

Effectiveness of Freshwater Mussel (*Pilsbryoconcha* exilis) as a Biofilter Medium in Improving Water Quality

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Abstract: This study aimed to evaluate the effectiveness of the freshwater mussel (Pilsbryoconcha exilis) as a natural biofilter in improving water quality in Dusun II, Bandar Klippa Village, Deli Serdang Regency. The research was conducted from August to October 2020 using a Completely Randomized Design (CRD) comprising four treatments based on biofiltration duration: 0 days (control), 5 days, 10 days, and 15 days, each with four replications. The observed parameters included physical characteristics (turbidity, odor, color, total dissolved solids/TDS) and chemical properties (pH, iron/Fe, manganese/Mn, nitrite/NO2⁻, and nitrate/NO₃⁻). Statistical significance was tested using ANOVA followed by DMRT. The results indicated that P. exilis significantly reduced turbidity and eliminated odor within 5 days of treatment. The pH also significantly decreased, although it remained within the safe threshold for clean water. Conversely, Fe and Mn concentrations did not change significantly, while nitrite and nitrate levels increased, presumably due to enhanced nitrification during filtration. Overall, P. exilis demonstrated potential as an effective biofilter for improving water's physical and chemical quality, with a 5-day filtration period identified as the optimal duration.

Introduction

Water is essential in supporting the continuity of biological systems and various life-related activities (1). Other compounds cannot substitute its function, as nearly all biological system processes depend on water availability, including cleaning processes, food preparation, and other domestic activities (2, 3). The rapid population growth in Indonesia has led to an increased demand for clean water (4). From a physiological perspective, living organisms have a lower water deficiency tolerance than nutrient deficiency (5, 6). Therefore, adequate quality and quantity of water availability are priorities when providing essential resources (7-9). Water quality is crucial in improving environmental health, as substandard water can be a vector for various waterborne diseases (10, 11).

Water quality standards are established to ensure safety and suitability for use. Clean water quality is assessed based on physical, chemical, and biological parameters. Physical parameters include odor, temperature, taste, color, turbidity, and total dissolved solids (TDS). Chemical parameters cover metals, toxic substances, reactive compounds, and pH. Biological parameters refer to microorganisms such as coliform bacteria (12, 13).

One method for improving water quality is the use of biofiltration systems. Biofilters function to reduce dissolved chemical compounds through adsorption processes and act mechanically to clarify water and biologically to neutralize toxic substances (14, 15). Aquatic organisms such as *P. exilis* (freshwater mussel), native to parts of Southeast Asia, including Indonesia, can be used as biofiltration agent (16). This species is a filter feeder capable of removing suspended particles and microorganisms through its internal circulation system. According to Prihartini 1999, the species demonstrates high survival rates and significant potential for application in the remediation of polluted water, including removing heavy metals (17). Additionally, it contributes to reducing excessive phytoplankton, thereby supporting the improvement of aquatic environmental quality (18).

However, despite its known filtration ability, limited research has quantified the effectiveness of *P. exilis* under controlled treatment durations or examined its specific impact on multiple water quality parameters in Indonesian freshwater systems. This study addresses that gap by evaluating the species' performance over different biofiltration periods, aiming to identify the optimal exposure time for practical use in local water treatment efforts.

Methodology Time and Location of the Study

This study was conducted from August to October 2020 in Dusun II, Bandar Klippa Village, with water sample analysis carried out at the Industrial Research and Standardization Center (Baristand) in Medan.

Materials and Equipment

Freshwater mussels were sourced from the local aquaculture in Deli Serdang, Indonesia. They were fed Nutrex Hawaii spirulina powder (USA). The filtration medium was river water from Dusun II, Bandar Klippa Village. All water analyses were conducted at Baristand Medan, Indonesia.

The study used 24-liter plastic buckets (local production, Indonesia) with aeration provided by a Resun ACO-001 air pump (Resun, China). Water sample weights were measured using a Camry EK5055 digital scale (Camry, China). Chemical analyses were performed with a Shimadzu UV-1800 spectrophotometer (Shimadzu, Japan), while pH and TDS were measured using Hanna HI98103 and HI98301 meters (Hanna Instruments, USA). Pyrex® glass measuring cylinders (Corning Inc., USA) ensured accurate volume measurements, and water samples were stored in food-grade Lion Star® jerry cans (Indonesia).

Research Methodology

This study employed an experimental approach using a Completely Randomized Design (CRD). Four treatment groups were tested, each replicated four times to ensure the validity.

The treatments were designed to evaluate the effectiveness of *P. exilis* as a biofilter for improving water quality. The first treatment (K0) was the control group without applying biofiltration and represented the initial condition (day 0). The second treatment (K1) involved a 5-day biofiltration period to assess short-term filtration period to evaluate medium-term impacts on water quality. The fourth treatment (K3) included a 15-day filtration period to determine the extended effectiveness of *P. exilis*. These four treatments were structured to identify the optimal exposure duration for maximum biofiltration efficacy. Data obtained from the experiment were analyzed using analysis of variance (ANOVA) based on the **Equation 1**.

$\mathbf{Y}_{\mathbf{ij}} = \mu + \tau_{\mathbf{i}} + \boldsymbol{\Sigma}_{\mathbf{ij}}$

Equation 1 | Y_{ij} = water quality resulting from the treatment, μ = overall mean, τ_i = effect of the i-th mussel treatment, Σ_{ij} = experimental error of the i-th mussel treatment at the j-th replication.

Measured Parameters

The parameters measured in this study include water quality based on both chemical and physical properties, which encompass pH, Iron (Fe), Manganese (Mn), Nitrate (NO_3^-), Nitrite (NO_2^-), Turbidity, Odor, and Color.

The study used four treatment containers filled with 20 liters of river water. Freshwater mussels were introduced at a stocking density of 0.5 kg per 20 liters of water (1:40 ratio). The mussels were maintained under continuous aeration for 15 days and fed spirulina powder every two days at a consistent rate to support filtration activity.

The dimensions of the medium were standardized across treatments, and 20 liters of water were used in identical containers to ensure consistency. Treatments consisted of four biofiltration durations: 0 days (control), 5 days, 10 days, and 15 days. Each treatment was replicated four times for statistical validity.

Water sampling was conducted at four time points:

Order	Day of Treatment	Mean
1	0	37.75°
2	15	3.50 ^b
3	10	3.25⁵
4	5	2.75 ^b

Note: Numbers followed by the same letter in the column indicate no significant difference according to Duncan's multiple range test at $\alpha = 0.05$.

baseline (before mussel introduction), and on days 5, 10, and 15 after treatment began. At each interval, 2 liters of water were collected from each container for laboratory analysis. Samples were handled and transported using clean, foodgrade containers to prevent contamination.

Measured parameters included pH, turbidity, odor, color, total dissolved solids (TDS), iron (Fe), manganese (Mn), nitrite (NO_2^-), and nitrate (NO_3^-). Physical parameters (turbidity, odor, color) were assessed immediately, while chemical parameters were analyzed at the Medan Industrial Research and Standardization Center (Baristand Medan).

Data analysis was performed using analysis of variance (ANOVA) to determine the effect of treatment on water quality. Where significant differences were found, Duncan's Multiple Range Test (DMRT) was applied at a 95% confidence level to identify specific treatment differences.

Results and Discussion

Physical Parameters

Turbidity Analysis

The observational data and results of the analysis of variance (ANOVA) for the turbidity parameter are presented in Appendix 1. The mean turbidity values for each treatment group are summarized in **Table 1**.

The study's results indicate that the 0-day treatment differed significantly from the 5-day, 10-day, and 15-day treatments. The highest mean turbidity value was observed in the 0-day treatment, reaching 37.75 NTU, while the lowest was found in the 5-day treatment, which recorded 2.75 NTU. This suggests that the 5-day treatment was the most effective in reducing water turbidity. Turbidity is an optical property of a liquid that affects the transmission of light through the water due to absorption and scattering processes (19). Turbid water contains numerous suspended particles that alter its visual appearance, including color and clarity. The degree of turbidity varies: clear water exhibits low turbidity, while opaque water indicates a high turbidity level (20).

Water Color

The observational data and analysis of variance (ANOVA) for water color parameters are presented in Appendix 2. The results indicate that treatments using *P. exilis* from day 0 to day 15 did not significantly affect water color. Thus, while *P. exilis* may contribute to improving water quality, but it does not appear to alter the color of the water.

Total Dissolved Solids

The observational data and analysis of variance (ANOVA) for Total Dissolved Solids (TDS) are provided in Appendix 3. The mean TDS values are summarized in **Table 2**. This study's

Table 2. Mean total	dissolved solids measured on days	0, 5,
10, and 15 following	treatment.	

Order	Day	Mean (mg/L)
1	15	162.75°
2	10	141.25 ^{ab}
3	5	122.5 ^{bc}
4	0	111.5 ^c

Note: Numbers followed by the same letter in the column indicate no significant difference according to Duncan's multiple range test at $\alpha = 0.05$.

results indicated no significant differences between the treatments applied on days 0, 5, 10, and 15 regarding Total Dissolved Solids (TDS) levels in water. However, the highest average TDS value was recorded on day 15 at 162.75 mg/l, while the lowest was observed on day 0 at 111.5 mg/l. Although TDS levels slightly increased after treatment, the 5-day period resulted in the lowest rise among the filtered groups and remained well within acceptable drinking water standards. The goal was not to increase TDS, but rather to evaluate whether biofiltration adversely affected it.

Total Dissolved Solids (TDS) refers to dissolved substances with particle sizes smaller than 10⁶ mm (dissolved) and colloidal particles ranging from 10⁻⁶ to 10⁻³ mm. These consist of various chemical compounds and other substances that cannot be filtered through a 0.45 μ m filter paper (21). TDS typically includes inorganic salts such as calcium, magnesium, sodium, potassium, carbonate, bicarbonate, chloride, sulfate, and nitrate.

The presence of TDS in water can affect its taste. Water quality panels often evaluate the taste of water based on its TDS level. According to established standards, water with a TDS level of less than 300 mg/l is considered excellent, 300 to 600 mg/l is good, 600 to 900 mg/l is fair or drinkable, 900 to 1200 mg/l is poor, and above 1200 mg/l is very poor (22). Water with very low TDS levels is also often considered less acceptable due to its flat taste and less nutrition for the biota (13, 23). Therefore, while water with low TDS may be considered safe for consumption, its taste might be inadequate for long-term use. Thus, while an increase in TDS can influence the taste quality of water, it is crucial to consider the balance between water quality and consumer acceptance of its taste (23).

Water Odor

The results of the water odor test are presented in **Table 3**. The analysis throughout the study revealed a significant change in water odor, transitioning from a detectable smell to odorless. On day 0, the water was found to have an odor, but after treatments of 5, 10, and 15 days, the water became odor-free. This suggests that *P. exilis* is an effective biofilter for eliminating odors in water, thus improving its quality and

making it more suitable for use. Regarding odor elimination, the mussels likely reduced odor-causing organic compounds by consuming suspended algae and bacteria, often responsible for foul smells in stagnant water (24, 25). The complete removal of odor after 5 days indicates efficient microbial and organic load reduction.

Chemical Parameters

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The observational data and analysis of variance (ANOVA) for pH values are presented in Appendix 4. The mean pH values are shown in **Table 4**. There is a significant difference compared to the 5-day, 10-day, and 15-day treatments. However, there was no significant difference among the 5-, 10-, and 15-day treatments, indicating that treatment duration did not cause a differential effect on pH reduction. The highest mean pH value was recorded in the 0-day treatment at 8.70, while the lowest was observed in the 15-day treatment at 8.05. The 5-day treatment was the most effective in reducing pH levels.

Fluctuations in pH values were influenced by the feeding activity involving *Spirulina sp.* powder and the biological activity of freshwater mussels that release feces during the filtration process, cumulatively affecting water pH (26, 27). According to Surbakti E. P. *et al* (2021), a decrease in pH can enhance the toxicity of heavy metals in water (28). Based on the data in the table, the observed pH values remain within the acceptable range, which is between 6.5 and 8.5 (29).

Iron (Fe) Content

The observational data and the results of the analysis of variance (ANOVA) on iron content are presented in Appendix 5. Based on the statistical analysis, water treatment using *P. exilis* from day 0 to day 15 did not significantly differ in iron levels. Therefore, improving water quality from unfit to potable using *P. exilis* did not significantly affect the iron concentration in the water.

Iron (Fe) plays a crucial role in the formation of hemoglobin (30). Iron is regulated through the absorption phase, as the body lacks a specific excretory mechanism for this metal. As a result, individuals who frequently undergo blood transfusions may experience iron accumulation, which can cause darkening of the skin (31). Drinking water with elevated iron levels can induce nausea (32). Although Fe is essential, excessive intake can damage the intestinal lining, and in severe cases, may lead to fatal outcomes. Iron concentrations above 1 mg/L may irritate the eyes and skin, while levels exceeding 10 mg/L can give the water a foul odor like rotten eggs (33).

Manganese (Mn) Content

The observational data and analysis of variance (ANOVA) results for manganese content are presented in Appendix 6. Statistical analysis revealed that water treatment using *P*.

Table 3. Water odor tested on days 0, 5, 10, and 15 following treatment.

Day	Replication				Maan
	II	II	III	IV	- Mean
day 0	Odorous	Odorous	Odorous	Odorous	Odorous
Day 5	Odorless	Odorless	Odorless	Odorless	Odorless
Day 10	Odorless	Odorless	Odorless	Odorless	Odorless
Day 15	Odorless	Odorless	Odorless	Odorless	Odorless

Table 4. Mean pH value measured on days 0, 5, 10, and 15following treatment.

Order	Day	Mean
1	0	8.70ª
2	5	8.35⁵
3	10	8.15 ^b
4	15	8.05⁵

Note: Numbers followed by the same letter in the column indicate no significant difference according to Duncan's multiple range test at $\alpha = 0.05$.

Table 5. Mean nitrite and nitrate contents measured on days 0, 5, 10, and 15 following treatment.

Order	Day	Mean (mg/L)		
		Nitrite	Nitrate	
1	10	0.8250ª	38.575°	
2	15	0.6325 ^{ab}	20.620 ^{ab}	
3	5	0.4075 ^b	13.688 ^b	
4	0	0.0092 ^c	0.316 ^c	
Note: Numbers followed by the same letter in the column				

indicate no significant difference according to Duncan's multiple range test at $\alpha = 0.05$.

exilis from day 0 to day 15 did not significantly differ in manganese levels. Therefore, the application of *P. exilis* to improve water quality from nonpotable to potable did not significantly affect manganese concentration.

At low concentrations (<0.5 mg/L), manganese (Mn) in water poses no health risks. It may even offer benefits such as supporting brain and bone health, promoting hair and nail growth, and aiding in enzyme production for carbohydrate and protein metabolism into energy. However, manganese becomes neurotoxic at higher concentrations (>0.5 mg/L) (34). Symptoms of exposure include nervous system disorders, insomnia, muscle weakness in the legs and face, and a frozen facial expression resembling a mask (35).

Nitrite (NO₂) Content

The results of the observations and analysis of variance for nitrite content are presented in Appendix 7. The average nitrite content values are displayed in **Table 5**. The research findings revealed that the 10-day treatment did not differ significantly from the 15-day treatment, and likewise, no significant difference was observed between the 15-day and 5-day treatments. The highest average nitrite concentration was recorded on day 10 at 0.852 mg/L, while the lowest concentration was observed on day 0 at 0.00925 mg/L. The application of *P. exilis* as a biofiltration agent in this study was ineffective in reducing nitrite levels. Instead, it increased nitrite concentration, exceeding the maximum limit set by the Indonesian Ministry of Health Regulation No. 32 of 2017, 1 mg/L.

Naturally, nitrite (NO₂⁻) concentrations in aquatic environments are relatively low due to its instability in the presence of oxygen, where it rapidly oxidizes to nitrate. Nitrite typically forms as an intermediate compound during the oxidation of ammonia to nitrate. The nitrification process occurs in two primary stages, each catalyzed by different types of bacteria. The first stage involves the oxidation of ammonium (NH_4^+) to nitrite (NO_2^-) by *Nitrosomonas*, following the reaction:

 $NH_{4^+} + 2O_2 \rightarrow NO_2^- + 2H_2O$ (by Nitrosomonas)

Subsequently, the nitrite produced is oxidized into nitrate (NO $_3^-$) by *Nitrobacter*, as shown in the reaction:

 $2NO_2^- + O_2 \rightarrow 2NO_3^-$ (by Nitrobacter)

This process plays a crucial role in the nitrogen cycle and is significantly influenced by oxygen availability and microbial activity.

Nitrate (NO₃) Concentration

The observational data and analysis of variance for nitrate content are presented in Appendix 8. The mean nitrate concentrations are shown in **Table 5**. Nitrate is an intermediate product in the nitrogen cycle, resulting from the transforming of nitrogen compounds. Nitrite is generated through bacterial activity starting from ammonia and organic nitrogen. These nitrogenous compounds are commonly found in wastewater, groundwater, and agricultural runoff (<u>36</u>). Nitrification refers to the biological oxidation process where ammonium is first converted into nitrite, and then nitrite is oxidized into nitrate. This process occurs naturally in soil, freshwater, and marine ecosystems, driven by specialized nitrifying bacteria. Several environmental conditions highly influence the rate of nitrification.

Nitrifying bacteria are particularly sensitive to temperature changes (37). Although abrupt temperature shifts do not directly affect bacterial growth rates, temperature remains a key factor in the efficiency of nitrification. The optimal temperature range for nitrification is between 0-20°C, within which bacterial activity peaks, leading to faster nitrification (37). Oxygen concentration is another crucial factor, as these bacteria are aerobic and require oxygen for survival (38). In addition, the pH of the environment significantly surrounding impacts the nitrification rate, with the most rapid reaction occurring at pH levels between 7.5 and 7 (39, 40). These factors collectively determine the viability and activity of nitrifying bacteria, thereby influencing the overall nitrification process.

Conclusion

Based on the results of this study, *P. exilis* demonstrated significant effectiveness as a biofilter for improving both the physical (turbidity, odor) and chemical (pH) quality of water. Among the tested durations, a five-day exposure period provided the best overall performance, making it the optimal timeframe for biofiltration. This suggests that *P. exilis* has strong potential for application in low-cost, community-based water treatment systems, especially in rural or peri-urban areas. Future research should examine long-term impacts, scalability, and how *P. exilis* interacts with other organic compounds or emerging contaminants.

Declarations

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Conflict of Interest

The authors declare no conflicting interest.

Data Availability

The unpublished data is available upon request to the corresponding author.

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Not applicable.

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Additional Information

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