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RESEARCH ARTICLE

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Effect of Eco-enzyme on Water Quality Parameters in Some Rivers Disembogued at Ambon Bay

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ABSTRACT

Due to the increasing population in Ambon City, the high volume of organic waste in rivers that empties into Ambon Bay can impact the marine ecosystem. On the other hand, processing organic waste in the form of eco-enzymes has been widely applied and has benefits, for instance, as a cleaning liquid. This study aimed to analyze the effect of eco-enzyme concentrations on water quality parameters, such as pH, ammonia, detergent, coliform, nitrate, phosphate, BOD, COD, and DO in several rivers flowing into Ambon Bay. The research was conducted from May to November 2022. Water samples were collected from four rivers that flow into Ambon Bay. Production of eco-enzyme was carried out at the Marine Science Laboratory at Pattimura University for three months. Eco-enzyme derived from banana peels obtained from tourist areas on Ambon Island. The research applied the method of observation and experiment. The data of each parameter obtained from the treatments (sample before and after treatment using 10% and 20% of eco-enzyme) were then compared with Second-Class of River Water Quality Standards. The results showed that eco-enzyme could neutralize the pH value. The DO, ammonia, detergent and coliform concentration decreased after adding 10% and 20% of the eco-enzyme. By contrast, adding eco-enzyme to the river water can increase the concentration of nitrate, phosphate, BOD, and COD.

Introduction

Human activities cause waste pollution both on land and in water [1]. The waste problem in Ambon Bay has become a serious concern for local governments and communities [2]. Previous studies have shown that organic and inorganic waste in the waters of Ambon Bay negatively impacts ecosystems. The category of inorganic waste that dominates Ambon Bay is plastic waste, which ranges from 76% to 90% [3,4]. Research has confirmed that plastic waste has been identified as a microplastic in the water column and aquatic biota, which can harm the growth of fish and mollusks [5,6]. Organic waste, as well as inorganic waste, also has a severe impact on water, such as food waste. Food waste accounts for 60% of garbage, of which 50% is made up of fresh fruits and vegetables [7]. Organic waste production accounts for 70% of the total waste generated in Indonesia [8]. The category of organic waste disposed of in water, such as food waste, causes a pungent odor that harms public health and can reduce oxygen levels needed by aquatic biota [9]. Moreover, the decomposition of organic waste can increase greenhouse gases and trigger global warming [10,11].

Currently, the conditions of Ambon Bay are affected by direct disposal or flow through the river [12]. Several rivers that flow into Ambon Bay contribute to high organic waste because of the high density of residential areas along the river basin [13]. Unfortunately, garbage disposal activities in rivers have become a habit of irresponsible people, resulting in lower water quality both physically and biologically [14]. Physically, changes in the color and odor of water indicate declining river water quality, which can cause disturbances to public health and the life of river, coastal, and marine ecosystems [15].

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Efforts to manage waste are required to reduce the negative impacts of daily waste production. Various efforts have been made to overcome waste production in Ambon Bay waters with the issuance of regulations regarding waste management [16,17] as well as reducing single-use plastic bag usage [18]. In addition, regulations regarding prohibition and agreement for everyone who lives on river banks have been issued, which aim to preserve the river ecosystem, one of which is the disposal of garbage [19]. At the community level, there have been calls for banning garbage dumps into rivers by installing banners and prohibiting boards. However, it can be assumed that waste management activities at the government and community levels have not yet been maximized, as indicated by the large amount of waste disposed of in rivers, both organic and inorganic, that pollutes river ecosystems and empties into coastal and marine waters.

Eco-enzymes, one of the solutions for managing organic waste, can neutralize polluted river water [20,21]. Eco-enzymes are fermented fruit peels mixed with water and brown sugar [22], which are obviously uncomplicated, cheap, and environmentally friendly; therefore, they can be produced quickly. Eco-enzymes have been proven to be effective cleaning fluids, pest repellents, antibacterial agents, air and water purifiers [23,24]. Applying eco-enzymes to clean polluted water can be a solution, considering the high amount of waste dumped in the river, which is finally discharged into the Ambon Bay area. The research aims to analyze the effect of eco-enzyme concentration on water quality parameters, including pH, ammonia, detergent, coliform, nitrate, phosphate, Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), and Dissolved Oxygen (DO) in several rivers flowing into the waters of Ambon Bay.

Materials and Methods

This research was conducted from May to November 2022 using observational and experimental methods. Water samples were collected from four rivers downstream that flow to Ambon Bay: Wai Batu Gantung, Wai Tomu, Wai Batu Merah, and Wai Ruhu (Table 1). The documentation of the research location was conducted on August 2, 2023. The four rivers were selected purposively because of the high level of community activity around the rivers, which directly impacts the quality of the river's water [25,26].

In this study, the produced eco-enzyme solution was derived from banana peels obtained from tourist areas on Ambon Island. People's craze for fried banana snacks results in banana peel waste that accumulates not only in tourist areas but also in several places that sell fried foods. The eco-enzyme solution was prepared using banana peel as the essential ingredient for three months at the Marine Science Laboratory, Faculty of Fisheries and Marine Science, Pattimura University. Brown sugar, fruit peel, and water were mixed in a storage container at a ratio of 1 : 3 : 10 [27]. Furthermore, the mixture was filtered for eco-enzyme harvesting and was ready for use.

Several river water quality parameters, such as pH, ammonia, detergent, coliforms, nitrate, phosphate, BOD, COD, and DO, were analysed. A river water sample was collected using a Nansen bottle and poured into a plastic bottle to measure the chemical parameters and a glass bottle to measure coliforms. pH and DO measurements were carried out directly in the field using a multiparameter water quality meter. Water sampling for coliform parameters was conducted by burning the mouth of the bottle before filling it with river water samples.

Each sample was treated by adding 10% and 20% eco-enzyme and then set aside for five days [28]. Before and after mixing with the eco-enzyme, the samples were analyzed at *Balai Teknik Kesehatan Lingkungan dan Pengendalian Penyakit* (BTKLPP) Ambon, a standardized laboratory, according to ISO/IEC 17025:2017. The analysis of the water quality parameters was referred to as the Standard Method for the Examination of Water and Wastewater [29] using a UV spectrophotometer (UV-1800).

The ammonia test used the phenate method in the range from 0.1 to 0.6 mg/l of NH_3N at 640 nm wavelength [30]. The nitrate test applied the brucine sulfate method at a wavelength of 410 nm, whereas phosphate was tested using the ascorbic acid method at a wavelength of 880 nm [31]. The methylene blue method was applied to analyze the detergent portion by determining the anionic surfactant content. The range used was from 0.025 to 2.0 mg/L at 652 nm wavelength [32]. The DO test applied the Winkler titration method based on a redox reaction [33]. To test the BOD, the water sample was incubated for five days at 20 °C, whereas to test the COD, the closed reflux method was applied by reducing $Cr_2O_7^{2-}$ [34] with different wavelengths according to COD values, that is, 600 nm wavelength to the range from 100 to 900 mg/l and 420 nm wavelength to a COD value smaller than 90 mg/l. The coliforms were analyzed by applying the Most Probable Number (MPN) method [35]. Furthermore, the results were referred to in Government Regulation Number

22 concerning the Implementation of Environmental Protection and Management (Class II River Water Quality Standards) [36]. Data analysis is displayed in graphical form for further discussion.

Table 1. Description of river water sampling locations.

Research sites	Picture	Description
Wai Batu Gantung		It has a length of 5.7 km and passes through Mangga Dua Village, Nusaniwe District. Dense residential areas have resulted in this river being used as a dumping ground for household waste.
Wai Tomu		It has a length of 7 km and passes through Karang Panjang and Uritetu Villages, Sirimau District. The high activity of the community, such as offices, markets, hotels, and schools, resulted in a large amount of waste being generated and disposed of into the river.
Wai Batu Merah		It has a length of 7.3 km and passes through Karang Panjang Village and Negeri Batu Merah, Sirimau District. The high river residential and human activity density resulted in high waste. Consequently, when it rains, the waste density increases and changes the water colour.
Wai Ruhu		It has a length of 12.7 km and passes through Hative Kecil Village, Sirimau District. This river is the second-longest river in Ambon City. In addition to the waste generated by household activities, there has been pretty high sedimentation in the downstream section, resulting from converting upper land into settlements.

Results and Discussion

The results showed that the pH value of the eco-enzyme from banana peels was 3.44, under acidic conditions. Another study also showed that the pH value of the eco-enzyme from banana peel was 3.3, which can be used as a fruit preservative [37]. When the pH of the eco-enzyme is below 4, it is effective [38]. In the process of making eco-enzymes, alcohol, and acetic acid are produced from the fermentation process, so the pH value is acidic. This is better when the eco-enzyme quality is produced under acidic pH conditions. Depending on the type of fruit waste utilized and the makeup of the mixture, the pH of the eco-enzyme might vary from 4.5 to 6.5 [20]. The amount of water supplied and the length of the fermentation process also affected the pH of the eco-enzyme. In general, the pH decreases with increasing fermentation time. A substance derived from microorganisms: an eco-enzyme is produced by the fermentation of vegetable waste. A liquid with bacteria and enzymes that can decompose organic materials in the environment and reduce pollution is the end result. The pH values in the four rivers ranged from 6.74 to 8.09 (Figure 1), which is within the range of quality standards of 6–9. After adding the eco-enzyme, the average pH value ranged from 6.76–6.78 for the 10% concentration and 6.74–6.77 for the 20% concentration.

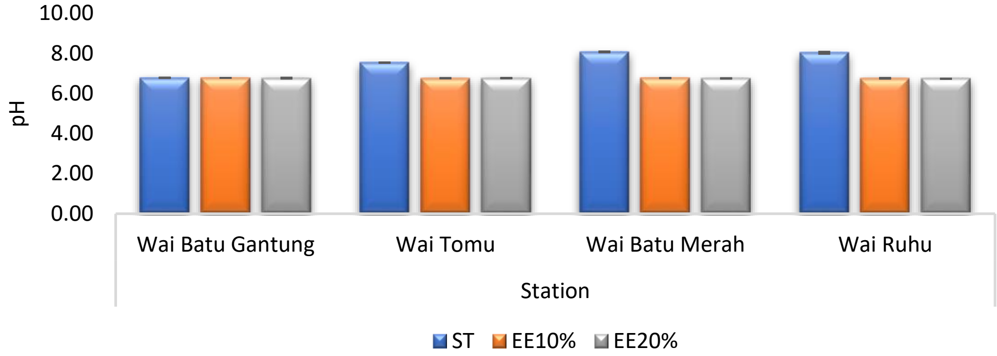

Figure 1. Range of pH values in river water sample tested (ST) and after being treated with eco-enzyme 10% (EE 10%) and 20% (EE 20%).

Eco-enzymes play a crucial role in pH adjustment of water and wastewater. They can effectively increase or decrease pH levels to achieve optimum conditions for various water treatment processes. The results showed that eco-enzymes tend to neutralize the pH of river water. Other research has shown that pH increased from 4.16 to 6.82 in 5 days after adding 10% of eco-enzyme [39]; eco-enzymes can reduce the pH value of wastewater from 9.2 to 6.6 for 270 minutes [40]. Eco-enzymes include amylase, lipase, and trypsin, which can reach their optimum activity at pH 6–8 [41]. Because of the application of eco-enzymes, the metabolism of aquatic organisms can be maintained at an optimum level, and vice versa. When the water pH is too acidic or alkaline, it can influence the metabolism of aquatic organisms [42,43].

The concentration of ammonia ranged from 0.1741 $\pm$ 0.002 to 1.270 $\pm$ 0.000 mg/l (Figure 2). Based on the water quality standards, the total ammonia must not exceed 0.2 mg/l; hence, it can be said that the ammonia value in the four rivers has surpassed the quality standard, especially in Wai Batu Merah, which has the highest value due to the density of settlements around the river and market activities along the river. The presence of ammonia in rivers and coastal and marine waters generally originates from community and industrial activities [44,45]. Ammonia can enter the aquatic environment through indirect pathways, including nitrogen fixation, air deposition, and runoff from agricultural areas, as well as direct pathways, such as animal emissions of nitrogenous waste and municipal effluent discharges [46].

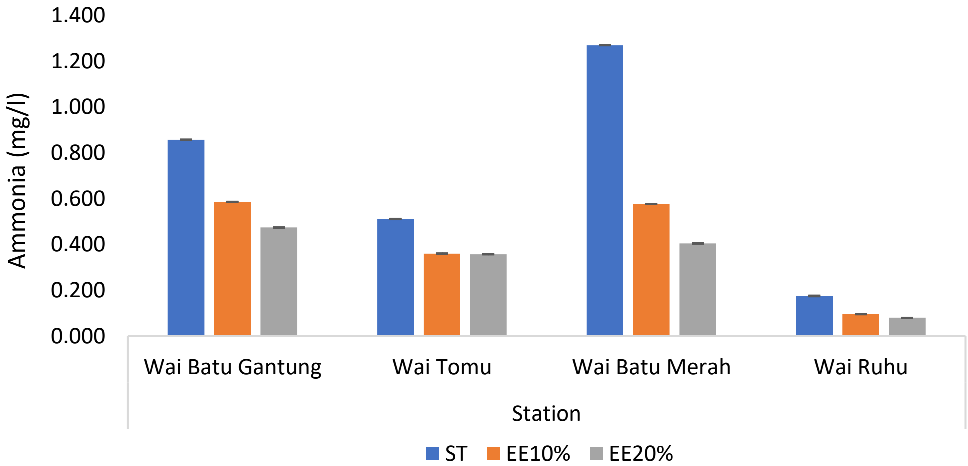

Figure 2. Range of ammonia values in river water sample tested (ST) and after treatment with eco-enzyme 10% (EE 10%) and 20% (EE 20%).

Detergents are cleaning products that households use daily. Heavy stains such as oil, bug excrement, animal dung, and plant-fruit dirt may be successfully removed by detergent product composition, which has good penetration, stripping, dissolving, and decontamination attributes [48]. The direct disposal of household sewage causes detergents to exist in the four rivers, which aggravates the active surfactant ingredients in detergents, leading to dissolved oxygen depletion [49]. It is inevitable that surfactants will be used in homes and businesses and that they will be released into the environment, particularly as effluents in water bodies. Because they are surface-active compounds, most of their applications include emulsifiers, wetting agents, soaps, detergents, and personal care items. The most common class of surfactants is cationic surfactants [50]. These surfactants, which give water bodies their characteristic foam and froth, may be harmful to both the biotic and abiotic elements of the environment. In addition, surfactants are hydrolyzed to produce phosphate, which can trigger nutrient enrichment in water or eutrophication.

The results showed that the detergent concentration in the four rivers ranged from 0.037 ± 0.000 to 0.044 ± 0.000 mg/l and was below the established quality standard of 0.2 mg/l (Figure 3). Adding an eco-enzyme solution containing high acetic acid reduced the detergent concentration immediately in the water to 0 mg/l. The Eco-enzyme contains the enzymes amylase, trypsin, and lipase. The lipase in the eco-enzyme solution functions as a biocatalyst and can degrade the active ingredients of surfactants [51,52]. Several studies have shown that eco-enzymes can also be used as detergents because they contain antibacterial agents. Detergents made from Chinese honeylocust and garbage enzymes are safe to use because they do not contain any harmful pathogens and have a comparatively small microbial community [53].

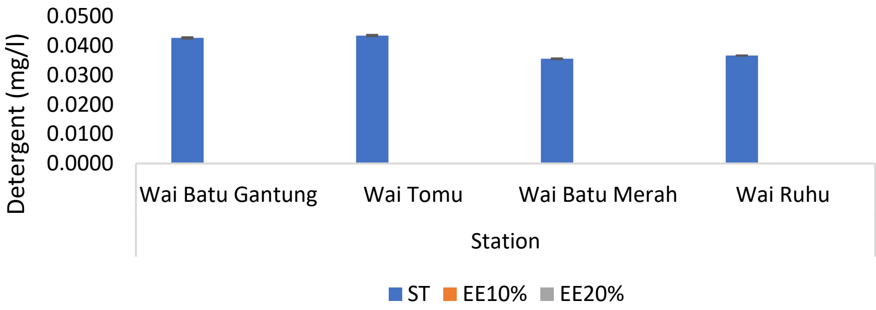

Figure 3. Detergent value range in river water sample tested (ST) and after being treated with 10% (EE 10%) and 20% (EE 20%) eco-enzyme.

Dense settlements on riverbanks contribute to high levels of pollutants, including coliform [54]. The location of densely populated communities, the distance between sewage disposal and adjacent water sources, and residents' habits on the riverbanks are causes of direct urine and feces disposal into the river, causing coliform bacteria contamination [55]. Cases of diarrhea outbreaks in the community result from coliform pollution in river waters due to the disposal of excrement and waste into the river [56]. The results showed that the total coliform in the four rivers was $16,000 \pm 0.000$ MPN/100 ml, exceeding the established quality standard of 5,000 MPN/100 ml. People along the river utilize water for urine and feces disposal, resulting in a high coliform concentration in the river's downstream waters.

The total coliform decreased significantly from 0 ± 0.000 to 366 ± 1.155MPN/100 ml after adding eco-enzyme (Figure 4). As previously mentioned, eco-enzymes are cleaners and disinfectants because they contain lactic acid bacteria, which produce solutions to suppress and eliminate pathogenic or harmful bacteria. According to other studies, eco-enzymes from orange peel could inhibit the growth of *Escherichia coli* bacteria found in chicken coops compared to detergents [57]. One of the key findings of these studies is that eco-enzymes can significantly decrease the levels of coliform bacteria in wastewater. This means that eco-enzymes can effectively reduce the risk of waterborne diseases by targeting and lowering the coliform levels in wastewater. This reduction in coliform levels is attributed to the antimicrobial activities of eco-enzymes, which inhibit bacteria, such as *Escherichia coli*, *Staphylococcus aureus*, *Rhizobium leguminosarum*, *Salmonella typhi, Shigella* spp [58]. This study proved that the greater the concentration of eco-enzyme, the greater the inhibition zone produced. A medium inhibition zone was found in *V. alginolyticus* and *A. caviae* at 100% concentration [59]. Furthermore, eco-enzymes include amylase, protease, cellulase, lipase, and caseinase, which contribute to the degradation and breakdown of organic matter, including coliform bacteria, in wastewater.

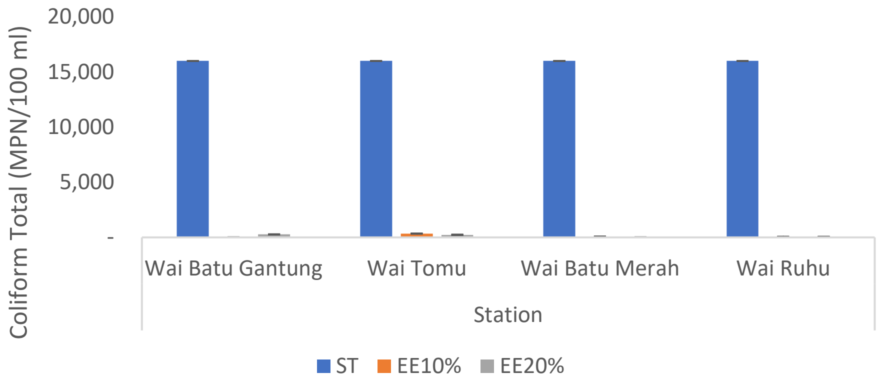

Figure 4. Range of coliform values in river water sample tested (ST) and after treatment with Eco-enzyme 10% (EE 10%) and 20% (EE 20%).

Using rivers as household organic waste disposal sites results in a high rate of decomposition, which produces nutrients in the water. By the results, the nitrate contained in rivers ranged from 0.43 $\pm$ 0.000 to 1.38 $\pm$ 0.001 mg/l and was still under the quality standard of 10 mg/l (Figure 5). Phosphate values ranged from 0.09 $\pm$ 0.001 to 0.33 $\pm$ 0.012 mg/l (Figure 6) and tended to exceed the established quality standard of 0.2 mg/l, especially at Wai Batu Gantung, Wai Tomu and Wai Batu Merah (Figure 6).

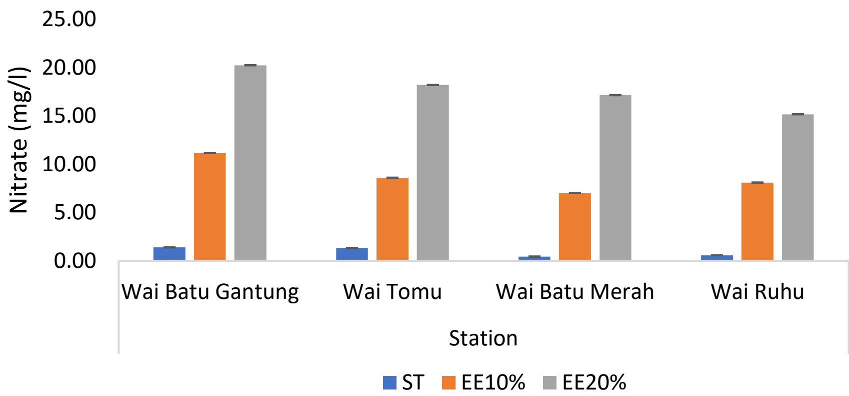

Figure 5. Range of nitrate values in river water sample tested (ST) and after being treated with Eco-enzyme 10% (EE 10%) and 20% (EE 20%).

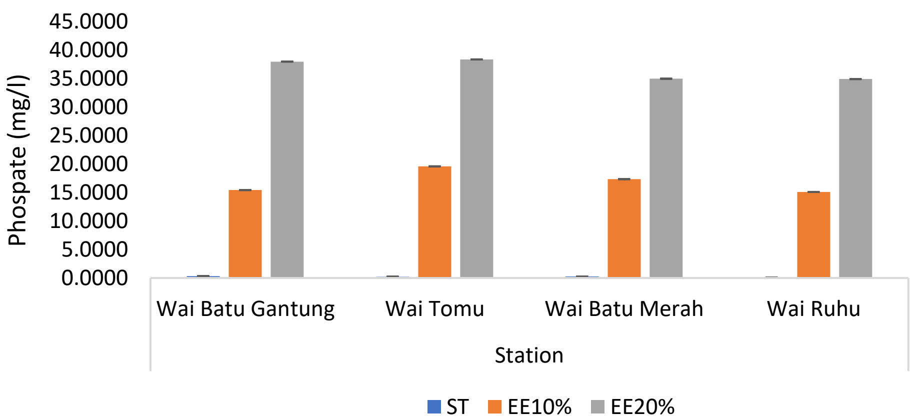

Figure 6. Range of phosphate values in river water sample tested (ST) and after treatment with Eco-enzyme 10% (EE 10%) and 20% (EE 20%).

The presence of eco-enzymes has been found to increase nitrate levels in water samples, leading to potential implications for water quality and the overall health of aquatic ecosystems. The exact mechanisms by which eco-enzymes increase nitrate in water are still being studied, but initial research suggests that these enzymes can break down organic matter and convert it into nitrate through a process known as nitrification. After adding 10% and 20% eco-enzymes, nitrate and phosphate concentrations increased significantly. Phosphate and nitrate are essential nutrients for the growth and metabolism of phytoplankton and are indicators for evaluating the quality and fertility of water. One of the eco-enzyme functions is as a plant fertilizer because of the eco-enzyme's ability to convert ammonia into nitrate with the help of bacteria [60]. Carbon trioxide (CO_3) and nitrate (NO_3), which are soil nutrients, are produced during the process of creating eco-enzymes [61]. This demonstrates that plant nutrients may be derived from eco-enzyme solutions. Consequently, the concentrations of nitrate and phosphate increased after addition of the eco-enzyme.

BOD and COD concentrations generally determine the oxygen consumption level required for decomposing organic matter in water. The results showed that the BOD value exceeded the established quality standard (3 mg/l) at Wai Batu Gantung and Wai Batu Merah (Figure 7). The concentration of BOD decreased to 9.77 $\pm$ 0.023 mg/l in Wai Batu Gantung after adding 10% eco-enzyme concentration and 10.53 $\pm$ 0.017 mg/l at 20% eco-enzyme concentration, but the other three rivers showed an increase in BOD concentration after eco-enzyme was added. However, because the eco-(garbage) enzyme contains a significant quantity of organic material, an increase in BOD may be detected. According to a pond water study, the waste enzyme is successful in lowering ammonia, nitrogen, and phosphorus, but not BOD [62].

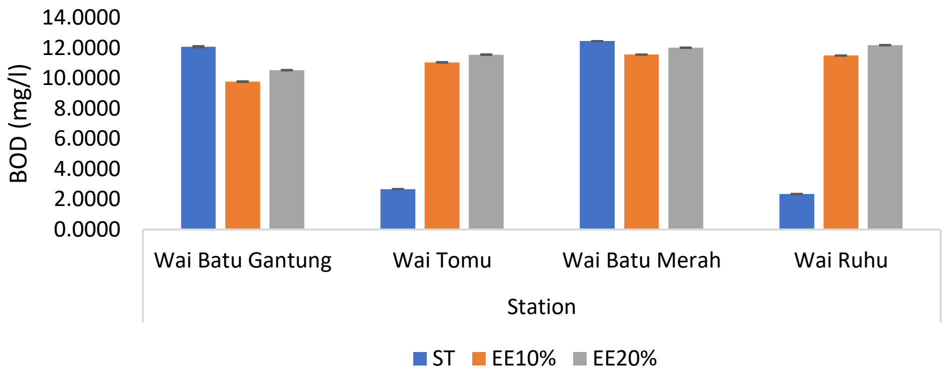

Figure 7. Range of BOD values in river water sample tested (ST) and after being treated with eco-enzyme 10% (EE 10%) and 20% (EE 20%).

COD concentrations in the four rivers were below the quality standard, 25 mg/l for the second-class category, before adding eco-enzyme. Similar to the BOD concentration, COD concentrations increased when eco-enzyme was added (Figure 8). After adding the eco-enzyme, the increase in BOD and COD concentrations affected the depletion of DO concentrations in the river waters from more than 4 mg/l at the initial stage (Figure 9). At 20% concentration, there was an increase in DO in Wai Batu Merah and Wai Ruhu. The presence of DO in water is closely related to organic matter. Although this study did not calculate the total organic matter, it is suspected that this occurred because of the lack of organic matter in these two waters compared with Wai Batu Gantung and Wai Tomu. Based on the color of the water during data collection, Wai Batu Merah and Wai Ruhu showed a light brown color because of the high sediment content in the water. Black water has high organic matter content [63].

The high content of BOD and COD in river water after adding eco-enzymes is thought to be influenced by some microorganisms, as it is mentioned that the number and activity of microorganisms significantly affect the value of BOD and COD [64]. When the number of microorganisms is insufficient, the biochemical breakdown process does not occur, or the intensity of the biochemical breakdown is not significant. Enzyme bacteria in eco-enzyme solutions can minimize aerobic microorganisms such as *E. coli*, which can live under aerobic and anaerobic conditions. Consequently, this process is suspected to reduce the microorganisms that decompose organic matter and increase the concentrations of BOD and COD. The eco-enzyme solution produced from the activity of enzyme bacteria can convert oxygen into hydrogen peroxide because the activities of bacteria occur under anaerobic conditions, causing a decrease in the DO concentrations.

The process of anaerobic metabolism, also known as fermentation, is how bacteria obtain their energy from carbohydrates without oxygen, resulting in alcohol or acetic acid as by-products. The application of eco-enzymes can have an impact on dissolved oxygen levels in water systems. When eco-enzymes are added to wastewater or liquid waste, they can break down organic matter and increase biological oxygen demand levels. This increase in BOD levels means that decomposing microbes require more oxygen for their metabolic processes, decreasing dissolved oxygen levels. This decrease in dissolved oxygen can have negative effects on aquatic ecosystems, as it can lead to hypoxia, which depletes oxygen in water bodies [65]. In addition, eco-enzymes can also inhibit the growth of microorganisms in liquid waste, including those that consume oxygen. Therefore, the application of eco-enzymes can further contribute to a decrease in dissolved oxygen levels.

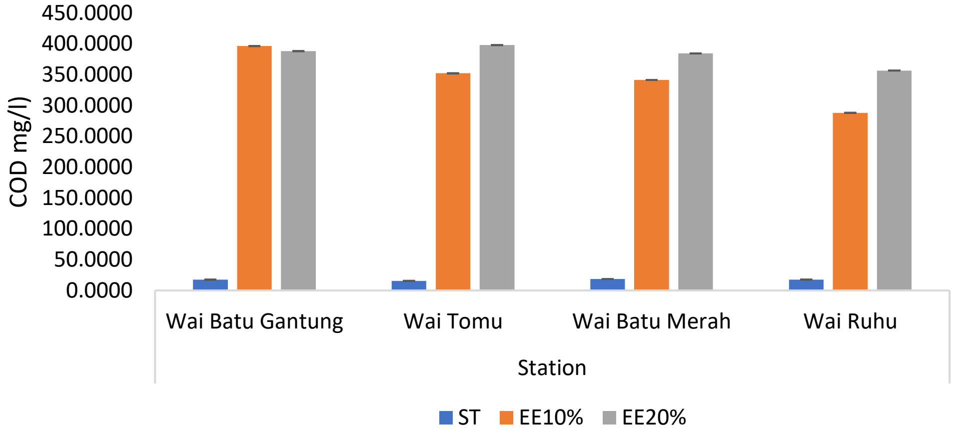

Figure 8. Range of COD values in river water sample tested (ST) and after being treated with eco-enzyme 10% (EE 10%) and 20% (EE 20%).

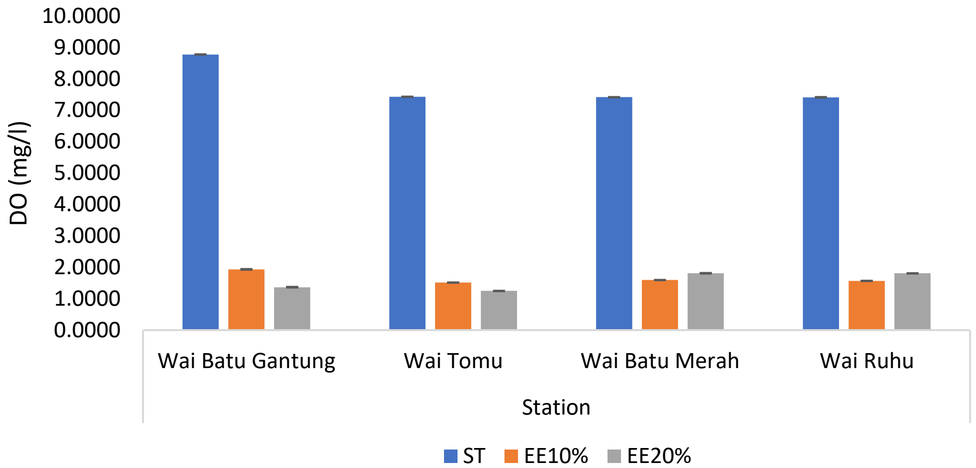

Figure 9. Range of DO values in river water sample tested (ST) and after being treated with eco-enzyme 10% (EE 10%) and 20% (EE 20%).

Conclusions

The addition of eco-enzymes to river water affects water quality parameters. Eco-enzyme is a simple, environmentally friendly technology effective for neutralizing wastewater quality parameters, considering that water quality is an important factor for aquatic ecosystems. The pH, ammonia, detergent, coliform, and DO quantities decreased after adding the eco-enzyme, but the concentrations of nitrate, phosphate, BOD, and COD increased. Further research is needed to determine the effectiveness of eco-enzymes in wastewater at different concentrations.

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