characteristics shape fish diversity in Central Java rivers remains limited. The Ijo River is an important natural system for evaluating ecological factors that determine fish communities. This study aims to assess the physical and chemical characteristics of the Ijo River and evaluate their influence on fish diversity. Research was conducted using survey methods and purposive random sampling at nine stations across three zones: upstream, middle, and downstream. The results obtained are that the environmental parameters varied across river zones. The downstream zone recorded the highest temperature (29.14°C), greatest depth (2.54 m), widest channel (32.97 m), and highest free CO₂ (12.64 ppm). The upstream zone showed the greatest light penetration (74.43 cm), fastest Current Velocity (0.75 m/s), highest dissolved oxygen (5.28 ppm), and highest pH (8.31). Fish diversity also differed between zones. The downstream zone had the highest abundance (4,885 individuals) and greatest dominance (0.17), whereas the upstream zone exhibited the highest Shannon-Wiener diversity index (2.46) and evenness (0.36). These results indicate that upstream zones, with lower temperatures, faster currents, higher dissolved oxygen, and stable pH, support greater species diversity and evenness. Downstream zones, with higher temperatures, elevated free CO₂, and wider river width, show greater abundance but are dominated by a few adaptive species. This pattern, consistent with other rivers, confirms that physical and chemical water characteristics shape distribution, abundance, and dominance of freshwater fish. Thus, fish diversity is a key indicator of river ecosystem health.

1. Introduction

Rivers support the life of fish populations, especially in relation to the diversity and abundance of species that occupy different parts of the river (Gustiano *et al.*, 2025). Such diversity not only reflects the number of species that exist but also indicates the balance of ecosystems that depend on stable environmental conditions. The quality of river water, including temperature, dissolved O₂, pH, and light penetration, is the main factor that determines the presence and abundance of fish, as each of these parameters plays a direct role in the physiological processes and behavior of fish. For example, temperature affects metabolism and growth rate, dissolved O₂ is essential for respiration, pH determines the physiological suitability of fish, while light penetration affects the primary productivity that forms

the basis of the food chain (Napit, 2024; Pratami *et al.*, 2018).

Natural river ecosystems maintain the continuity of river flows so that fish can utilize various microhabitats according to their life needs, because maintained flows allow fish to move freely from upstream to downstream to seek feed, shelter, and reproduce (Sofi *et al.*, 2020). Water physicochemical factors, including variations in current, depth, and substrate, play an important role in determining fish distribution patterns along river flows (Chemagin *et al.*, 2025; Pichon *et al.*, 2016). These natural conditions favor a more balanced interaction between species. The existence of different habitat types also enriches the structure of fish communities in rivers (Scholl *et al.*, 2023; Elviana *et al.*, 2019).



The Ijo River, located in Kebumen Regency, Central Java, is a natural river approximately 36 km long and 14-41 m wide. It covers an area of about 32,902 hectares and includes several tributaries, one of which is the Pringtutul, Kecepek, Tambak, Gumelar, Bulu, Bodo, Demangsari, and Logending Rivers. The Ijo River has relatively stable environmental conditions that support high fish diversity, including species such as Milkfish (*Chanos chanos*), goby (*Glossogobius circumpectus*), barramundi (*Lates calcarifer*), two-spot catfish (*Mystus nigriceps*), Nile tilapia (*Oreochromis niloticus*), Pangas catfish (*Pangasius pangasius*), and mudskipper (*Periophthalmus sp.*) (BPDAS Serayu Opak Progo, 2015). The stability of water quality allows sensitive fish species to survive, while maintaining the balance of freshwater ecosystems in a sustainable manner (Hutapea et al., 2019). The existence of natural rivers is important as a reference for freshwater ecology. An understanding of water quality and its relationship to fish diversity is indispensable for designing sustainable aquatic management strategies (Vyas et al., 2025; Nasriyah & Anas, 2018).

Changes in water physicochemical can affect habitat preferences, fish behavior, and interactions between fish species in their natural habitat (Pahrela et al., 2023). Good water conditions are the basis for the balance of river ecosystems, allowing fish to move freely and maintaining diverse populations (Inwati et al., 2025). Water quality factors also determine the ability of fish to adapt to environmental dynamics; thus, natural river water quality is the main key in maintaining the

sustainability of freshwater ecosystems (Mamun & Kwang, 2022). Freshwater fish populations are highly sensitive to habitat variation, yet information on how physicochemical habitat characteristics shape fish diversity in the Ijo River remains limited. Therefore, this study aimed to evaluate the physical and chemical characteristics of the Ijo River and their influence on fish abundance, diversity, evenness, and dominance across different river zones.

2. Material and Methods

2.1. Study Area

The Ijo River is approximately 36 km long, flowing from the north (Mahameru Mountains) to the south (South Coast) and passing through three districts: Banyumas, Kebumen, and Cilacap. It covers a large area of about 32,902 hectares and includes five sub-districts: Rowokele, Ayah, Tambak, Sumpiuh, and Nusawungu (BPDAS Serayu Opak Progo, 2015). This research collected samples from the Ijo River from November 2022 to October 2023, with one sampling conducted each month for a frequency total of 12 times. The survey method was applied using purposive random sampling. The purposive aspect involved dividing the river into 9 stations across three zones the upstream (stations 1-3), the middle part (stations 4-6), and the downstream (stations 7-9). Random sampling was conducted by collecting fish samples from riverbank and midstream habitats to obtain representative data. The study area is shown in Figure 1, and detailed sampling site characteristics are provided in Table 1.

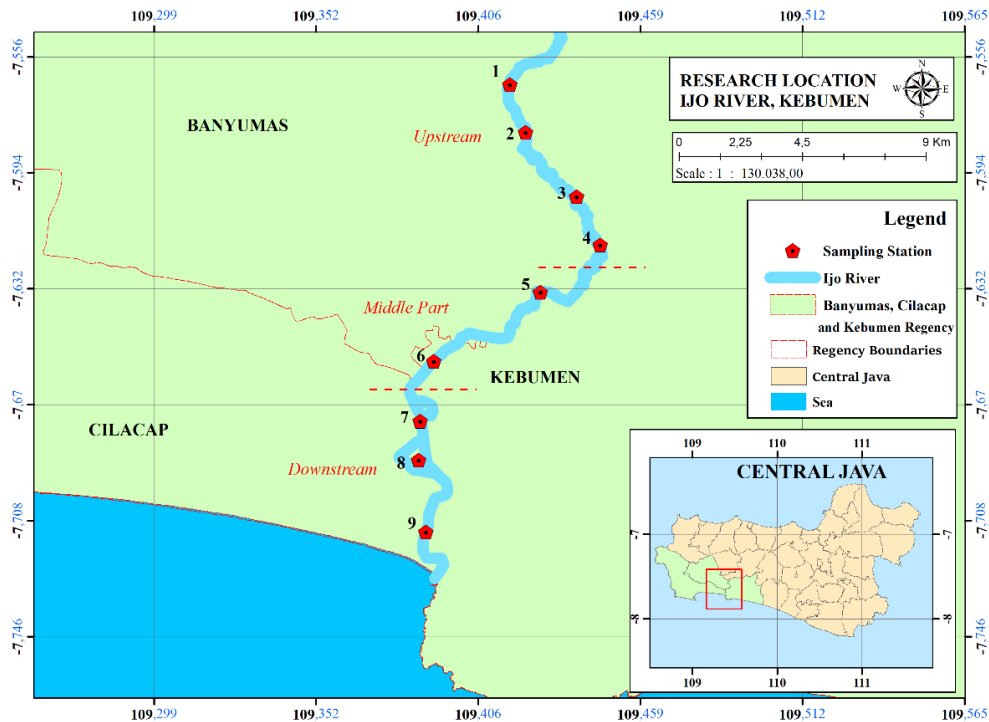


Figure 1. Map of the study area showing the nine sampling stations distributed across the upstream, middle reach, and downstream zones of the Ijo River, Indonesia.

Table 1. Sampling stations, geographic coordinates, and surrounding land use characteristics along the Ijo River, Kebumen Regency, Central Java, Indonesia

Zone	Station	Geographic Coordinates	Surrounding Land Use
Upstream	1	7°33'37.8"S 109°25'05.9"E	Forest, rice fields
	2	7°34'33.8"S 109°25'07.9"E	Forest, rice fields
	3	7°36'22.4"S 109°26'24.2"E	Forest, rice fields, residential area
Middle Reach	4	7°37'14.6"S 109°26'46.9"E	Rice fields, plantations, residential area
	5	7°37'57.5"S 109°25'36.4"E	Rice fields, plantations, residential area
	6	7°39'14.1"S 109°23'33.3"E	Rice fields, plantations, residential area
Downstream	7	7°40'32.7"S 109°23'14.6"E	Rice fields, plantations, residential area
	8	7°41'15.3"S 109°23'17.9"E	Rice fields, plantations, residential area
	9	7°42'48.3"S 109°23'16.5"E	Rice fields, plantations, residential area

2.2. Sampling of Water Quality Parameters

Water samples were collected directly (in situ) and analyzed for two categories of parameters: physical and chemical. The physical parameters included temperature, water transparency, current velocity, river width, and depth, while the chemical parameters comprised pH, dissolved oxygen (DO), and free carbon dioxide (CO₂). The methods used for water sampling are summarized as follows.

2.2.1. Temperature

Water temperature was measured using a water thermometer (°C). The thermometer was immersed directly into the river for approximately two minutes until a stable reading was obtained. According to APHA (2005), measurement of water temperature is carried out in situ by placing the thermometer in the water body until equilibrium is reached, thereby ensuring accurate representation of the actual river conditions.

2.2.2. Water Transparency

Water transparency was measured using a Secchi disk (cm). The disk was lowered vertically into the river until it was no longer visible, and the depth at which it disappeared was recorded. According to Mikhail & Sergey (2024), water transparency measured using a Secchi disk by slowly lowering the Secchi disk into the water until the black and white pattern on the Secchi disk is no longer visible (D1), then the Secchi disk is slowly lifted again until the black and white pattern is visible again (D2).

$$\text{Water Transparency} = \frac{D1 + D2}{2}$$

2.2.3. Current Velocity

River current velocity was measured using the bottle and rope method (m/s). A weighted bottle was attached to a rope and released into the river, allowing it to drift with the current. The time taken for the bottle to travel a predetermined distance was recorded, and current velocity was calculated by dividing the distance travelled by the elapsed time.

2.2.4. River Width

River width was measured using a measuring tape (m). The tape was stretched across the river from one bank to the other at each sampling station, ensuring that the measurement was taken perpendicular to the flow direction. River width measurement should be conducted in situ by directly spanning the water body with a calibrated instrument, thereby providing an accurate representation of channel morphology.

2.2.5. River Depth

River depth was measured using a calibrated pole with a scale (m). The pole was lowered vertically into the river until it reached the substrate, and the depth was recorded based on the scale markings.

2.2.6. pH

Water pH was measured in situ using a portable pH meter (unitless). The electrode of the pH meter was immersed directly into the river water and allowed to stabilize before recording the value. According to APHA (2005), pH measurement should be conducted by placing the electrode in the water body under field conditions to obtain accurate readings. This parameter is essential for assessing the acidity or alkalinity of the river, which influences chemical processes, nutrient availability, and the suitability of aquatic habitats.

2.2.7. Dissolved Oxygen

DO concentration was measured in situ using a Water Quality Checker (WQC) and expressed in parts per million (ppm). The probe was immersed directly into the river water and allowed to stabilize before recording the value. According to APHA (2005), DO measurement should be conducted under field conditions to ensure accurate representation of oxygen availability in the aquatic environment.

2.2.8. Free-CO₂

Free carbon dioxide (CO₂) concentration was measured in situ using a WQC and expressed in parts per million (ppm). The probe was immersed directly into the

river water and allowed to stabilize before recording the value. According to APHA (2005), free CO₂ measurement should be conducted under field conditions to obtain accurate readings of dissolved carbon dioxide in the aquatic environment. This parameter is important for evaluating the balance of respiration and photosynthesis, buffering capacity, and the overall chemical dynamics of the river.

2.3. Fish Sampling

Fish samples were obtained from the help of fishermen in each predetermined zone. Fish were taken in the upstream zone of the river with traditional fishing gear (rattan fish trap, scoop net, and stocking net). Sampling in the middle and downstream zones using rattan fish traps and gillnets. Installation of fishing gear at 04.00-08.00 WIB (duration 4 hours). The fish specimens (*M. nigriceps*) were put in an ice box, then taken to a base camp (fisherman's house) to be observed measured length and weight. Fish samples were identified by (Froese & Pauly, 2024; Kottelat et al., 1993; Saanin, 1968).

2.4. Data Analysis

Water quality data were analyzed descriptively and quantitatively. Fish community data were assessed using four metrics, assessed using four metrics, which include abundance, the Shannon-Wiener diversity index, Pielou's evenness index, and the Simpson dominance index. The calculation of the Shannon-Wiener diversity index, evenness index, and dominance index followed the references provided by Magurran (1988). The Shannon-Wiener diversity index (H') was calculated as:

$$H' = - \sum_{i=1}^s \left(\frac{ni}{N}\right) \ln \left(\frac{ni}{N}\right)$$

where:

- H' = Shannon-Wiener diversity index;
- s = Total number of fish species caught;
- ni = Number of individuals of species i caught;
- N = Total number of individuals of all species caught;
- ln = Natural logarithm.

Pielou's evenness index (E) was calculated as:

$$E = \frac{H'_a}{\ln (s)}$$

where:

- E = Pielou's evenness index;
- H' = Shannon-Wiener diversity index;
- s = Total number of species.

Simpson's dominance index (C) was calculated as:

$$C = \sum \left(\frac{ni}{N}\right)^2$$

where:

- C = Simpson's Dominance index;
- ni = Number of individuals of species i;
- N = Total number of individuals of all species.

Fish diversity data obtained in this research were analyzed in relation to water quality parameters using Canonical Correspondence Analysis (CCA). This multivariate method was applied to explain the relationship between fish community structure and environmental variables. Analyses of fish communities and CCA were conducted using PAST software, version 5.0.

3. Material and Methods

3.1. Habitat Characteristics

The habitat characteristics measured, consisting of the physics and chemistry of the water, can be seen in Table 2. Water quality parameters in the Ijo River display distinct spatial gradients from upstream to downstream reaches. The upstream zone has the lowest temperatures (25.2-25.8°C), which increase in midstream sections (28.8-29.5°C) and remain high downstream (28.1-28.8 °C). Transparency shows an inverse pattern, with the highest values upstream at 67.9-80.0 cm and the lowest downstream at 30.3-36.9 cm. River depth and width increase progressively, from the lowest values upstream (0.8-1.3 m deep; 13.6-18.4 m wide) to the highest downstream (2.3-2.8 m deep; 26.2-41.4 m wide). Flow velocity decreases along this gradient, from the highest at 0.83 m/s upstream to the lowest at 0.35-0.47 m/s downstream. Chemical profiles reinforce these trends: DO reaches the highest levels upstream (4.7-6.1 mg/L) and the lowest downstream (2.8-3.4 mg/L), while free CO₂ shows the opposite, with the lowest upstream (2.5-6.8 mg/L) and the highest downstream (11.6-13.7 mg/L). pH follows suit, highest upstream (8.1-8.5) and lowest downstream (6.7-7.2). Overall, upstream zones maintain the highest oxygen and transparency levels with the lowest temperatures and CO₂, while downstream areas show the opposite patterns.

Increasing water temperature accelerates the metabolism and respiration of aquatic organisms, thereby raising oxygen demand and contributing to a decrease in dissolved oxygen (Sarkar et al., 2017; Rouf et al., 2022). The brightness results obtained in Ijo River were higher than previous research in Logawa River, Banyumas, which obtained a brightness value ranging from 13.6-58.6 cm. The brightness range obtained supports fish life in the Logawa River because the brightness can trigger sunlight to enter the river waters, helping the fish move to forage and grow well. Water-soluble substances often block the penetration of light, limiting photosynthesis zones for aquatic habitats (Susanto & Novitasari, 2017). The width of the Ijo River is higher than previous research on the Towkak River, India, with a width of 24.38 - 30.48 m (Dutta, 2016). The range obtained supports the life of more fish in the habitat of the Pakil River because the wider river supports a greater variety of habitats compared to narrow rivers, so that the longer and wider the size of the river, the more fish inhabit it (Zulfikri et al., 2016; Dutta, 2016). The depth in the Ijo River is lower than the previous study in the Lok Ulo River,

Kebumen; the depth was obtained in the range of 4.00-6.00 m (Prakoso & Wahyuni, 2019). The depth of the river will increase if it rains and the depth of the water affects the light penetration of a water, the deeper a water, the lower the light intensity. Increasing depth will result in a decrease in dissolved oxygen levels, because the

photosynthesis process decreases and the existing oxygen levels are widely used for respiration and oxidation of organic and inorganic materials (Syahrul et al., 2021).

Table 1. Physicochemical characteristics of water across sampling stations in the Ijo River

Zone	St	Temperature (°C)	Water Transparency (cm)	Depth (m)	Current Velocity (m/s)	River Width (m)	Dissolved Oxygen (mg/L)	Free-CO ₂ (mg/L)	pH
Upstream	1	25.05 - 25.48 (25.24)	78.5 - 81.2 (80.0)	0.78 - 0.82 (0.8)	0.81 - 0.85 (0.83)	13.5 - 13.7 (13.63)	6 - 6.15 (6.08)	2.4 - 2.6 (2.5)	8.45 - 8.55 (8.5)
	2	25.39 - 25.71 (25.53)	75.1 - 76 (75.6)	0.81 - 0.85 (0.83)	0.69 - 0.73 (0.71)	18 - 18.22 (18.14)	5 - 5.1 (5.05)	4.6 - 4.8 (4.7)	8.25 - 8.35 (8.3)
	3	25.68 - 25.95 (25.83)	67.5 - 68.2 (67.88)	1.28 - 1.35 (1.32)	0.68 - 0.72 (0.70)	18.3 - 18.48 (18.4)	4.65 - 4.75 (4.7)	6.65 - 6.85 (6.75)	8.1 - 8.15 (8.13)
Middle Part	4	28.65 - 28.95 (28.80)	55.5 - 56.2 (55.9)	1.32 - 1.37 (1.34)	0.57 - 0.61 (0.59)	20.8 - 21.08 (20.96)	4.6 - 4.68 (4.64)	7.7 - 7.95 (7.84)	7.75 - 7.8 (7.78)
	5	29.05 - 29.28 (29.15)	48.8 - 49.5 (49.15)	1.65 - 1.72 (1.69)	0.49 - 0.53 (0.51)	22.1 - 22.45 (22.28)	4.18 - 4.25 (4.22)	7.95 - 8.2 (8.09)	7.5 - 7.6 (7.55)
	6	29.38 - 29.55 (29.46)	42 - 42.5 (42.25)	1.9 - 1.97 (1.94)	0.46 - 0.5 (0.48)	22.5 - 22.85 (22.68)	3.7 - 3.75 (3.73)	8.6 - 8.9 (8.78)	7.4 - 7.45 (7.43)
Downstream	7	28 - 28.25 (28.11)	36.8 - 37.1 (36.95)	2.22 - 2.28 (2.25)	0.45 - 0.49 (0.47)	26 - 26.45 (26.24)	3.35 - 3.4 (3.38)	11.5 - 11.8 (11.65)	7.15 - 7.25 (7.2)
	8	28.3 - 28.5 (28.43)	34.8 - 35.1 (34.95)	2.5 - 2.6 (2.56)	0.34 - 0.38 (0.36)	31 - 31.4 (31.20)	3.17 - 3.22 (3.2)	12.5 - 12.8 (12.56)	7.05 - 7.1 (7.08)
	9	28.65 - 28.85 (28.75)	30 - 30.5 (30.25)	2.75 - 2.88 (2.82)	0.33 - 0.37 (0.35)	41.2 - 41.55 (41.39)	2.72 - 2.77 (2.75)	13.5 - 13.8 (13.65)	6.65 - 6.75 (6.7)

Data are presented as range values, with mean values shown in parentheses.

Current Velocity is grouped into 5 categories, namely very fast (>1 m/s), fast (0.5-1 m/s), medium (0.25-0.5 m/s), slow (0.1-0.2 m/s), and very slow (<0.1 m/s) (Kamboj et al., 2020). Based on the results, the flow speed obtained in the Ijo River is classified as moderate to fast. Current Velocity in the Ijo River decreases from upstream (0.83 m/s, classified as fast) to downstream (0.35-0.47 m/s, moderate), consistent with river hydrology. This pattern arises because upstream sections are narrower (13.6-18.4 m) and shallower (0.8-1.3 m) with steeper gradients, concentrating discharge into higher current velocity. Downstream widening (26.2-41.4 m) and deepening (2.3-2.8 m), combined with reduced slopes and increased bed roughness from sediments and vegetation, slow the water. The results obtained in this study are higher than the research in the Lamunde River, Southeast Sulawesi, the current velocity is obtained in the range of 0.105 - 0.388 m/s (Jukri et al., 2013). However, the results obtained in the Ijo River were lower than the research in the Cikawung River, Cilacap, the current velocity was obtained in the range of 0.06-1.0 m/s, which is relatively fast (Nuryanto et al., 2015).

Dissolved oxygen in the Ijo River decreases from upstream (4.7-6.1 mg/L) to downstream (2.8-3.4 mg/L), lower overall than in the Cikawung River (3.8-9.8 mg/L), where levels support diverse fish life (Nuryanto et al., 2015). This downstream decline stems from reduced atmospheric reaeration due to lower flow velocities (0.83 m/s upstream vs. 0.35-0.47 m/s downstream), warmer

temperatures (25.2-25.8 °C upstream vs. 28.1-28.8 °C downstream). Slower current velocity limits turbulence-driven gas exchange at the air-water interface, while temperature inversely affects dissolved oxygen solubility (Olopade et al., 2017). Conversely, free CO₂ rises downstream (2.5-6.8 mg/L upstream to 11.6-13.7 mg/L), though lower than in the polluted Ampenan River (Idrus, 2018). This increase results from elevated respiration by biota, organic matter decomposition, and groundwater inputs in low-velocity, warmer downstream zones—processes amplified by physical factors like reduced flow, which hinders CO₂ off-gassing. Flow velocity directly influences both: high upstream speeds enhance aeration (boosting dissolved oxygen, venting free-CO₂), while low downstream speeds trap respiratory gases. Optimum DO for fish survival and growth varies by species and life stage but generally ranges from 5-8 mg/L for most tropical freshwater fishes; levels below 3-4 mg/L cause stress, reduced growth, and mortality.

The pH values recorded in the Ijo River tend to decline from upstream to downstream, reflecting increased organic matter decomposition and CO₂ accumulation in slower-flowing downstream waters. In comparison, the Aur Lemau River in Bengkulu maintains a stable pH of 6.6-6.8, which falls within the acceptable quality standard (6-9). This decline in pH downstream is ecologically significant, as excessively low values can increase heavy metal solubility and threaten aquatic life, while excessively high values can elevate ammonia

concentrations (Febrian et al., 2022). For most freshwater fish, the optimum pH for survival and growth lies between 6.5 and 8.5, with near-neutral conditions (around 7.0-7.5) being most favorable (Milbrath et al., 2025).

3.2. Fish Diversity

Table 3 summarizes fish abundance and ecological indices, including the Shannon-Wiener diversity, evenness, and dominance indices, across the Ijo River. A

total of 11,096 fish individuals were recorded gradually increasing from the upstream zone (1,663 individuals) to the downstream zone of 4,885 individuals. Similarly, in the Opak River, Yogyakarta, fish were more abundant in the downstream zone, with 2,295 individuals and the lowest in the upstream zone, with as many as 1,204 individuals, the results obtained are suspected because downstream of the river usually has a deeper water depth, a varied substrate (Yudha et al., 2020).

Table 2. Fish community abundance and ecological indices in the upstream, middle reach, and downstream zones of the Ijo River

Ecological Indices	Ijo River		
	Upstream	Middle Reach	Downstream
Number of Species	32	41	41
Abundance	1,663	4,548	4,885
Shannon-Wiener Diversity (H')	2.46	2.43	2.37
Pielou's Evenness (E)	0.36	0.28	0.26
Simpson's Dominance (C)	0.14	0.16	0.17

The ecological index in the Ijo River consists of 3 types. The highest Shannon-Wiener diversity index is in the upstream zone (2.46) and the lowest in the downstream zone (2.37). The Pielou's evenness index shows the same value as the Shannon-Wiener diversity index, which is the highest in the upstream zone (0.36) and the lowest in the downstream zone (0.26). The Simpson's dominance index shows the difference between the two indices above with the highest value in the downstream zone (0.17) and the lowest in the upstream zone (0.14). The upstream zone of the Ijo River exhibited the highest Shannon-Wiener diversity index and Pielou's evenness, a pattern likewise reported in the Keriau River, West Kalimantan, and the Batang Uleh River, Jambi. Variations in fish species diversity are strongly associated with habitat features, including river width and substrate composition, which determine the degree of diversity (Prayogo et al., 2022; Budiman et al., 2021).

The higher Simpson's dominance index in the downstream zone of the Ijo River indicates that several fish species dominate this area. A similar pattern was observed in the waters of Krueng Raya, Aceh, where the downstream zone recorded a Simpson's dominance index (C) of 0.21. These findings suggest that a high Simpson's dominance index reflects the predominance of certain species in the downstream zone. Moreover, an increase in Simpson's dominance index is typically associated with lower Shannon-Wiener diversity and evenness values (Ulfah et al. 2019).

3.3. The Relationship of Habitat Characteristics to Fish Diversity

Canonical Correspondence Analysis (CCA) is a multivariate statistical method used in ecology to explore

and quantify the relationships between biological communities (such as fish diversity) and environmental variables (such as habitat characteristics). It essentially links species composition data with environmental gradients, allowing researchers to see how differences in habitat conditions influence the distribution and diversity of species is obtained in the CCA diagram (Figure 2).

Based on the in Figure 2, CCA in Ijo River revealed that Shannon-Wiener diversity and Pielou's evenness are closely associated with upstream parameters such as water transparency, current velocity, dissolved oxygen, and pH. Conversely, fish abundance and Simpson's dominance index show affinity with downstream parameters including river width, depth, temperature, and free CO₂. These findings indicate that the upstream zone supports a more equitable distribution of species, while the downstream zone is characterized by high abundance and dominance of species adapted to warmer, deeper waters with elevated CO₂. Comparable patterns have been documented globally. In the Wampu River, North Sumatra, higher Shannon-Wiener diversity and Pielou's evenness in upstream zones were linked to cooler temperatures, faster flows, and higher dissolved oxygen (Desrita et al., 2019). Similarly, in the Bichom River, India, upstream habitats with greater light penetration and current velocity supported more diverse fish assemblages (Nimasow et al., 2025). In China's Lijiang River, CCA analysis confirmed that upstream zones with high current speed and dissolved oxygen were significantly correlated with Shannon-Wiener diversity and Pielou's evenness. These parallels highlight a consistent ecological principle: habitat heterogeneity drives fish diversity across river systems (Huang et al., 2019).

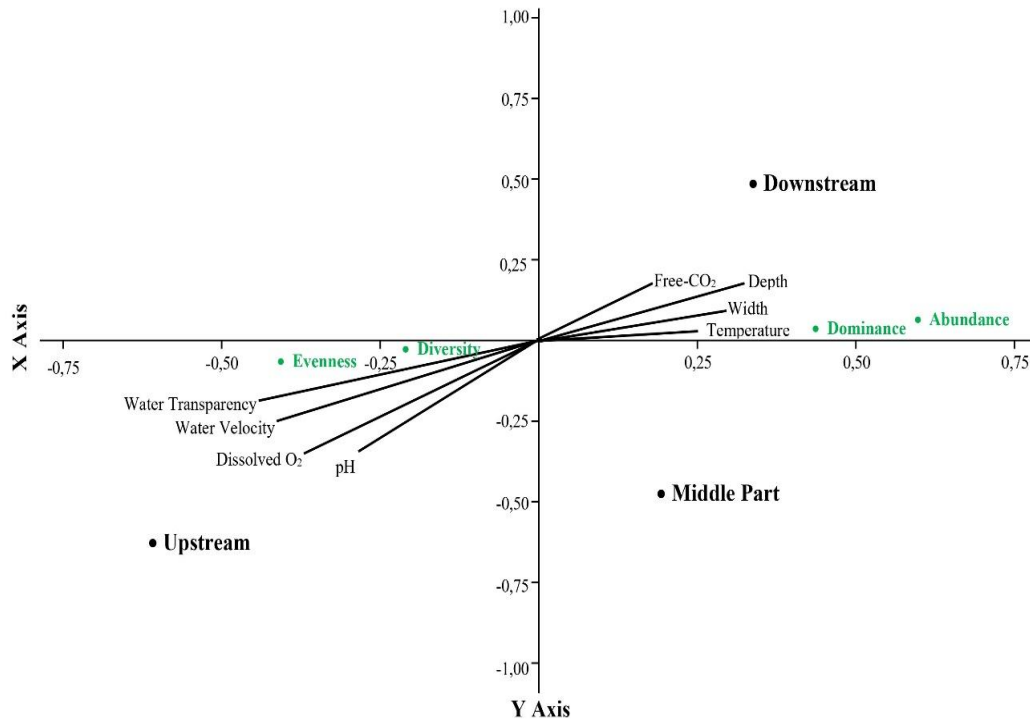


Figure 1. Canonical Correspondence Analysis (CCA) showing the relationships between physicochemical water parameters and fish community indices across river zones in the Ijo River.

Beyond local and regional contexts, these findings carry global implications. Freshwater ecosystems are increasingly threatened by anthropogenic pressures such as dam construction, pollution, acidification, and climate change. Rising temperatures and altered flow regimes can shift habitat suitability, often favoring a few tolerant species while reducing overall diversity. This has cascading effects on ecosystem services, including nutrient cycling, food security, and livelihoods dependent on inland fisheries. The observed relationship between habitat characteristics and fish diversity underscores the importance of conserving upstream habitats, which often act as biodiversity reservoirs. Protecting these zones contributes not only to local ecological balance but also to global efforts in maintaining freshwater resilience under climate change. Moreover, indices such as Shannon-Wiener diversity, Pielou’s evenness, and Simpson’s dominance provide sensitive bioindicators of ecosystem health, offering valuable insights for monitoring and managing rivers in the face of global challenges such as acidification and warming.

In the downstream zone of the Ijo River, a higher Simpson’s dominance index indicates that several fish species dominate the community. The dominant taxa recorded in this study include Long-whiskered catfish (*Mystus gulio*), two-spot catfish (*Mystus nigriceps*), climbing perch (*Anabas testudineus*), Nile tilapia (*Oreochromis niloticus*), snakeskin gourami (*Trichogaster pectoralis*), and flathead grey mullet (*Mugil cephalus*). This pattern reflects their ability to

adapt to downstream habitats characterized by wider and deeper channels, higher temperatures, and the presence of free CO₂. Comparable findings have been reported in other rivers, such as the Garonne River in France, where species like *European grayling* (*Thymallus thymallus*), *common barbel* (*Barbus barbus*), and *common bream* (*Abramis brama*) dominate downstream zones (Santoul et al., 2025). In the Bengawan River, Solo, species such as Nile tilapia (*Oreochromis niloticus*), Pangas catfish (*Pangasianodon hypophthalmus*), suckermouth catfish (*Pterygoplichthys pardalis*), Patin juaro (*Pangasius polyuranodon*), and enoplos barb (*Cyclocheilichthys enoplos*) are prevail (Aida et al., 2022). These dominances are linked to adaptive traits such as tolerance to habitat variability, flexible foraging strategies across water columns and benthic zones, and reproductive strategies suited to downstream environments.

4. Conclusion

This study demonstrates that spatial variations in physicochemical water characteristics along the Ijo River strongly influence fish community structure and ecological distribution patterns. Upstream zones, characterized by lower temperatures, faster current velocity, higher dissolved oxygen concentrations, greater water transparency, and more stable pH conditions, supported higher Shannon-Wiener diversity and Pielou’s evenness indices, indicating a more balanced and diverse fish assemblage. In contrast, downstream zones, which exhibited higher

temperatures, elevated free CO₂ concentrations, and greater river width and depth, showed higher fish abundance but were dominated by a limited number of adaptive species, as reflected by the higher Simpson's dominance index. These findings confirm that habitat heterogeneity and physicochemical conditions are key determinants of freshwater fish distribution, abundance, diversity, and dominance in tropical river ecosystems.

Our study further demonstrates that ecological indices such as Shannon-Wiener diversity, Pielou's evenness, and Simpson's dominance are effective bioindicators for evaluating river ecosystem health. The upstream zone functions as an important biodiversity reservoir, whereas downstream areas indicate ecological pressure that favors tolerant and opportunistic species. This ecological differentiation has important implications for conservation prioritization and sustainable freshwater resource management.

More broadly, the study provides a practical ecological framework for river ecosystem monitoring using fish community indices and environmental parameters. The observed patterns also suggest that increasing temperature and hydrological alteration associated with climate change may intensify downstream environmental stress, potentially reduce biodiversity and increase species dominance. Furthermore, the findings are consistent with global freshwater challenges, including habitat degradation, eutrophication, and ecological fragmentation, which continue to threaten riverine ecosystems worldwide. Therefore, the integration of fish diversity indices with physicochemical assessments can support evidence-based policy development, adaptive river management strategies, and long-term freshwater conservation planning.

5. Data Availability Statement

All data utilized in this study are openly accessible and do not contain any confidential or ethically sensitive information

6. Funding Institutions

This research received no external funding.

7. Conflict of Interest

This manuscript has no declarations or potential conflicts of interest among the authors.

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Vertical Distribution of Chl-a in Relation to Environmental Factors and Water Column Stratification in the Downstream Section of the Air Bengkulu River

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Abstract

The Bengkulu Watershed, which spans two administrative districts, plays a crucial role in supporting ecological and socio-economic functions in Bengkulu Province, Indonesia. Its main river, the Air Bengkulu, has experienced environmental pressures from land conversion, coal mining activities, domestic waste discharge, and industrial effluents, which may alter downstream water quality and disrupt estuarine ecological processes, including primary productivity. Historically, the adjacent coastal area was utilized by local fishers as a fishing ground in the early 2000s, but this function has since declined. However, studies on the ecological implications of these pressures on phytoplankton production in the Air Bengkulu estuary remain limited. This study aimed to assess phytoplankton productivity using Chl-a concentration as a proxy and examine its relationship with key environmental parameters. Fieldwork was conducted during the peak dry season from June to August 2025, with two sampling sessions per event in the morning and afternoon under spring tide conditions. Observations were carried out at three stations on three sampling events with measurements taken at three depth levels. Salinity, current velocity, dissolved oxygen, and nitrate concentration were measured at all depths, while temperature, pH, and water transparency were limited to surface levels. Chlorophyll-a (Chl-a) concentration was analyzed using spectrophotometric methods and its relationship with environmental parameters was evaluated using correlation analysis and quantile regression. The results showed that Chl-a concentrations were consistently very low across stations, ranging from 0.025 to 0.139 $\mu\text{g/L}$, indicating ultra-microtrophic to oligotrophic conditions. Among the measured parameters, nitrate concentration and current velocity exhibited the strongest relationships with Chl-a, suggesting their role as primary limiting factors, while other parameters showed comparatively weaker influences. These findings indicate that low nutrient availability and hydrodynamic conditions constrain phytoplankton biomass development and primary productivity in the Air Bengkulu River estuary, potentially limiting its capacity to sustainably support the food web.

1. Introduction

An estuary is a tidally brackish water body where riverine and marine waters interact (Nontji, 2008). Accordingly, the downstream section of a river is considered part of the estuarine system, as it remains under the combined influence of river discharge and

marine dynamics. As dynamic and biogeochemically active environments, estuaries regulate primary productivity through coupled physical, chemical, and biological processes, along with substantial inputs of nutrients and organic matter (Cloern et al., 2014; Junior, 2025).



Phytoplankton, microscopic algae inhabiting aquatic systems, are highly responsive to environmental changes, making them a reliable indicator of water quality (EPA, 2023). Chlorophyll-a (Chl-a), a common photosynthetic pigment in all phytoplankton, is widely used as an indirect measure of algal carbon biomass and a proxy for primary productivity (Damar et al., 2020; Gall and Pinkerton, 2024), which underpins estuarine food webs and indicates ecosystem function and water quality.

Phytoplankton communities in estuaries typically consist of both freshwater and marine species due to continuously changing conditions driven by tidal dynamics. Their distribution depends on hydrodynamic processes and salinity tolerance (Nontji, 2008). The mixing of freshwater and seawater causes fluctuations in salinity, current velocity, and light availability, thereby influencing phytoplankton metabolism and photosynthesis, and shaping community abundance, distribution, and diversity (Cereja et al., 2021; Neun et al., 2022; Thrush et al., 2014; Xia et al., 2024; Ye et al., 2025).

Salinity regulates algal metabolism through osmotic stress and nutrient-sediment interactions, leading to spatial and temporal variability in nutrient availability (Orizar et al., 2024; Thrush et al., 2014). Differences in salinity tolerance affect metabolic performance, productivity, and survival, particularly in freshwater phytoplankton, with only a limited number of species able to persist across the full salinity gradient of estuarine environments (Cereja et al., 2021; Lancelot and Muylaert, 2011; Lionard et al., 2005; Steidle and Vennell, 2023).

Current velocity, governed by tidal dynamics, morphology, and upstream discharge (Purnaini and Purwono, 2018; Tendean, 2017), drives the transport of oxygen, nutrients, and organic matter while shaping Chl-a distribution (Reseck, 1988; Dahuri, 2003; Wang et al., 2024). Together with salinity, hydrodynamic variability enhances water mixing and turbidity, often constraining photosynthesis by reducing light penetration (Cloern et al., 2014). Interactions between current velocity and residence time can limit phytoplankton accumulation despite high nutrient availability (Zhong and Chien, 2024), whereas lower velocities promote retention within the optimal light zone and increase Chl-a concentrations (Zhang et al., 2015; Steidle and Vennell, 2023).

Phytoplankton production is further influenced by water column stratification and mixing processes driven by freshwater inflow and tidal dynamics (Cloern et al., 2014). Stratification is defined as the development of vertically distinct water layers driven by density gradients, such as salinity (WetlandInfo, 2023). Strong stratification promotes phytoplankton growth by retaining cells within the euphotic zone, whereas intensified mixing redistributes cells below light-limited depths, reducing photosynthetic efficiency (Cloern et al., 2014; Gall et al., 2023). Environmental changes modify stratification intensity and mixed layer depth, influencing vertical gradients in primary productivity (Gall and Pinkerton,

2024; Xia et al., 2024; Zhong and Chien, 2024). This vertical variability is reflected in phytoplankton distribution across depths where phytoplankton productivity is typically higher near the surface, while in deeper layers, chlorophyll concentrations increase only when sufficient light penetrates (Domingues and Barbosa, 2023; Gall et al., 2023; Gonçalves-Araujo and Markager, 2020). However, this pattern is not always uniform due to mixing and stratification dynamics.

In addition to physical drivers, nutrient availability also regulates phytoplankton dynamics. Nitrate is an essential nutrient required for the synthesis of proteins, chlorophyll, and nucleic acids (Effendi, 2003; Nasution et al., 2019; Zakem et al., 2018). Elevated nitrate concentrations in estuarine systems often indicate anthropogenic inputs from river basins with intensive human activities, including wastewater discharge, agricultural runoff, septic leakage, and industrial effluents (EPA, 2000; González-Ramírez et al., 2023). This study uses nitrate as a nutrient parameter to establish baseline data for future research in the study area.

Land-based anthropogenic activities are a major source of nutrient inputs in estuarine systems. Land use conversion in the upstream area, intensive oil palm plantations, coal and river sand mining, and rubber processing activities in the middle reaches, as well as poorly planned coastal urban development in the downstream section, have contributed to the degradation of the Air Bengkulu River (Pareke and Putra, 2014). Belladonna (2017) also reported substantial pollution from industrial effluents at multiple monitoring sites. The river water quality has been classified as moderately polluted by Dinas Lingkungan Hidup Bengkulu (2023) and has also been considered unsuitable as a raw water source due to pollution (Wijayanto et al., 2025). Consistent with these findings, the Bengkulu watershed has been categorized as being in poor condition and in need of restoration (BPDAS Tahun, 2024). These cumulative pressures across the Air Bengkulu River watershed have the potential to alter downstream physicochemical conditions through riverine transport processes, leading to both deterioration of estuarine ecological functions and increased coastal vulnerability.

In the Air Bengkulu estuary, previous studies have predominantly focused on pollution status and surface-level physicochemical conditions, with limited attention to vertical variability and biological responses, particularly in relation to primary productivity. This may overlook important depth-related processes associated with stratification and mixing, resulting in an incomplete understanding of primary productivity in this area when assessed using Chl-a as a proxy.

Therefore, this study aims to assess phytoplankton dynamics represented by Chl-a concentration, with a focus on its vertical distribution and its relationship with key environmental parameters in the downstream section of the Air Bengkulu estuarine system. The study

was conducted during the peak of the dry season, representing conditions of reduced freshwater discharge and stronger marine influence.

2. Materials and Methods

2.1. Study Area

The study area is characterized by a mixed, predominantly semidiurnal tidal regime, in which two high tides and two low tides may occur each day with unequal heights and periods (Hasibuan et al., 2020). Field measurements indicate that water depths in the river mouth zone range from approximately 1 to 4 meters (m), classifying the system as a shallow estuary. The study area encompasses approximately 1 kilometer (km) of the

river channel extending upstream from the Air Bengkulu River mouth as presented in Figure 1.

Sampling was conducted during the peak of the dry season, as indicated by climatological records from BMKG Provinsi Bengkulu (2025). During this period, rainfall was relatively low, with total monthly precipitation ranging from 0 to 155 mm, with a mean of 9.4 mm, based on daily rainfall data obtained from the BMKG (2025) online database. Rainfall in the study area was recorded only once throughout the sampling period, occurring during the third sampling event at Station 2 at 17.00 local time. Details of the sampling schedule are provided in Table 1.

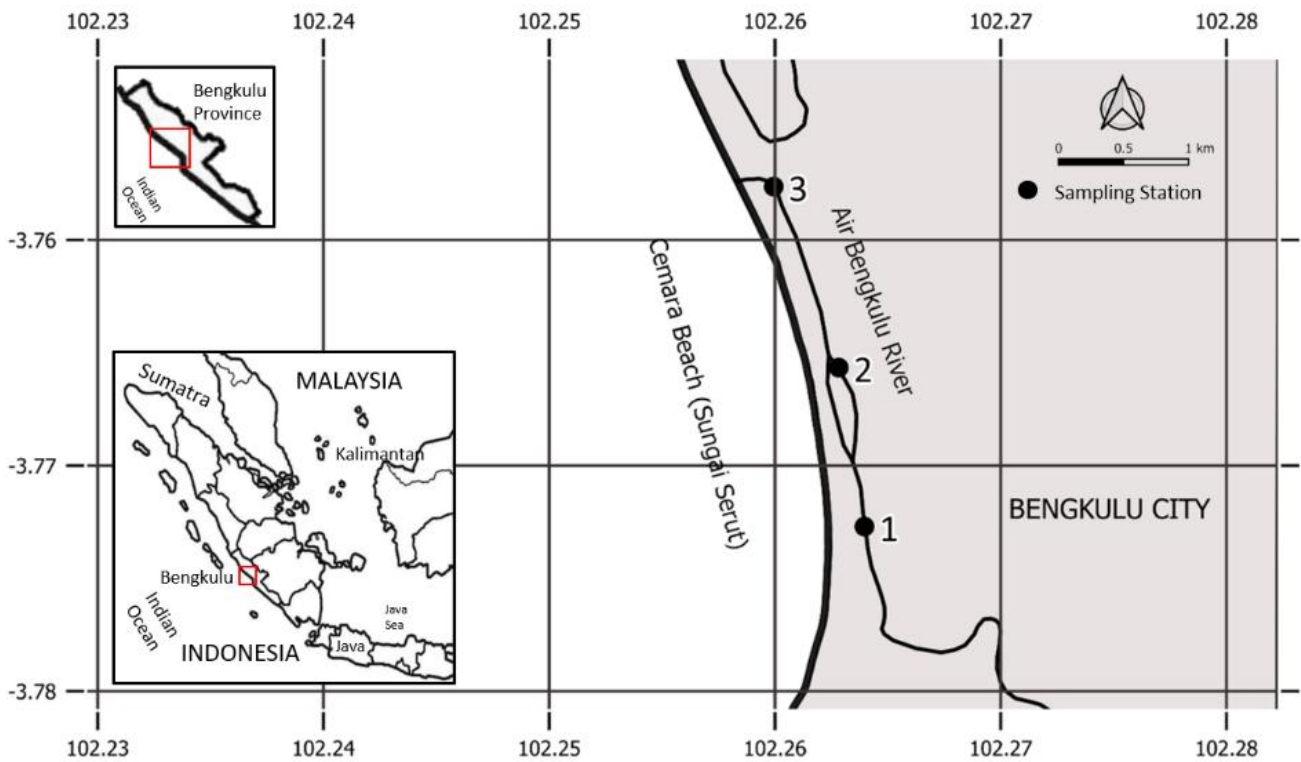


Figure 1. Location of the study area along the downstream of Air Bengkulu River, showing the sampling stations within approximately 1 km upstream from the river mouth.

Table 1. Geographic coordinates of sampling stations and schedule

Sampling Location	Coordinates	1 st Sampling (June)		2 nd Sampling (July)		3 rd Sampling (August)	
		Morning	Afternoon	Morning	Afternoon	Morning	Afternoon
Station 1	102.26348 -3.77175	09.20	15.00	09.24	14.47	09.20	14.47
Station 2	102.26338 -3.76578	11.10	16.10	10.47	15.24	10.49	15.28
Station 3	102.25924 -3.75657	12.50	17.00	11.54	17.00	11.19	16.18

2.2. Field Sampling Design

Sampling was conducted at three stations at depths of 0, 1, and 2 m, twice daily, with collections performed in the morning during high tide and in the afternoon during low tide. Sampling was carried out on three occasions in June, July, and August 2025, timed to coincide with spring tide conditions associated with the new moon phase. Tidal information was obtained from the WXTide32 application.

Phytoplankton samples were collected by filtering 60 L of water through a plankton net. Nitrate (mg/L) samples were obtained from the filtrate collected during phytoplankton sampling. Both sample types were transported to the laboratory under chilled conditions. Phytoplankton subsamples (100 mL) were then filtered and stored frozen.

Environmental parameters were measured in situ, including current velocity (m/s) using a current meter, salinity (‰) using a refractometer, dissolved oxygen (mg/L) using a DO meter, temperature (°C) using a thermometer, pH (-) using a pH meter, and water transparency (%) determined using a Secchi disk. Salinity, current velocity, dissolved oxygen, and nitrate were measured at all depths, whereas temperature, pH, and water transparency were recorded only at the surface.

2.3. Laboratory and Statistical Analysis

Trichromatic spectrophotometric analysis was used to determine the concentration of Chl-a, referring to EPA Method 446.0 Revision 1.2 (EPA, 1997). Absorbance was measured at three primary wavelengths (A664, A647, and A630) and values were corrected using absorbance at A750, as specified in the method. Chl-a concentration (µg/L) was calculated using the equation proposed by Jeffrey and Humphrey (1975), as referenced in EPA (1997) and Rey and Aminot (2002). Nitrate concentration was determined spectrophotometrically using the brucine method, following the technical guidelines from Balai Penelitian Tanah (2005).

Statistical analyses were conducted using the Kruskal–Wallis test, followed by a post hoc Mann–Whitney test, to evaluate spatial differences among parameters. Quantile regression was applied to assess the influence of environmental variables on the response variable. Chl-a data were log-transformed after adding a small constant to accommodate values below the detection limit. These non-detect values are reported as zero in the tables to reflect environmental conditions. Spearman's rank correlation was used to examine general relationships among the parameters. Statistical significance was determined at $p < 0.05$.

3. Results and Discussion

3.1. Environmental Parameter Heterogeneity

Environmental parameters were analyzed across stations at three depths at each station by comparing morning high-tide and afternoon low-tide conditions, as shown in Figure 2. Observed variations in current velocity suggest non-uniform vertical water mass movement, which influences key water-column processes such as nutrient transport, oxygen distribution, and salinity gradients. Higher dissolved oxygen concentrations observed during the afternoon are likely associated with enhanced photosynthetic activity under increased light availability. Depth-dependent differences in current velocity, however, may limit vertical mixing, leading to uneven oxygen distribution and increased heterogeneity. Similarly, increased nitrate heterogeneity during the afternoon reflects spatial variability in nutrient availability. Nitrate is transported by current from both marine and upstream river sources, while phytoplankton uptake occurs at different rates across depths, further amplifying concentration variability. In addition to river discharge, surface currents are influenced by diurnal tidal forcing and wind-driven circulation, whereas subsurface currents are largely influenced by riverbed morphology, which may contribute to vertical heterogeneity in environmental conditions.

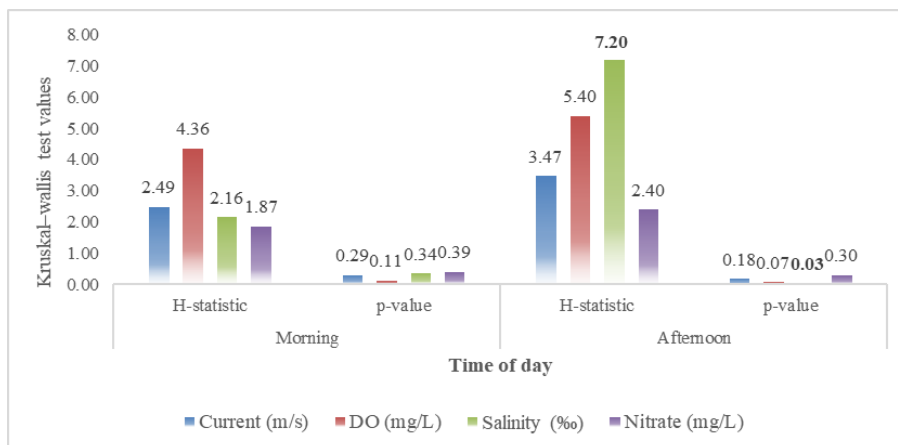


Figure 2. Depth-related variation of environmental parameters based on the Kruskal-Wallis test results, with a critical H value of 5.991 ($\alpha < 0.05$)

Although variability increased across depths during the afternoon, only salinity showed a statistically significant difference ($H = 7.20, p = 0.03$; Figure 2). With the exception of salinity, environmental conditions across the three water column layers were relatively homogeneous during the study period, suggesting that phytoplankton were exposed to broadly uniform physicochemical conditions both spatially and

temporally. Post hoc Mann–Whitney tests revealed significant differences in afternoon salinity among all depths, whereas Chl-a concentrations remained consistently low and showed no significant variations across depths (Figure 3). These results indicate that salinity was not significantly associated with depth-related changes in Chl-a during the observation period.

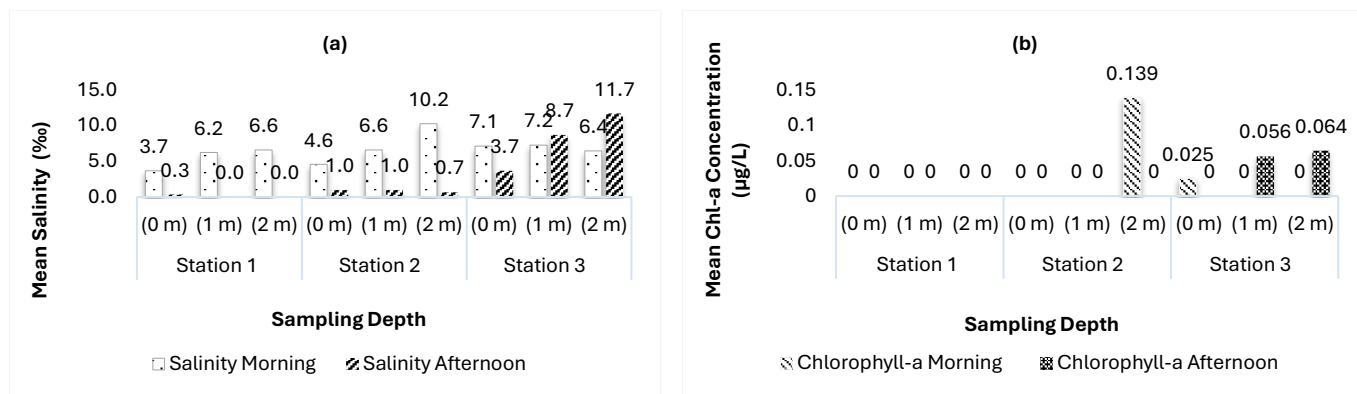


Figure 3. Comparison of average salinity (a) and Chl-a concentrations (b) between morning and afternoon observations across three depths at three stations.

Table 2. Overall mean nitrate concentrations and current velocities compared to Chl-a concentration* at three river depths across stations

Sampling Location	Depth (meter)	Morning			Afternoon		
		Nitrate (mg/L)	Current Velocity (m/sec)	Chl-a (µg/L)	Nitrate (mg/L)	Current Velocity (m/sec)	Chl-a (µg/L)
Station 1	0	0.359	0.147	0.000	0.245	0.143	0.000
	1	0.396	0.064	0.000	0.430	0.237	0.000
	2	0.677	0.166	0.000	0.248	0.209	0.000
Station 2	0	0.429	0.379	0.000	0.265	0.114	0.000
	1	0.419	0.316	0.000	0.266	0.232	0.000
	2	0.488	0.099	0.139	0.261	0.232	0.000
Station 3	0	0.420	0.164	0.025	0.294	0.120	0.000
	1	0.375	0.167	0.000	0.285	0.120	0.056
	2	0.381	0.396	0.000	0.284	0.086	0.064

*Values reported as zero indicate Chl-a concentration below the spectrophotometric detection limit

3.2. Diurnal Patterns of Nitrate and Current Velocity

During the study period, neither nitrate levels nor current velocity exhibited consistent patterns across the three depths, and the mean value range of both parameters was lower in the afternoon than in the morning (Figure 4). The highest nitrate concentration was recorded at a depth of 2 m at Station 1 on the first sampling (1.142 mg/L), while the lowest was at the surface of the same station on the third sampling (0.039 mg/L). The maximum and minimum current velocities were recorded at the surface of Station 2 on the second sampling (0.900 and 0.012 m/s, respectively). Both parameters generally exhibited lower values on the third sampling than on preceding sampling events.

Progressive decline occurred in current velocity from the first (0.170–0.422 m/s) and second (0.096–0.244 m/s) sampling to a weak and stable flow condition on the third sampling (0.060 m/s). This pattern indicates increased water column stability and a longer residence time following water inflow from the nearby Air Hitam River into the Air Bengkulu River via a small canal one day prior to the third sampling. Under low-current conditions, water masses from both rivers are likely to persist longer within the water column, allowing phytoplankton and organic materials to remain available for biological processes. These conditions coincided with the highest detection of Chl-a across all sampling points, consistent with findings that zones characterized by longer residence time tend to support greater phytoplankton accumulation (Zhang et

al., 2015; Stumpner et al., 2020; Steidle and Vennell, 2023). Such conditions may also facilitate the mixing of phytoplankton and light organic matter from the Air Hitam River into the Air Bengkulu River, potentially contributing to the slightly elevated productivity observed at Station 3. While Figure 4 depicts the diurnal fluctuations in nitrate and current velocity, Table 2 presents their overall mean values over the study period. During the morning and afternoon sampling, nitrate concentrations and current velocities did not exhibit consistent patterns at the three depths. Nitrate concentrations ranged from 0.359 to

0.677 mg/L in the morning and from 0.245 to 0.430 mg/L in the afternoon, while current velocities ranged from 0.064 to 0.396 m/s and from 0.086 to 0.237 m/s, respectively. Chl-a concentrations generally remained stable even at higher nitrate levels, indicating that lower current velocities had a slightly greater influence on phytoplankton growth than nitrate, and that the overall phytoplankton response was limited despite variations in these parameters.

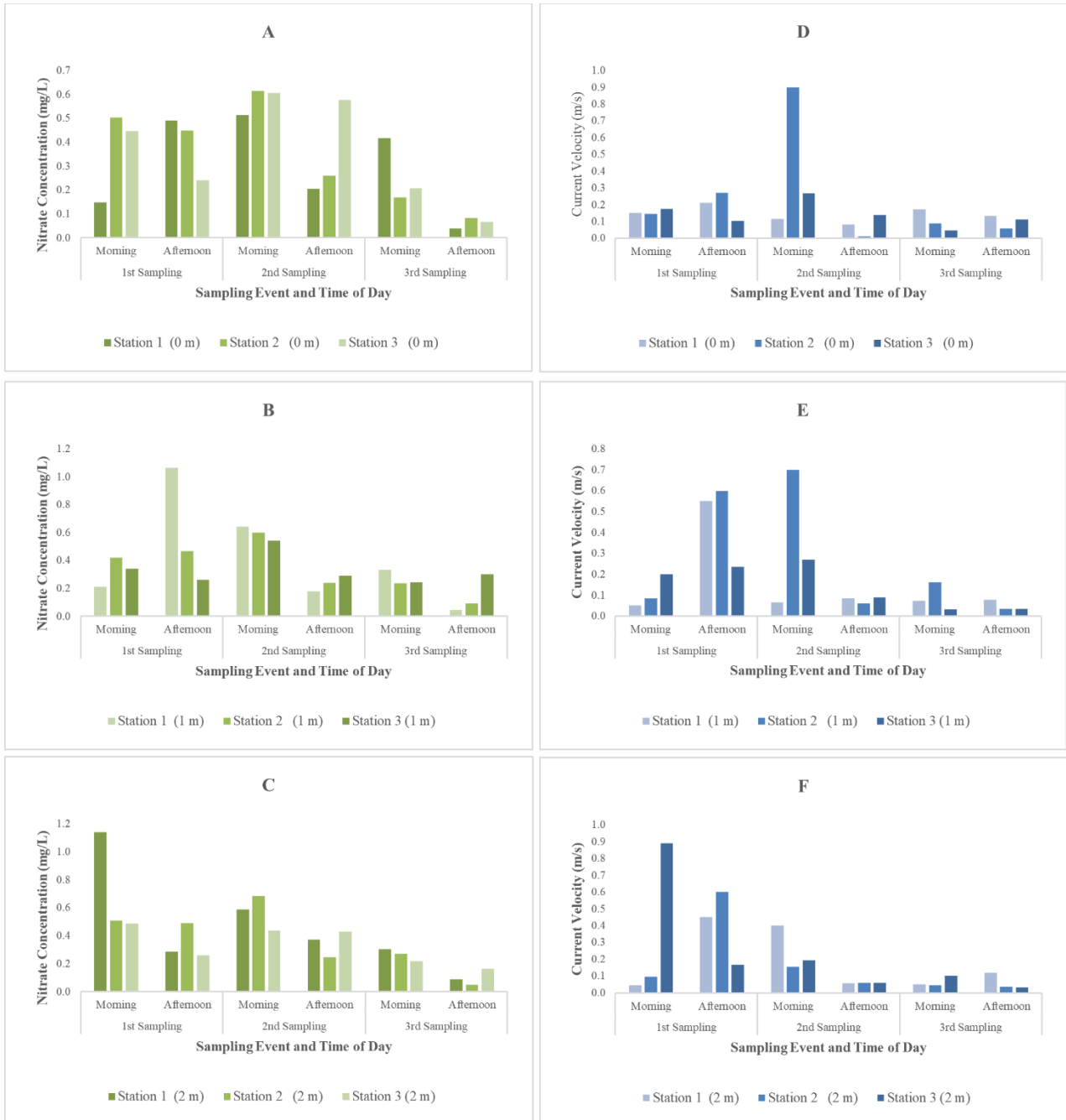


Figure 4. Dynamics of nitrate concentrations (A, B, C) and current velocity (D, E, F) at depths of 0 m (A, D), 1 m (B, E), and 2 m (C, F) which were collected twice daily (morning and afternoon) on three stations, over three sampling events

3.3. Chl-a Concentration

Spectrophotometric analysis revealed that the concentrations of Chl-a on all depth layers at the three stations were mostly below the detection limit and were therefore recorded as zero, as shown in Table 2. This low variation persisted under both high- and low-tide conditions. Only slight increases were observed at Station 2 at a depth of 2 m (0.139 µg/L) and at the surface of Station 3 (0.025 µg/L) during morning measurements. Similarly, slight increases were also observed at Station 3 at depths of 1 and 2 m (0.056–0.064 µg/L) during afternoon measurements. No significant increase was observed at any of the sampling points at Station 1. These limited variations suggest that Chl-a concentrations were generally constrained across stations and depths, likely associated with the influence of current velocity and nitrate, as indicated by the statistical analyses.

Quantile regression was performed using river depth, dissolved oxygen, current velocity, salinity, and nitrate as predictors of Chl-a at three quantiles ($\tau = 0.25, 0.50,$ and 0.75) representing low, median, and high concentrations, respectively. All collected data were included. The intercept coefficient remained stable across quantiles ($\beta = 0.544, 0.543,$ and 0.568), indicating that the model predicted baseline concentrations within a narrow range across all levels of distribution. River depth, dissolved oxygen, and salinity showed limited effects on Chl-a concentrations across the water column. Significant effects were observed only for current velocity at two quantiles and nitrate at one quantile: current velocity shifted from positive at the lower quantile to negative at the median and upper quantiles ($\beta = 0.170, -0.213,$ and -0.212), suggesting that it limits Chl-a at moderate-to-high concentrations; nitrate shifted from negative to positive ($\beta = -0.546, 0.000, 0.290$), indicating that nitrate limits Chl-a only at low concentrations.

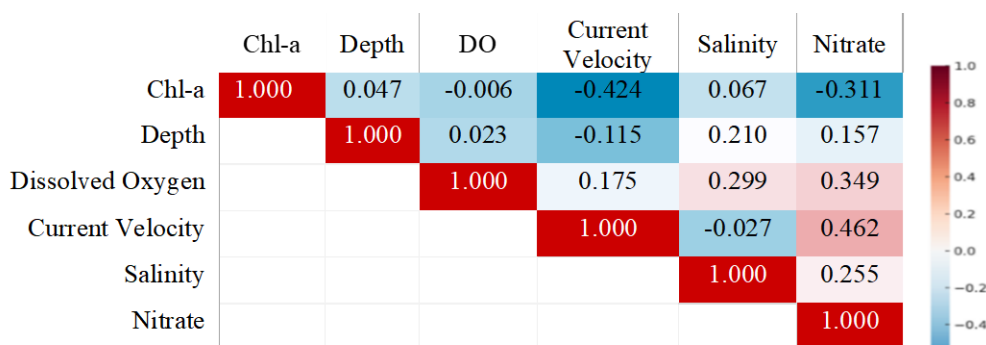


Figure 5. Spearman correlation among parameters; blue indicates negative correlations, red indicates positive correlations, and color intensity reflects the strength of the correlation.

Spearman's correlation analysis was conducted to assess the relationships between the parameters and the concentration of Chl-a, as well as the influence of river depth on all parameters (Figure 5). Chl-a exhibited moderate negative correlations with current velocity ($\rho = -0.424$) and nitrate ($\rho = -0.311$), which is consistent with the quantile regression results. Water column depth exhibited weak correlations with all other parameters, including Chl-a ($\rho = 0.047$), dissolved oxygen ($\rho = 0.023$), current velocity ($\rho = -0.115$), salinity ($\rho = 0.210$), and nitrate ($\rho = 0.157$), indicating that variations in depth had an insignificant effect on these parameters' conditions.

Observations indicate that most Chl-a concentrations at all depths across the three stations were below the detection limit under both high tide and low tide conditions. These results suggest that the area around the mouth of the Air Bengkulu River has very low primary productivity. Detected concentrations ranged from 0.025 to 0.139 µg/L (see Table 2), corresponding to the ultra-microtrophic to oligotrophic trophic classes, according to Gall and Pinkerton (2024). Nitrate and current velocity appear to be the main limiting factors for Chl-a concentrations across the three depths in the water

column. This finding aligns with the study by González-Ramírez et al. (2023), who reported that riverine nitrate supply and hydrodynamic factors are the primary drivers of chlorophyll variation in estuarine ecosystems. However, nitrate effectiveness is found to depend on current dynamics, and its high concentrations do not always result in enhanced Chl-a concentrations in estuaries, a pattern also observed by Mallin et al. (1993).

The negative correlation between nitrate and Chl-a at low concentrations likely reflects rapid phytoplankton uptake, as phytoplankton can remove nitrate and other dissolved nitrogen species from the water column at substantial rates during periods of high productivity, as reported by Torres-Valdes and Purdie (2006). This results in rapid nitrate assimilation and limited accumulation in the water column, indicating that nitrate may act as a limiting factor when the utilization of phytoplankton exceeds the available supply. Our observations of relatively low Chl-a concentrations are consistent with this pattern and likely reflect a limited phytoplankton population. Kennish, as cited by Rasyid et al. (2018), reported that the optimal nitrate range for phytoplankton growth is 0.9–3.5 mg/L, while concentrations below 0.44

mg/L may constrain growth. Furthermore, Rasyid et al. (2018) demonstrated that low nitrate concentrations in estuarine waters were associated with reduced phytoplankton abundance. Consistent with these findings, the observed nitrate concentrations in the Air Bengkulu River fall below this range and are insufficient to adequately support phytoplankton productivity, thereby contributing to low primary production.

3.4. Influence of Other Environmental Factors

We observed higher salinity heterogeneity in the afternoon samples, indicating that freshwater and seawater do not mix uniformly in the estuary of the Air Bengkulu River. Despite the estuarine waters being relatively shallow and lacking permanent stratification, interactions between seawater intrusion and freshwater flow can still generate salinity variations across depths. The intensity of tides, current velocity, and wind at specific times may enhance or weaken mixing in the estuary, particularly during low tide (Lupiola et al., 2025). This combination of factors likely explains why salinity variability was most pronounced among the measured parameters at different depths in the afternoon. Current velocities were higher during the morning high tide than during the afternoon low tide, resulting in reduced mixing and greater heterogeneity in the measured variables. Consequently, salinity differences across depths became more pronounced.

Salinity strongly influences algal metabolism, requiring cells to adapt to osmotic pressure while affecting the availability of nutrients, as well as the structure, distribution, and diversity of phytoplankton communities in freshwater and brackish estuaries (Orizar et al., 2024; Xia et al., 2024). Changes in salinity can limit the activity or induce the mortality of non-tolerant phytoplankton species (Cereja et al., 2021), enabling only tolerant species to survive. The low phytoplankton productivity observed in this study is likely the result of their small population in the Air Bengkulu River and selection by salinity. The highest tides in the morning bring higher salinity from the sea into the river, likely reducing the number of freshwater phytoplankton that could otherwise survive. Conversely, the lowest tides in the afternoon increase salinity dilution, further increasing salinity variability across the estuarine water column.

In addition to salinity-driven processes, other environmental parameters were also evaluated. Temperature, water transparency, and pH were measured at the surface layer. The pH values (6.7–7.7) indicated near-neutral to slightly alkaline conditions, which are generally favorable for most aquatic organisms, including phytoplankton, as they fall within the optimal range of 6.5 to 8.0 (EPA, 2026). Similarly, water temperature (26–32°C) was within or slightly above the optimal range for aquatic organisms in tropical waters (20–30°C; Effendi, 2003). However, statistical analysis revealed that none of these parameters had a significant relationship with Chl-a concentration, indicating that

they were not the primary factors controlling phytoplankton dynamics in the study area, particularly based on the surface measurements. Among these parameters, water transparency is further discussed due to its direct role in regulating light availability.

Water transparency is an important factor controlling primary productivity in estuarine systems, as limited light availability constrains phytoplankton physiology and growth in estuaries despite elevated nutrient levels (Xia et al., 2024). In this study, surface water transparency (0.8–45.4%) indicates heterogeneous light conditions, with lower values indicating potential light limitation in more turbid waters (Kirk, 2011). The waters appeared visually turbid throughout the study period, except at Station 3 near the estuary mouth during a rainfall event, when the highest water transparency value (45.4%) was recorded. This event coincided with a slight increase in afternoon Chl-a concentrations at 1 and 2 m (0.056 and 0.064 µg/L; Table 2). However, overall were dominated by low transparency values, suggesting restricted light availability for phytoplankton photosynthesis and consequently reduced primary productivity. Such conditions may be associated with salinity gradients and concurrent changes in light availability, as higher salinity in estuarine waters is often associated with increased turbidity and reduced light penetration, which together can limit phytoplankton photosynthetic activity, alter community dynamics, and render phytoplankton presence difficult to detect in estuarine environments (Cereja et al., 2021; Ye et al., 2025). A similar pattern was reported by Utami et al. (2025), where low Chl-a concentrations were associated with limited light availability. These conditions may also be further influenced by anthropogenic inputs from land-based activities, which can increase turbidity and reduce light penetration, potentially limiting phytoplankton productivity in the estuarine system.

4. Conclusion

The estuarine ecosystem of the Air Bengkulu River is shaped by complex spatial and temporal interactions among environmental factors. This study indicates that physical mixing processes and nutrient availability, particularly current velocity and nitrate concentration, are the important drivers of Chl-a variability, whereas the general water quality parameters are not the dominant limiting factors. The consistently low Chl-a concentrations indicate ultra-microtrophic to oligotrophic conditions and limited primary productivity, with implications for food web support and local capture fisheries. Anthropogenic influences affecting turbidity and nutrient dynamics may further constrain productivity. This study contributes to a better understanding of environmental controls on phytoplankton dynamics, in particular in the downstream section of the Air Bengkulu River and provides a basis for future investigations on river-sea interactions and water quality management. Future studies should include

observations during rainy and transitional seasons and broaden the temporal scope of the study to better capture seasonal variability, while also incorporating additional parameters not included in this study, such as turbidity, suspended solids, and other nutrients.

5. Data Availability Statement

All data included and used in the study are open and contain no confidential and ethical private information.

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7. Conflict of Interest

Each author has declared that there is no conflict of interest in the writing or submission of this manuscript.

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