

Type of the Paper (Original Research)

The Effectiveness of *Pseudomonas aeruginosa* in Degrading Bio-chemical Oxygen Demand and Chemical Oxygen Demand in Small Island Domestic Wastewater

Nurdianti¹, Agus Bintara Birawida^{1†}, Hasnawati Amqam¹, Gurendro Putro² and Shaharuddin Mohd Sham³

¹Department of Environmental Health, Faculty of Public Health, Hasanuddin University, Makassar, Indonesia

²Research Center for Public Health and Nutrition, Nasional Research and Innovation Agency, Jakarta, Indonesia

³Department of Environmental and Occupational Health, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Selangor, Malaysia

†Corresponding author: Agus Bintara Birawida; agusbirawida@unhas.ac.id

ORCID IDs: <https://orcid.org/0009-0004-5659-4466>, <https://orcid.org/0000-0002-8223-2387>, <https://orcid.org/0000-0003-4961-8517>, <https://orcid.org/0000-0002-2718-8619>, <https://orcid.org/0000-0002-3603-0821>

Key Words	Bioremediation, Bioaugmentation, Batch reactor, Domestic wastewater
DOI	https://doi.org/10.46488/NEPT.2026.v25i02.D1843 (DOI will be active only after the final publication of the paper)
Citation for the Paper	Nurdianti, A.B., Amqam, H., Putro, G. and Mohd Sham, S., 2026. The effectiveness of <i>Pseudomonas aeruginosa</i> in degrading bio-chemical oxygen demand and chemical oxygen demand in small island domestic wastewater. <i>Nature Environment and Pollution Technology</i> , 25(2), D1843. https://doi.org/10.46488/NEPT.2026.v25i02.D1843

ABSTRACT

The high organic content of domestic wastewater on small islands increases the risk of environmental pollution and the spread of disease. Limited resources and infrastructure exacerbate waste management challenges in these areas. This study aims to identify local bacteria from domestic wastewater and test the effectiveness of *Pseudomonas aeruginosa* in degrading BOD and COD. This study was conducted in the Spermonde Islands, Makassar City. Wastewater samples were collected from six locations and analyzed using the MALDI-TOF MS test. Effectiveness tests were carried out in a simple 5 L ex situ bioreactor using a completely randomized design (CRD) with two factors: bacterial inoculum concentration (0%, 5%, 10%, 15%) and incubation time (2, 4, 6, 8 days). *Pseudomonas aeruginosa* effectively reduced BOD by 64 - 77% and COD by 63 - 77%. Variations in inoculum concentration did not significantly affect BOD and COD reduction ($p > 0.05$), while incubation time had a significant effect ($p < 0.05$). The highest removal efficiency occurred on day 6. These findings suggest that optimizing incubation time is more critical than increasing inoculum concentration for successful bioremediation. *Pseudomonas aeruginosa* is a practical and economical option to support sustainable wastewater management on small islands and can be integrated into household-scale communal wastewater treatment plants (WWTPs) to improve treatment effectiveness.

INTRODUCTION

Small island have a wide range of define problems (Khotimah et al., 2021; Singh et al., 2022). Ecologically, these islands are fragile and vulnerable due to limited land area, scarce resources, restricted distribution, and high isolation, which pace them at high risk (Kumar and Mishra, 2024). These factors increase island communities' vulnerability to disease and mortality, often linked to inadequate sanitation (Birawida et al., 2020).

Population growth increases environmental pressure. Domestic wastewater, unlike solid waste that is often managed, is frequently discharged directly into waterways and can pollute coastal and groundwater environments (Harahap et al., 2021). Rivers, lakes, and ponds are the most vital and vulnerable freshwater systems, playing a crucial role in sustaining all forms of life (Kumar and Mishra, 2024).

Wastewater commonly contains excreta and greywater from kitchens and bathrooms and is rich in organic material (Buslima et al., 2024; Rahma and Purwanti, 2020). Millions of intestinal bacteria and a small number of other organisms are found in domestic waste, posing a danger to residents. Laundry wastewater, which is rich in detergents, phosphates, and nitrates, produces foam and is harmful to aquatic organisms in freshwater ecosystems through the process of eutrophication. Domestic wastewater production accounts for 70 - 75% of total waste production. Domestic wastewater has a large volume, contributing to water body pollution by approximately 70 - 80% (Khotimah et al., 2021).

Domestic wastewater is a significant problem in Indonesia, especially in the archipelago, which is home to around 60% of the country's 250 million people. Population growth has led to increased waste production, which has impacted environmental quality. Most communities still use rivers, seas, and beaches as disposal sites (Prastiwi and Rosariawari, 2023). Therefore, treatment is needed to ensure that domestic wastewater can be safely discharged into the environment. Conventional biological treatment methods can meet emission standards but have limitations such as high costs, large land area requirements, complexity, high energy needs, and costly maintenance (Bai et al., 2024; Rafiaee et al., 2020; Xiao et al., 2021).

Simpler and lower-cost alternatives are therefore needed. One promising approach is bioremediation using local microorganisms to degrade pollutants to environmentally safe concentrations (Vasdravanidis et al., 2022). Bioremediation is inexpensive and uses natural microbial activity without causing significant side effects in the treatment process (Shah and Shah, 2020).

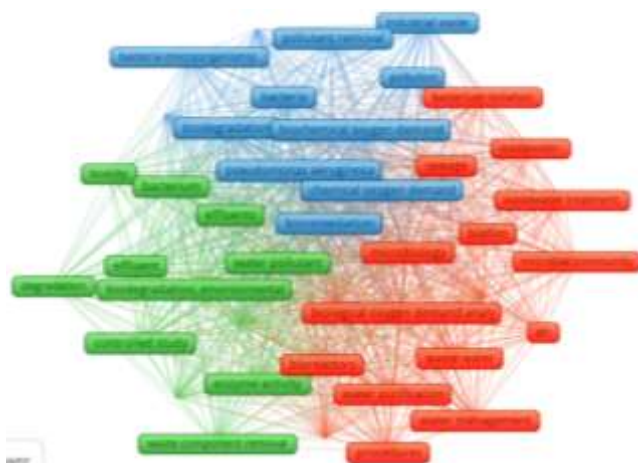


Fig. 1: Keyword Network VOSviewer Analysis Wastewater Bioremediation

Figure 1 shows a map of the relationships between keywords from VOSviewer related to wastewater treatment, bioremediation, and microbiology. Keywords are grouped by category with different colors: blue highlights the analysis of waste characteristics and the degradation process by microorganisms, green focuses on the impact of waste and the potential of microorganisms, while red covers technologies to improve treatment efficiency.



Fig. 2: Global Distribution of Wastewater Bioremediation Research by Bacteria

Figure 2 shows the distribution of research by country, indicating that waste treatment and environmental technology issues are a global concern with varying priorities. In line with the keyword map in Figure 2, research on wastewater bioremediation using BOD and COD degrading bacteria has attracted widespread attention. One of the potential bacteria that has been extensively studied is *Pseudomonas aeruginosa*.

This bacterium has the ability to degrade various pollutants found in domestic wastewater. *Pseudomonas aeruginosa* utilizes various carbon and energy sources and has an efficient enzymatic system for degrading pollutants (Lalucat et al., 2020). The environmental characteristics of small islands are marked by limited waste treatment infrastructure. Although there are communal wastewater treatment plants (WWTPs) on a household scale, these facilities are not operational or are not utilized optimally. This condition, combined with the vulnerability of water quality to pollution and incidents of seawater intrusion that increase salinity, results in a dynamic environmental system that requires biological agents with high adaptability.

Pseudomonas aeruginosa is a Gram-negative bacterium with broad metabolic flexibility; it can metabolize under both aerobic and anaerobic conditions and tolerates variations in nutrient availability and salinity. In addition to these adaptive abilities, *Pseudomonas aeruginosa* has the capacity for enzymatic degradation of various complex organic compounds, including components of domestic organic matter, thereby potentially reducing measurable organic loads such as Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD) in domestic wastewater. The combination of these physiological properties and degradation capabilities makes *Pseudomonas aeruginosa* a relevant candidate in bioremediation strategies for wastewater treatment systems that are prone to operational disruptions and the effects of seawater intrusion.

Previous studies have focused on the use of degrading bacteria to treat industrial, textile, and heavy metal-contaminated waste. However, the potential of *Pseudomonas aeruginosa* in degrading domestic waste, especially in small island ecosystems, has not been widely explored. Therefore, this study aims to make a new contribution

by analyzing the effectiveness of *Pseudomonas aeruginosa* in reducing BOD and COD levels in domestic waste on Small Island.

2. MATERIALS AND METHODS

2.1 Research Location

This research was conducted in the Spermonde Islands, Makassar City, South Sulawesi from August to November 2024. The research focused on domestic wastewater bioremediation efforts using local bacteria. Samples were collected by grab sampling at six observation points selected for their propensity to accumulate domestic waste, limited treatment facilities, and coastal ecosystem vulnerability.

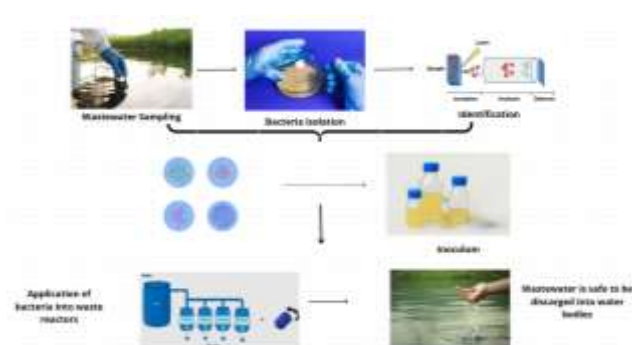


Fig.3: Research Illustration

2.2 Isolation and Identification of Bacteria

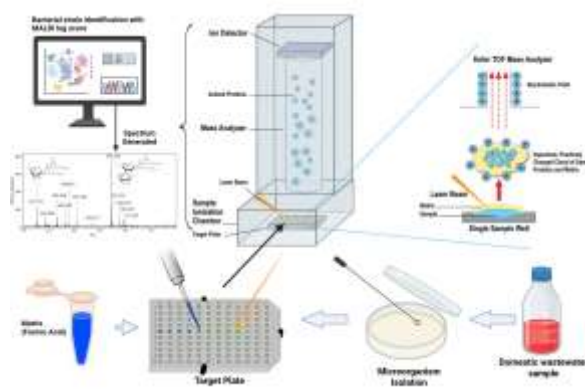


Fig.4: Illustration of the bacterial identification process using MALDI-TOF MS

The equipment used includes filter/filtration sets, membrane paper, TPC, incubators, water baths, tweezers, cotton, Bunsen burners/spirit lamps, test tube racks, Petri dishes, sample bottles, tissues, markers, lighters, paper labels, VITEK MS machines, and VITEK MS test slides. The materials used included bacterial samples, 70% alcohol, sterile water, masks, gloves, 1 μ L tips, VITEK MS-CHCA matrix, nutrient agar (NA), and nutrient broth (NB). Bacterial isolation was performed by filtering 100 mL of domestic wastewater using a 0.45 μ m filter membrane, then the membrane paper was planted on Cn agar medium and incubated at 35°C for 24 hours. Bacterial identification was performed using MALDI-TOF Mass Spectrometry, by applying bacterial colonies to a target

slide coated with VITEK MS-CHCA (α -cyano-4-hydroxycinnamic acid) matrix to facilitate ionization. After drying and crystallization, the slides were analyzed using VITEK MS integrated with the analysis software (Izzati, 2018).

2.3 Culture and Experimental Design

Pseudomonas aeruginosa isolates previously obtained from domestic wastewater were cultured on Nutrient Agar (NA) for 48 h at room temperature. Six colonies were transferred into 200 mL Nutrient Broth (NB) and incubated at 110 rpm for 24 h. Inoculum concentrations were prepared as 5% (2.5×10^3 CFU), 10% (5×10^3 CFU), and 15% (7.5×10^3 CFU). The experiment used a completely randomized design (CRD) with two factors: inoculum percentage (0%, 5%, 10%, 15%) and incubation time (2, 4, 6, 8 days), each with three replicates (Khadijah et al., 2023; Rahardja et al., 2010). The ex situ bioreactor consisted of four 5-L plastic reactors connected in series; reactor A served as the control (0% inoculum), reactors B–D received 5%, 10%, and 15% inoculum, respectively.

2.4 Physicochemical Analysis

pH was measured in situ with a pH meter and temperature with a thermometer. BOD was measured by the Winkler titrimetric method (SNI 06-6989.72-2009). COD was determined by the dichromate reflux titrimetric method (SNI 6989.2:2019).

2.5 Data Analysis

Data were analyzed using GraphPad Prism 10 software while statistical analysis used the One - Way Anova test.

3. RESULTS

3.1. Identification of Bacteria



Fig. 5: Distribution of organic pollutant degrading bacteria

Based on the identification results visualized on the spatial map in Figure 4, MALDI-TOF MS identification found diverse bacteria in six zones, including *Acinetobacter iwoffii*, *Acinetobacter haemolyticus*, *Enterobacter aerogenes*, *Proteus mirabilis*, *Escherichia coli*, and *Bacillus sp.*, *Pseudomonas sp.* was also found in Zone VI.

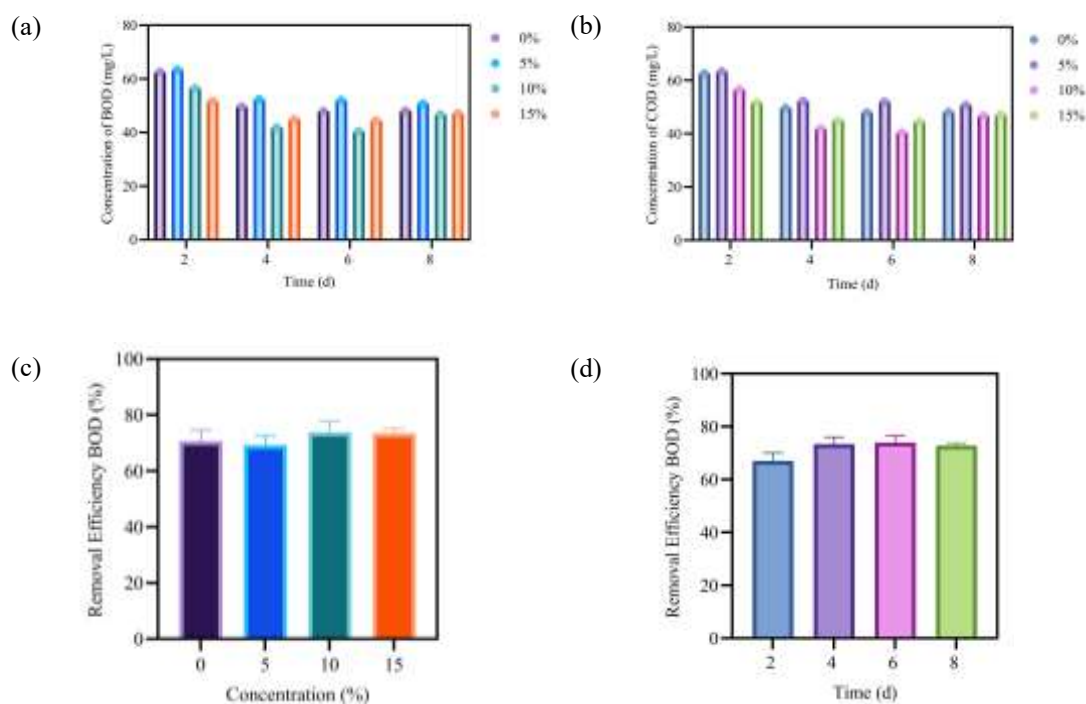
3.2. Effectiveness of Bioremediation

pH and temperature remained stable during the experiment (Table 1), providing suitable conditions for microbial activity.

Table 1. pH and Temperature Values

Time (days)	pH	Temperature (°C)
2	7.74	28.6
4	7.73	28.7
6	7.73	28.8
8	7.73	27.8
Average	7.7	28.4

Table 1 shows the results of pH and temperature measurements over 8 days at 2 day intervals. The pH value was stable in the range of 7.73 - 7.74 (average 7.7), while the temperature ranged from 27.8 to 28.8°C (average 28.4°C), indicating constant conditions suitable for microbial activity.



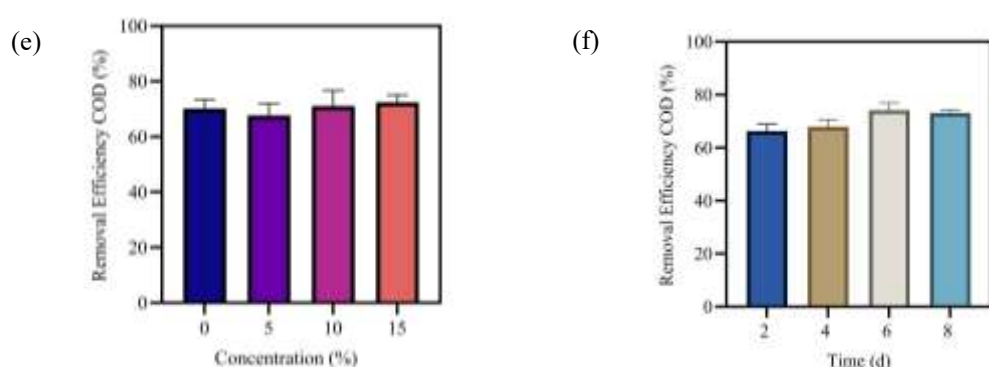


Fig. 6: (a)(b) BOD and COD concentrations, (c)(d)(e) (e) BOD and COD efficiency

Figure 6(a) show BOD and COD concentration trends. The greatest decreases in BOD and COD were observed with 10% inoculum at day 6 from an initial concentration of 177.5 mg/L to 40.50 mg/L. Conversely, the smallest decrease in BOD occurred at a bacterial concentration of 5% on day 2, with a value of 63.40 mg/L. Figure 6(b) shows a similar pattern for COD, with the largest decrease in COD occurring at a bacterial concentration of 10% on day 6, namely from 449.32 mg/L to 101.35 mg/L. Conversely, the smallest decrease in COD occurred at a bacterial concentration of 5% on day 2, namely 164.31 mg/L. BOD removal efficiency across inoculum concentrations ranged from 64.73% to 77.18%; one-way ANOVA showed no significant differences between inoculum concentrations ($p > 0.05$). COD removal efficiency ranged from 63.43% to 77.44%, likewise with no significant differences between inoculum concentrations ($p > 0.05$). For both BOD and COD, removal efficiencies increased from day 2 to day 6 and decreased slightly by day 8; one-way ANOVA showed incubation time had a significant effect ($p < 0.05$), with highest efficiencies observed on day 6.

4. DISCUSSION

4.1 Bacterial Identification

Domestic wastewater obtained from Spermonde Island was then subjected to laboratory testing to identify the bacteria present. Results showed, several types of bacteria in the wastewater samples. *Escherichia coli* and *Bacillus sp.* were found in zones I and II, while *Acinetobacter iwoffii* and *Bacillus sp.* were found in zone III, *Acinetobacter haemolyticus* and *Enterobacter aerogenes* in zone IV, *Proteus mirabilis* and *Bacillus sp.* in zone V, and *Pseudomonas sp.* and *Bacillus sp.* in zone VI. These bacteria belong to the group of gram negative bacteria except *Bacillus sp.*, which is a gram positive bacteria. In this study, further analysis of the lokal isolate from zone VI in Figure 5 showed that the strain was *Pseudomonas aeruginosa*.

Pseudomonas aeruginosa is a bioremediation agent due to its superior physiological and biochemical capabilities, including its ability to metabolize various complex organic compounds aerobically and anaerobically. This bacterium is also known to have high tolerance to extreme environmental conditions and rapid adaptability to new habitats, making it a potential candidate for environmental biotechnology applications. In the context of small islands with limited domestic wastewater management systems and high potential for environmental pollution due to population density, the use of *Pseudomonas aeruginosa* offers an efficient and sustainable biological solution to significantly reduce BOD and COD levels. *Pseudomonas aeruginosa* is effective in reducing pollutants in domestic wastewater, specifically BOD by 70 - 85% and COD by 65 - 80%. *Pseudomonas aeruginosa* possesses enzymes that effectively break down various organic compounds responsible for high BOD and COD levels (Naloka et al., 2024). The effectiveness of *Pseudomonas aeruginosa* bacteria in wastewater bioremediation is detailed in Table 2.

Table 2. Wastewater Bioremediation by *Pseudomonas aeruginosa* Bacteria

Waste Type	Results	Reference
Petroleum refinery wastewater	83% reduction in COD,	(Aswani et al., 2025)
Alcoholic beverage industry wastewater	COD reduction efficiency of control group 45% - 49%, (67% - 71%) treatment group	(Huang et al., 2025)
Industrial wastewater	BOD and COD reduction efficiency 82.35%	(Shah et al., 2025)
Industrial waste	COD degradation efficiency $76.3 \pm 2.8\%$	(Yin et al., 2025)
Textile dye waste	Reduction of BOD 70.16% and COD 49.23%	(Chellapandian et al., 2024)
Landfill leachate water	COD and BOD reduction efficiency $>70\%$	(Arliyani et al., 2023)
Distillery wastewater	Reduced COD 61.32% and BOD 53.53%	(Ratna and Kumar, 2022)
Industrial effluents	BOD and COD reduction efficiency $91.3 \pm 2.1\%$, $97.6 \pm 3.3\%$, and $94.3 \pm 4.4\%$, respectively.	(Al-Ansari et al., 2021)
Factory wastewater	COD removal efficiency of 92% in BA reactor and 85% without reactor	(Ruan et al., 2020)
Wastewater automobile service station	COD removal was 94%	(Mallick and Chakraborty, 2019)

Although *Pseudomonas aeruginosa* has great potential in waste treatment, it should be noted that *Pseudomonas aeruginosa* bacteria are classified as opportunistic pathogens that can pose health risks, especially to individuals with weak immune systems. Some infections in humans are shown in figure 7 (Tuon et al., 2022).

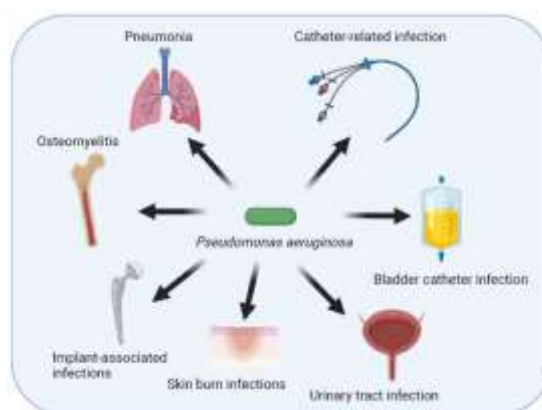


Figure 7. Schematic representation of the main infections caused by *Pseudomonas aeruginosa*

Therefore, it is necessary to implement appropriate risk management or mitigation strategies to minimize the potential negative impacts of using *Pseudomonas aeruginosa* in waste treatment. In this study, a closed reactor was used as a mitigation measure to prevent the release of bacteria into the environment. In addition, additional mitigation was carried out through the sterilization of final waste to ensure safe microbiological quality and compliance with environmental utilization standards. Sterilization was carried out by applying a chemical disinfection method as a final stage, commonly used to eliminate microorganisms in wastewater (Putra et al., 2024).

4.2 Bioremediation Effectiveness

4.2.1 pH and temperature

Environmental factors such as temperature and pH are important parameters that influence the success of the bioremediation process. These two parameters determine the level of metabolic activity of microorganisms and the stability of enzymes that play a role in pollutant degradation. In this study, they were within the optimal range for the growth and metabolic activity of *Pseudomonas aeruginosa*. This bacterium is able to grow well at neutral to slightly alkaline pH and mesophilic temperatures. pH stability indicates that during the degradation process, there were no significant changes that could inhibit enzymatic activity, while a constant temperature supported the optimal rate of biological reactions. Thus, the stability of these two parameters indicates that the incubation environment was sufficiently conducive to supporting the BOD and COD bioremediation processes by *Pseudomonas aeruginosa* on the small island (Alsukaibi et al., 2023).

In general, mesophilic microorganisms that live at temperatures between 20 and 40°C are the most widely used in the biodegradation process. Temperature plays an important role in influencing the rate of this process; increasing the temperature to the optimal limit can accelerate microbial activity, while excessively high temperatures can actually inhibit growth and enzymatic activity. Conversely, low temperatures tend to slow down

the biodegradation process, although microbial activity can still occur at a slower rate. In addition to temperature, the pH of the environment is also an important factor that affects the efficiency of biodegradation because it directly impacts the activity and survival of microorganisms (Aswani et al., 2025).

The optimal pH range varies depending on the type of microbe and organic compound being degraded, but generally ranges from pH 6 to 9. pH values outside this range can reduce the efficiency of microbial activity. In the context of bioremediation, pH also affects the adsorption of contaminants by microorganisms. High pH (alkaline) tends to promote precipitation, while low pH can increase the attraction of contaminants to the surface of microbial cells, affecting the interaction between microorganisms and pollutants (Moreno et al., 2024).

4.2.2 Biochemical oxygen demand (BOD) analysis

The initial BOD concentration of 177.5 mg/L decreased. Figure 6(a) shows that in the control (0%), BOD decreased from 62.60 mg/L (day 2) to 48.18 mg/L (day 8). This decrease was caused by the activity of indigenous microorganisms naturally present in the wastewater sample, which were able to degrade organic compounds even without the addition of external bacteria. However, in the treatment with the addition of *Pseudomonas aeruginosa*, the efficiency of organic degradation increased by about 5 - 8% compared to the control. This indicates that the presence of additional inoculum can accelerate the process of breaking down complex organic compounds into simpler forms.

The addition of 5% inoculum showed a decrease from 63.40 mg/L to 50.84 mg/L in the same period. The 10% treatment experienced a decrease from 56.60 mg/L (day 2) to a low of 40.50 mg/L (day 6), before increasing again to 46.84 mg/L (day 8). Meanwhile, the 15% concentration decreased from 51.68 mg/L (day 2) to 44.50 mg/L (day 6), then increased to 47.18 mg/L (day 8). The increase in the 10% and 15% concentrations on day 8 was influenced by the maximum degradation capacity of the bacteria. Each bacterium has a limited capacity to break down organic matter. Once this capacity is reached, an increase in the number of bacteria is no longer proportional to an increase in degradation effectiveness. This situation is often referred to as the saturation point, where the ratio of available substrate to bacterial population no longer supports an increase in the degradation rate (Altowayti et al., 2022).

In addition, the increase in BOD during this period can be determined by observing the pattern or curve of bacterial growth, which consists of several phases: lag phase (adaptation phase), log phase (exponential growth phase), stationary phase, and death phase. On days 2 and 4, the bacteria were in the lag and log phases, where they

adapted to the new environment, followed by the log phase, where the bacterial population grew rapidly and enzymatic activity was high, enabling maximum degradation of organic matter, as seen in the significant decrease in BOD, especially at a concentration of 10%, which reached 41.98 mg/L on day 4 and 40.50 mg/L on day 6. During the incubation period, bacterial activity accumulated exponentially. The number and enzymatic activity of *Pseudomonas aeruginosa* increased over time. Under favorable environmental conditions, bacteria will continue to multiply and accelerate the degradation of organic compounds, thereby increasing the efficiency of pollutant reduction, such as BOD.

On days 6 - 8, bacteria entered the stationary or death phase, especially at higher concentrations (10% and 15%). In this phase, bacterial growth begins to slow, possibly due to nutrient depletion and the accumulation of toxic metabolites such as organic acids. In addition, at high bacterial concentrations, competition between bacterial cells becomes more intense, both in obtaining nutrients and dissolved oxygen. Oxygen deficiency in the environment causes the degradation process to be ineffective and can even trigger the activity of other microorganisms such as anaerobic or facultative bacteria that produce reductive compounds that actually increase the BOD load. On the other hand, in the death phase, dead bacterial cells undergo lysis or rupture, releasing their contents, such as proteins, lipids, and nucleic acids, back into the medium. These compounds are organic and easily degradable, thereby increasing the concentration of dissolved organic matter detected as an increase in BOD (Aswani et al., 2025).

Based on Figure 6(c), the BOD removal efficiency at various bacterial inoculum concentrations ranged from 64.73 to 77.18%. The highest efficiency was observed at a bacterial concentration of 10%. Meanwhile, Figure 6(d) shows that the efficiency increased from $\pm 64\%$ on day 2 to 70.51-78% on days 4 and 6, then decreased slightly on day 8 (71 - 73%). The highest efficiency at an incubation time of 6 days was 77.18%. Although there was a decrease in BOD and an increase in efficiency with variations in bacterial concentration, the results of the one-way ANOVA test showed that the bacterial concentration did not have a significant effect ($p > 0.05$). The same study stated that bacterial concentrations ranging from 0.7% to 1% did not significantly affect BOD reduction. Although BOD decreased in both treatments, the reduction efficiency at a concentration of 0.7% was higher at 80% compared to a concentration of 1%, which only reached 72% (Fathina, 2024).

Meanwhile, the incubation time had a significant effect on the reduction of BOD in domestic wastewater on small islands ($p < 0.05$). The incubation time of bacteria plays an important role in determining the effectiveness

of the degradation process of organic matter contained in waste. The longer the incubation time, the more time bacteria have to increase their metabolic activity, thereby optimizing the degradation process (Safitri et al., 2025) . The same study shows that incubation time has a significant effect on the effectiveness of BOD reduction in hospital wastewater. The results of the study noted that BOD reduction reached 73.86% with a residence or incubation time of 24 hours using a zeolite bioreactor bioball with added *Pseudomonas aeruginosa* bacteria in a case study at Prambanan Regional General Hospital (Desica et al., 2020) .

Pseudomonas aeruginosa degrades dissolved organic matter in water through aerobic metabolism and enzyme production. *Pseudomonas aeruginosa* uses oxygen to degrade organic compounds under aerobic conditions and involves enzymes that break down complex organic compounds into simpler compounds that can then be used as a source of energy and carbon by microbes. *Pseudomonas aeruginosa* produces various enzymes, namely lipase, protease, and amylase, which assist in the breakdown of lipids, proteins, and carbohydrates. This process increases the availability of digestible organic substrates, thereby reducing the BOD (Grbavcic et al., 2011; Hu et al., 2023).

4.2.3 Chemical oxygen demand (COD) analysis

Based on COD measurements after degradation by *Pseudomonas aeruginosa* in domestic wastewater on the small island, COD decreased with increasing incubation time and bacterial concentration. The initial COD was recorded at 449.32 mg/L, indicating a high organic load. In Figure 6(b), the control (0% additional bacteria) COD decreased from 146.85 mg/L (day 2) to 120.66 mg/L (day 8), indicating degradation by indigenous microorganisms. The 5% treatment showed a decrease in COD from 164.31 mg/L to 127.10 mg/L during the same period, which was greater than that of the control but did not achieve maximum degradation efficiency. At a concentration of 10%, COD decreased significantly from 157.40 mg/L (day 2) to 101.35 mg/L (day 6), then increased again to 117.04 mg/L on day 8. This indicates that the degradation effectiveness of *Pseudomonas aeruginosa* was optimal until day 6, but subsequently decreased due to substrate limitations or environmental conditions that were no longer supportive. The increase in COD on day 8 was comparable to a concentration of 15% (117.84 mg/L), indicating that high bacterial concentrations have the potential to trigger population imbalances, nutrient competition, or the accumulation of toxic metabolites.

The COD decline in wastewater from small islands reflects the growth phases of *Pseudomonas aeruginosa* during the biodegradation process. On day 2, the bacteria were in the lag phase, transitioning towards the logarithmic phase, so the COD value was still high, although a faster adaptation was observed at a concentration

of 15% (137.10 mg/L). Entering day 4, the bacteria entered the logarithmic phase, characterized by maximum metabolic activity, as indicated by a significant decrease in COD, particularly at concentrations of 10% and 15% (lowest at 128.98 mg/L). On the 6th day, the system reached the stationary phase, marked by a decrease in degradation efficiency due to substrate limitations and metabolite accumulation. Furthermore, with a longer contact time (day 8), the degradation process began to slow and even tended to stagnate due to nutrient competition and saturated environmental conditions (Khadijah et al., 2023) .

Figure 6(e) shows the COD removal efficiency at various bacterial inoculum concentrations, with an efficiency range of 63.43 - 77.44%. The highest efficiency was achieved at a bacterial concentration of 10%. Figure 6(f) shows that the efficiency increased from 63.43% on day 2 to a range of 64.79 - 77.44% on days 4 and 6, then decreased slightly on day 8 to 71 - 73%. The highest efficiency was achieved at an incubation time of 6 days. The results of the one-way ANOVA test showed that the bacterial concentration did not have a significant effect ($p > 0.05$). The same study stated that *Pseudomonas aeruginosa* has a high ability to reduce COD by up to 79%, but an increase in microbial population or bacterial concentration does not always correlate directly with the effectiveness of COD reduction. This indicates that the number of bacteria alone is not sufficient to determine the success of COD degradation.

Conversely, the results of the one-way ANOVA test on COD levels based on incubation time showed significant values ($p < 0.05$). This indicates that incubation time plays an important role in the COD reduction process, so that variations in incubation duration can affect the level of organic oxidant degradation in the system. The same study states that incubation time has a significant effect on the reduction of COD values (Harahap et al., 2023) . Thus, although the bacterial concentration factor can modify the degradation rate, the duration of microbial exposure to the substrate is a critical factor in achieving optimal COD reduction levels. A longer incubation period tends to increase COD efficiency to a certain point. After that, the increase effect becomes saturated depending on environmental conditions and substrate availability (Yin et al., 2025) .

4.3 Implementation Implications for Small Island

The characterization of small island with limited waste treatment facilities, water quality that is highly susceptible to pollution, and seawater intrusion that increases salinity, requires the presence of adaptive microorganisms to support bioremediation efforts. *Pseudomonas aeruginosa* is a bacterium with high metabolic flexibility, capable of growing in both aerobic and anaerobic conditions, and tolerant to variations in nutrients and

salinity levels. This ability makes it a potential candidate for domestic waste treatment on small island, where water quality is easily degraded due to high community activity and limited waste treatment infrastructure. In addition, this bacterium has an enzymatic system that is effective in breaking down complex organic compounds, thereby significantly reducing BOD and COD levels even in dynamic and challenging environmental conditions. Thus, the use of *Pseudomonas aeruginosa* is ecologically and sustainably relevant, as it is in line with the need for adaptation in island regions that are vulnerable to changes in environmental quality.

The results of this study indicate that optimizing the waste treatment process is more effectively achieved through retention time adjustment. In the context of small islands equipped with household-scale communal wastewater treatment plants (WWTPs), these results are practically relevant as they open up the possibility of implementing more efficient, economical, and easily replicable treatment strategies. The research results also confirm the ability of *Pseudomonas aeruginosa* to adapt to a range of inoculum concentrations. However, adequate incubation time is required for metabolic activity to reach optimal conditions in reducing pollutant loads. Thus, this study provides an empirical basis for the development of more effective and sustainable domestic wastewater management practices in small island regions, with a focus on optimizing process duration and environmental conditions that support the activity of degrading microbes.

4.4 Research Limitations

Although the results of the study show promising potential, this study has several limitations that need to be considered before it is implemented on a larger scale. First, the use of *Pseudomonas aeruginosa* to the simpler health risks, thus requiring strict supervision and control in its application. Second, the research parameters only focused on measuring BOD and COD, so other aspects such as measuring nitrogen and phosphorus levels were not examined, meaning that nutritional factors could not be analyzed as variables affecting the effectiveness of bioremediation. These limitations provide a basis for further research with a more comprehensive scope of analysis.

5. CONCLUSIONS

Pseudomonas aeruginosa effectively reduced BOD and COD in domestic wastewater from a small-island context. Variations in bacterial concentration did not have a significant effect on BOD and COD reduction, whereas incubation time had a significant effect, with optimal degradation occurring on day 6. Based on these findings, we recommend that *Pseudomonas aeruginosa*- based bioremediation focus on optimizing incubation time to achieve maximum BOD and COD reduction.

Author Contributions: Conceptualization and methodology, ABB, N, and HA; Software, N; Validation, ABB and HA; Formal analysis, GP and SMS; preparation and writing of initial draft, ABB and N; Writing, Revision, and Editing, SMS, Visualization, N; Supervision, ABB. The final manuscript has been revised and approved by all contributing authors.

Funding: The BIMA Research Grant 2024 awarded by the Ministry of Education, Culture, Research, and Technology.

Acknowledgments: We would like to express our gratitude to the Institute for Research and Community Service (LPPM) of Hasanuddin University for its guidance and support during the implementation of this research. We would also like to express our gratitude to all parties who, directly or indirectly, have contributed to the success of this study. Your contributions and cooperation are greatly appreciated.

Conflicts of Interest: The author confirms that there is no conflict of interest.

REFERENCES

- Al-Ansari, M.M., Benabdelkamel, H., AlMalki, R.H., Abdel Rahman, A.M., Alnahmi, E., Masood, A., Ilavenil, S., Choi, K.C., 2021. Effective Removal of Heavy Metals from Industrial Effluent Wastewater by A Multi Metal and Drug Resistant *Pseudomonas Aeruginosa* Strain RA-14 using Integrated Sequencing Batch Reactor. *Environ. Res.* 199, 111240. <https://doi.org/10.1016/j.envres.2021.111240>
- Alsukaibi, A.K., Alimi, F.R., Mechi, L., Otaibi, A.A., K, A., A.A.Alshamari, Alshammari, E.M., Khan, M.W.A., 2023. Optimization of *Pseudomonas aeruginosa* Isolated for Bioremediation from Ha'il Region of Saudi Arabia. *Bioinformation* 19, 893–900. <https://doi.org/10.6026/97320630019893>
- Altowayti, W.A.H., Shahir, S., Eisa, T.A.E., Nasser, M., Babar, M.I., Alshalif, A.F., AL-Towayti, F.A.H., 2022. Smart Modelling of a Sustainable Biological Wastewater Treatment Technologies: A Critical Review. *Sustainability* 14. <https://doi.org/10.3390/su142215353>
- Arliyani, I., Tangahu, B.V., Mangkoedihardjo, S., Zulaika, E., Kurniawan, S.B., 2023. Enhanced Leachate Phytodetoxification Test Combined With Plants and Rhizobacteria Bioaugmentation. *Heliyon* 9, 12921. <https://doi.org/10.1016/j.heliyon.2023.e12921>
- Aswani, K.V., Manu Sankar, V., Kalamdhad, A.S., Das, C., 2025. Microbial Enrichment-Based Bioremediation of Petroleum Refinery Wastewater- Enhanced Effluent Quality Assessment by Phytotoxicity Studies. *Bioresour. Technol. Reports* 29, 102043. <https://doi.org/10.1016/j.biteb.2025.102043>
- Bai, F., Liu, S., Gu, X., Wang, F., 2024. Highly Efficient Low-Temperature Biodegradation of Nitrogenous Pollutions in Domestic

- Wastewater Via Immobilized-Microbial Bioaugmentation Coupled with Hybrid Membrane Bioreactor. *Chem. Eng. J.* 485, 149705. <https://doi.org/10.1016/j.cej.2024.149705>
- Birawida, A.B., Selomo, M., Natsir, M.F., Rahmawati, I., Rachmat, M., 2020. Sanitation and The Existence of Bacteria in Drinking Water Towards Diarrhea in Barrang Lompo Island. *J. Nas. Ilmu Kesehat.* 3, 10–19. <https://doi.org/https://journal.unhas.ac.id/index.php/jnik/article/download/10385/5534>
- Buslima, F.A., Abu Hasan, H., Sheikh Abdullah, S.R., Othman, A.R., 2024. Water Recovery from Domestic Wastewater Using Integrated Biofilm-Phytoremediation Technology: A review. *J. Water Process Eng.* 65, 105875. <https://doi.org/10.1016/j.jwpe.2024.105875>
- Chellapandian, K., Devapriam, J.J., Lakshmipathy, V., Arumugam, B., Ramaraj, S.K., Al-Qahtani, W.H., 2024. Bioremediation of Textile Effluent using Indigenous Microbes. *J. Indian Chem. Soc.* 101, 101217. <https://doi.org/10.1016/j.jics.2024.101217>
- Desica, S., Masykuri, M., Setyono, P., 2020. Efisiensi Penyisihan Bod Dan Cod Limbah Cair Rumah Sakit Menggunakan Variasi Komposisi Media Zeolit-Bioball Dan Waktu Tinggal Hidraulik Dari Bioreaktor Anaerob-Aerob (Studi Kasus Rsud Prambanan). *Ekosains* 12, 10–19. <https://jurnal.uns.ac.id>.
- Fathina, D.S., 2024. Bioremediasi Logam Timbal (Pb) menggunakan Bakteri Indigenous Tanah TPA Piyungan pada Air Limbah Industri Tekstil. Islamic University of Indonesia. <https://dspace.uui.ac.id>.
- Grbavcic, S., Bezbradica, D., Zivkovic, I., Avramovic, N., Milosavic, N., Karadzic, I., Jugovic, Z.K., 2011. Production of Lipase and Protease from an Indigenous *Pseudomonas aeruginosa* Strain and their Evaluation as Detergent Additives: Compatibility Atudy with Detergent Ingredients and Washing Performance. *Bioresour. Technol.* 102, 11226–11233. <https://doi.org/10.1016/j.biortech.2011.09.076>
- Harahap, D., Rahmad, Z., Yahya, H., Harahap, J., 2023. The Ability of *Pseudomonas aeruginosa* PAO1 to Absorb Iron (Fe) in Leachate Waste at the Gampong Jawa Landfill in Banda Aceh City. *J. Biol. Sci. Appl. Biol.* 3, 15–24. <https://doi.org/10.22373/kenanga.v..i>
- Harahap, R.G., Nurmawati, N., Dianiswara, A., Putri, D.L., 2021. T Pelatihan Pembuatan Eco-Enzyme sebagai Alternatif Desinfektan Alami di Masa Pandemi Covid-19 bagi Warga Km.15 Kelurahan Karang Joang. *Journal of Community Service*

- Center. 5, 67. <https://doi.org/10.24127/sss.v5i1.1505>
- Hu, F., Wang, P., Li, Y., Ling, J., Ruan, Y., Yu, J., Zhang, L., 2023. Bioremediation of Environmental Organic Pollutants by *Pseudomonas aeruginosa*: Mechanisms, Methods and Challenges. Environ. Res. 239, 117211. <https://doi.org/10.1016/j.envres.2023.117211>
- Huang, S., Li, H., Zhou, C., Zhang, H., Sun, D., Liu, X., Dang, Y., 2025. Enhancement of Partial Denitrification by Nirs -Deficient Strain of *Pseudomonas Aeruginosa* in Treatment of Liquor Brewing Wastewater. J. Water Proce 75, 108035.
- Izzati, M., 2018. Perbandingan Hasil Identifikasi Bakteri Gram Negatif Menggunakan Teknik Biokimia Otomatis (Vitek® 2) Dan MALDI-TOF MS (VITEK® MS). Universitas Sebelas Maret, Surakarta, 02. <https://digilib.uns.ac.id>
- Khadijah, N., Manalu, K., Nasution, R.A., 2023. Pemanfaatan Bakteri *Pseudomonas Putida* Sebagai Agen Bioremediasi Untuk Penurunan Kadar Fosfat Pada Limbah Cair Jasa Laundry di Medan. J. (Biology Educ. Sci. Technol. 6, 542–548. <https://jurnal.uisu.ac.id>
- Khotimah, S.N., Mardhotillah, N.A., Arifaini, N., Sumiharni, 2021. Karakterisasi Limbah Cair Greywater pada level Rumah Tangga Berdasarkan Sumber Emisi. J. Saintis 21, 71–78. [https://doi.org/10.25299/saintis.2021.vol21\(02\).7876](https://doi.org/10.25299/saintis.2021.vol21(02).7876)
- Kumar, M., Mishra, G. V., 2024. Causes and Impacts of Water Pollution on Various water Wodies in the State of Rajasthan, India: A Review. Environ. Ecol. 42, 645–654. <https://doi.org/10.60151/envec/qiyj5706>
- Lalucat, J., Mulet, M., Gomila, M., García-Valdés, E., 2020. Genomics in bacterial taxonomy: Impact on the genus pseudomonas. Genes (Basel). 11. <https://doi.org/10.3390/genes11020139>
- Mallick, S.K., Chakraborty, S., 2019. Bioremediation of Wastewater from Automobile Service Station in Anoxic-Aerobic Sequential Reactors and Microbial Analysis. Chem. Eng. J. 361, 982–989. <https://doi.org/10.1016/j.cej.2018.12.164>
- Moreno, A.L., Ordonez, A.V., Villaverde, J., Madrid, F., Carlier, J.D., Santos, J.L., Alonso, E., Morillo, E., 2024. Bacterial Bioaugmentation for Paracetamol Removal from Water and Sewage Sludge. Genomic Approaches to Elucidate Biodegradation Pathway. J. Hazard. Mater. 480, 136128. <https://doi.org/10.1016/j.jhazmat.2024.136128>
- Naloka, K., Kuntaveesuk, A., Muangchinda, C., Chavanich, S., Viyakarn, V., Chen, B., Pinyakong, O., 2024. *Pseudomonas* and

- Pseudarthrobacter* are the Key Players in Synergistic Phenanthrene Biodegradation at Low Temperatures. *Sci. Rep.* 14, 1–14. <https://doi.org/10.1038/s41598-024-62829-y>
- Prastiwi, A.C., Rosariawari, F., 2023. Identifikasi dan Penentuan Strategi Pengelolaan Limbah Domestik Pada Kawasan Pesisir Dusun Kisik , Kabupaten Pasuruan. *J. Sains dan Teknol.* 2, 947–957. <https://doi.org/10.55123/insologi.v2i5.2670>
- Putra, S.D.A., Putri, R.D.C., Airlangga, H.M.R., 2024. Sebaran Profil Bakteri *Pseudomonas aeruginosa* Dan Profil Resistensi Antibiotiknya Pada Saluran Pembuangan Limbah Air Pada Salah Satu Rumah Sakit Tipe C Di Malang. *J. Kedokt. Komunitas* 12, 1–11. <https://jim.unisma.ac.id>
- Rafiaee, S., Samani, M.R., Toghraie, D., 2020. Removal of hexavalent chromium from aqueous media using pomegranate peels modified by polymeric coatings: Effects of various composite synthesis parameters. *Synth. Met.* 265. <https://doi.org/10.1016/j.synthmet.2020.116416>
- Rahardja, boedi s, Parogo, Mahasri, G., Hardhianto, mohammad dwi, 2010. Efektifitas Bakteri *Pseudomonas* sebagai Pengurai Bahan Organik (Protein, Karbohidrat, Lemak) Pada Media Air Limbah Pembenihan Ikan Lele Dumbo (*Clarias Sp.*) Sistem Resirkulasi Tertutup. *J. Ilm. Perikan. dan Kelaut.* 2, 72–73.
- Rahma, A.N., Purwanti, I.F., 2020. Sistem Pengelolaan Air Limbah Domestik Kecamatan Kota, Kota Kediri. *J. Tek. ITS* 9, 231–236. <https://doi.org/10.12962/j23373539.v9i2.56924>
- Ratna, S., Kumar, R., 2022. Production of Di-Rhamnolipid with Simultaneous Distillery Wastewater Degradation and Detoxification by Newly Isolated *Pseudomonas aeruginosa* SRRBL1. *J. Clean. Prod.* 336, 130429. <https://doi.org/10.1016/j.jclepro.2022.130429>
- Ruan, X., Yin, J., Cui, X., Li, N., Shen, D., 2020. Bioaugmentation and Quorum Sensing Disruption as Solutions to Increase Nitrate Removal in Sequencing Batch Reactors Treating Nitrate-Rich Wastewater. *J. Environ. Sci. (China)* 98, 179–185. <https://doi.org/10.1016/j.jes.2020.06.007>
- Safitri, D., Reulina, Y., Sebayang, A., Pinaring, N., Sari, M.N., Febriyosa, A., 2025. Efektivitas Bioremediasi Menggunakan Bakteri *Pseudomonas* untuk Menurunkan Kadar COD Limbah Organik di Pabrik Cincin Kota Medan. *Biopendix* 11, 161–167.
- Shah, A., Shah, M., 2020. Characterisation and Bioremediation of Wastewater: A Review Exploring Bioremediation as A

- Sustainable Technique for Pharmaceutical Wastewater. *Groundw. Sustain. Dev.* 11, 100383. <https://doi.org/10.1016/j.gsd.2020.100383>
- Shah, S.M., Khan, S., Bibi, N., Rehman, B., Ali, R., Shireen, F., Yilmaz, S., Ali, Q., Ullah, A., Ali, D., 2025. Indigenous Bacteria as Potential Agents for Trace Metal Remediation in Industrial Wastewater. *Sci. Rep.* 15, 1–22. <https://doi.org/10.1038/s41598-025-97711-y>
- Singh, A., Srivastava, A., Saidulu, D., Gupta, A.K., 2022. Advancements of Sequencing Batch Reactor for Industrial Wastewater Treatment: Major Focus on Modifications, Critical Operational Parameters, and Future Perspectives. *J. Environ. Manage.* 317, 115305. <https://doi.org/10.1016/j.jenvman.2022.115305>
- Tuon, F.F., Dantas, L.R., Suss, P.H., Ribeiro, V.S.T., 2022. Pathogenesis of the *Pseudomonas aeruginosa* Biofilm: A Review. *Pathogens* 11, 300. <https://doi.org/10.3390/pathogens11030300>
- Vasdravanidis, C., Alvanou, M. V., Lattos, A., Papadopoulos, D.K., Chatzigeorgiou, I., Ravani, M., Liantas, G., Georgoulis, I., Feidantsis, K., Ntinis, G.K., Giantsis, I.A., 2022. Aquaponics as a Promising Strategy to Mitigate Impacts of Climate Change on Rainbow Trout Culture. *MDPI Anim.* 12, 1–27. <https://doi.org/10.3390/ani12192523>
- Xiao, H., Peng, Y., Zhang, Q., Liu, Y., 2021. Pre-Anaerobic Treatment Enhanced Partial Nitrification Start-up Coupled with Anammox for Advanced Nitrogen Removal from Low C/N Domestic Wastewater. *Bioresour. Technol.* 337, 125434. <https://doi.org/10.1016/j.biortech.2021.125434>
- Yin, X., Wang, F., Ge, M., Zhang, F., Liang, G., 2025. *Pseudomonas Aeruginosa* Promoted Microbial Fuel Cells for Cytidine Acid Production Wastewater Treatment, *Scientific Reports*. <https://doi.org/10.1038/s41598-025-90361-0>