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The Effect of Dietary Probiotic and Oxygen Supply on the Growth and Survival Rate of Eels (*Anguilla bicollor*)

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Abstract: The aquaculture of Anguilla bicollor has significant economic potential, but persistent challenges continue to hinder profitability for farmers. Two of the most pressing issues are low dissolved oxygen levels and poor feed conversion efficiency. These problems slow eel growth and driveup operational costs, creating a bottleneck for sustainable production. This study examines a combined strategy to tackle these challenges by integrating probiotics (Lactobacillus sp.) and paddlewheel aeration. Research was conducted at BLUPPB Karawang, using 600 juvenile eels in ponds subjected to four distinct treatments: (1) probiotics and paddlewheels together, (2) paddlewheels-only, (3) probiotics-only, and (4) a control group with no intervention. Over a 45-day period, survival rates (SR), relative growth rates (RGR), and feed conversion ratios (FCR) were carefully monitored. The findings were promising. Ponds that used both probiotics and paddlewheels achieved the best results: an RGR of 1.19%, an SR of 100%, and an FCR of 1.09. Probiotics contributed to better gut health and digestion, while the paddlewheels boosted water oxygenation-creating an ideal growth environment. While no direct interaction between the two methods was observed, their combined application proved highly effective in improving productivity and sustainability. This integrated approach offers a practical solution to some of the most significant challenges in Anguilla bicollor aquaculture.

Keywords: Eel farming; Feed efficiency; Paddlewheel aeration; Probiotics; Sustainable aquaculture

Introduction

Eel farming (*Anguilla bicollor*) is one of the more profitable sectors of aquaculture, particularly in Indonesia – a country with extraordinary aquatic biodiversity (Jamaluddin et al., 2024). Despite this potential, eel farming in the region, which dates to 1992, still struggles with various technical and operational hurdles. Among these, poor water quality, especially low dissolved oxygen levels, has emerged as a critical challenge. Low oxygen not only slows eel metabolism but also negatively impacts their growth rates. Additionally, feed conversion inefficiency remains a significant problem, as feed costs account for the largest share of production expenses in eel farming. Poor feedback increases operational costs, making profitability a difficult target.

This study explores a novel solution: the combined use of probiotics and paddlewheel aeration. Probiotics are well known for their ability to balance gut microbiota, suppress harmful pathogens, and improve feed efficiency by producing digestive enzymes (Fuller,

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1992). Paddlewheels, on the other hand, are widely used for maintaining optimal dissolved oxygen levels, which not only supports eel metabolism but also enhances the effects of probiotics. Together, these methods aim to address the core technical challenges in eel farming by creating a more efficient and sustainable aquaculture environment.



Figure 1. Schematic illustration

The illustration shows how probiotics and paddlewheels can work together in eel farming to improve aquaculture systems. Paddlewheels help aerate the water, creating currents that boost dissolved oxygen levels. This oxygen is crucial—not just for the eels' metabolism, but also for maintaining a healthy pond environment. The need for research like this stems from the growing demand for effective ways to improve eel growth rates and feed efficiency.

Probiotics, for example, have already been shown to help with fish growth and digestion. Arai (2016) highlighted how probiotics regulate gut bacteria and release beneficial enzymes, which improves feed conversion. On the other hand, aeration systems like paddlewheels play a key role in increasing oxygen levels, which support metabolism. A recent study by Lin et al. (2024) also pointed out that probiotics can enhance fish immunity by strengthening their mucosal layers.

This study tries something new by combining probiotics with paddlewheel technology in eel farming. The idea is simple: improve oxygen levels while allowing probiotics to enhance digestion and feed efficiency. Over 45 days, the research looks at how this approach affects growth rate, SR, FCR, and overall profitability. With a goal of reaching an SR above 75% and an FCR below 3, the study offers a promising solution to make eel farming both more productive and more sustainable.

Method

The study was carried out from February to May 2019 at the Karawang Aquaculture Production Center

(BLUPPB), located in North Pusakajaya Village, Cilebar District, Karawang Regency, West Java, Indonesia. A total of 600 juvenile Anguilla bicollor eels were used, all sourced directly from BLUPPB Karawang. These eels, weighing between 78 and 130 grams on average, went through a one-week acclimation period. This gave them time to adjust comfortably to the new environment before the experiment officially began.

The research used earthen ponds, each measuring 500 square meters, to test how different treatments impacted eel growth. Four ponds were set up for the study: Pond J-1 combined probiotics and paddlewheels, pond J-3 had only paddlewheels, pond J-5 used probiotics alone, and pond J-7 served as the control with no treatments. Within each pond, three smaller enclosures (hapas) measuring 3x5 meters were installed to stock and manage the eels. Water levels in the ponds were maintained between 80 and 120 cm, and about 20–30% of the water was replaced when needed using a stagnant water system.

The eels were fed a commercial diet containing 46% protein every day at 3:00 PM. For the ponds with probiotic treatments, Lactobacillus sp. Bio7 was added to the feed at a dose of 15 ml per 5 kg of food. In the paddlewheel-treated ponds, the devices ran continuously to ensure proper aeration. Observations were made over 45 days, with weight samples taken every 11 days from 5% of the eel population.

Water quality, including pH, dissolved oxygen (DO), ammonia, nitrite, and temperature, was monitored every two days to ensure the ponds remained in good condition. Figure 2 shows the specific placement of the paddlewheels within the ponds.



Figure 2. Schematic of experiment eels pond

Key performance metrics such as SR, weight growth, RGR, and FCR were calculated using the following formulas:

$$SR = \frac{Nt}{No} x \ 100\% \tag{1}$$

Here, Nt is the total number of surviving eels at the end of the study, and No is the initial number of eels stocked. This method follows the guidelines of Safir et al. (2024).

$$W = W_t - W_0 \tag{2}$$

$$RGR = \left((Wt - W0)gr^{\frac{1}{45}} - 1 \right) x \ 100\% \tag{3}$$

This formula calculates the daily growth rate over the 45day experiment period. The approach aligns with the methods described by Setyono et al. (2023).

$$FCR = \frac{F(gr)}{Wt - W0(gr)}$$
(4)

Here, F is the total weight of feed provided, and W_t - W_0 is the weight gain of the eels. The FCR calculation is based on methodologies used by Inayah et al. (2023) and Sekartadji et al. (2023).

Statistical analysis was conducted using ANOVA (Analysis of Variance) with a 2x2 factorial Completely Randomized Design (CRD). All data were analyzed with SPSS version 27, and differences were considered statistically significant at a 0.05 confidence level.

Result and Discussion

The results from this experiment provided key performance metrics, including SR, RGR and FCR for the four treatments J1, J3, J5 and J7. These findings offer valuable insights into how different treatment combinations impact the growth and feed efficiency of *Anguilla bicollor* during the 45-day rearing period.

Table 1. Growth Performance and Feed Utilization of

 Anguilla bicollor during 45 Days of Rearing

| Parameter | J1 | J3 | J5 | J7 |
|-------------|-------|-------|-------|-------|
| Weight (gr) | 55.75 | 45.55 | 32.20 | 18.05 |
| RGR (%) | 1.19 | 0.67 | 0.76 | 0.29 |
| FCR | 1.09 | 1.48 | 1.64 | 3.11 |
| SR (%) | 100 | 100 | 100 | 100 |
| | | | | |

The results of the normality and homogeneity tests for weight, RGR, and FCR showed that all treatment groups—including the probiotic and paddlewheel combinations—met the necessary assumptions for parametric statistical analysis. Normality tests were conducted using the Kolmogorov-Smirnov and Shapiro-Wilk methods, and all groups had p-values greater than 0.05. This indicates that the data were normally distributed. For example, in the probiotic group, the pvalues were 0.200 and 0.623 for weight, 0.174 and 0.127 for RGR, and 0.200 and 0.603 for FCR.

In addition, Levene's test confirmed that the variances were homogeneous across all groups, with p-values exceeding 0.05. For instance, in the probiotic group, p-values for weight, RGR, and FCR were 0.405, 0.159, and 0.311, respectively.

These results provide confidence that the data for weight, RGR, and FCR are suitable for further analysis using parametric statistical methods, such as ANOVA. This ensures that the statistical conclusions drawn from the experiment will be valid and reliable.

Survival Rate

The average SR of eels reared in ponds treated with probiotics and paddlewheels showed no significant differences across all treatment groups (J1, J3, J5, and J7), with SR values consistently at 100%. Essentially, no mortality was observed in any of the treatment ponds. This high SR is likely due to various supportive factors that contributed to eels' health and well-being.

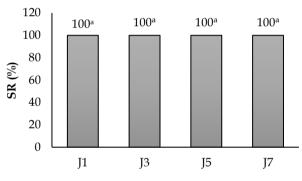


Figure 3. SR graph of Anguilla bicollor reared for 45 days

Water quality parameters played a crucial role in maintaining optimal conditions. Other biotic and environmental factors, such as the absence of competitors and parasites, appropriate population density, the eels' adaptability, and proper handling during cultivation, likely supported these results (Kim, 2019). Similar findings on the beneficial effects of probiotics have been reported in other fish species. For instance, research by Van Doan et al. (2021) found that supplementing tilapia feed with Lactobacillus paracasei L61-27b significantly improved growth performance and boosted immunity by stimulating the mucus layer. Probiotics containing lactic acid bacteria, such as Lactobacillus and Enterococcus, are known to produce antimicrobial compounds like organic acids and enzymes, which help strengthen the immune systems of fish (Qosimah et al., 2023). Additionally, highlighted that probiotics benefit the immune system in multiple ways, supporting the intestinal tract, skin, and gills of fish. These probiotics and their derivatives can act locally on the mucus layer or systemically throughout the body. According to Capaldo et al. (2021) and Harun et al. (2023), certain microorganisms can enter the host's bloodstream or activate immune cells that migrate from mucosal sites to systemic lymphoid tissues. They can exert either immunostimulatory or immunosuppressive effects, influencing both nonspecific and specific immune cells. This enhanced immunity ultimately contributes to higher SRs, as observed in the present study.

Absolute Weight growth

The absolute weight growth of eels in the treatment ponds (J1, J3, J5, and J7) showed clear trends. Factorial ANOVA revealed that both probiotics and paddlewheels significantly influenced weight growth, but there was no interaction between the two. Probiotics had a stronger effect, accounting for 87.8% of weight variation (F = 57.555, p < 0.001), while paddlewheels explained 80.8% of the variation (F = 33.693, p < 0.001). The lack of interaction (F = 0.606, p = 0.459) means their effects are independent.

Pairwise comparisons showed clear differences between the groups. Eels in the probiotic-treated ponds had an average weight that was 21.358 grams higher than those in the non-probiotic group, while those in the paddlewheel-treated ponds were 16.342 grams heavier on average than those in the non-paddlewheel group. Both differences were highly significant (p-value < 0.001). Among the treatment combinations, the group with both probiotics and paddlewheels (J1) achieved the highest average weight (55.750 grams), while the control group without either treatment (J7) had the lowest average weight (18.050 grams). These findings highlight the independent and additive benefits of probiotics and paddlewheels on eel weight growth. While the two factors do not interact, they each contribute significantly to improving growth, and their combined use offers the greatest benefits.

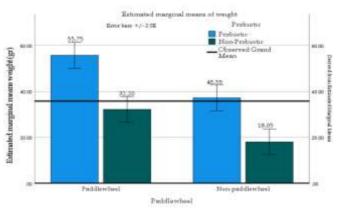


Figure 4. Weight (estimated marginal means) of Anguilla bicollor reared over 45 days

The graph shows the average weight of eels across different treatment groups (probiotic, paddlewheel, both, and control), with error bars (± 2 SE) indicating variability. The highest weight, around 60 grams, was observed in the group treated with both probiotics and paddlewheels, showing the strongest combined effect. Paddlewheels alone resulted in a lower average weight, while probiotics still boosted weight in the absence of paddlewheels, although the effect was smaller. The control group, without any treatment, had the lowest weight, around 20 grams.

The grand mean line highlights that groups with probiotics and/or paddlewheels performed above average, while non-probiotic groups fell below it. Minimal overlap in error bars suggests significant differences between groups. Probiotics and paddlewheels independently and additively improve weight, with their combination delivering the best results. Specifically, the combination treatment (55.75 g) was significantly better than probiotics or the control. However, paddlewheels alone were not significantly different from the control group.

Probiotics from the genus *Lactobacillus* have been widely studied for their beneficial effects on digestive health in aquatic species. Strains such as *Lactobacillus plantarum* (Fujii et al., 2024), have demonstrated the ability to enhance the viability of beneficial microorganisms that support gut health and nutrient absorption (J. S. Lee et al., 2015; Lestari et al., 2020; Park et al., 2020; Peristiwati et al., 2019). This improvement in the gut environment allows fish to absorb nutrients more efficiently, which is partially due to the elongation of the digestive tract's surface area (Suryadi et al., 2023; Chen et al., 2006; Cruz-Guerrero et al., 2014; Dewanti et al., 2022; Triyatmo et al., 2020; Zheng et al., 2019).

Research has also shown that probiotics stimulate intestinal microbiota and enhance digestive enzyme activity (Conforto et al., 2021; Espírito-Santo et al., 2024) which further improves nutrient breakdown and feed utilization. For example, Politis et al. (2023), and Soeprijanto et al. (2018), found that probiotics positively affect enzymatic activity in the intestine, allowing fish to maximize nutrient absorption. This process ultimately leads to better growth performance, as highlighted by Mandele et al. (2024). In addition, probiotics are linked to increased villi surface area in the intestinal tract. This occurs because probiotics, particularly those producing short-chain fatty acids during fermentation, encourage the proliferation of intestinal epithelial cells (Taufik et al., 2023; S. Wang et al., 2023). Short-chain fatty acids not only play a role in cell growth but also create a more efficient digestive environment, allowing fish to absorb more nutrients per unit of feed consumed. Finally, the improvement in nutrient absorption may also stem from optimized digestive enzyme activity. By maintaining a healthy gut environment and supporting the production of enzymes, probiotics enhance the digestive process, allowing fish to make better use of the nutrients in their diet. Together, these factors—improved microbiota, increased villi surface area, short-chain fatty acid production, and enhanced enzyme activity—explain the significant impact of probiotics on fish growth and overall health.

Relative Growth Rate

The daily growth rates (DGR) of eels in the treatment ponds J1, J3, J5, and J7 were 0.98%, 0.5%, 0.57%, and 0.22%, respectively, with the highest growth seen in the pond combining probiotics and paddlewheels (J1). The statistical model explained 99% of the variation in RGR (adjusted to 98.4% for complexity), showing its reliability. Probiotics accounted for 82.8% of the variation (F = 38.616, p <0.001), while paddlewheels explained 92.3% (F = 95.359, p < 0.001), indicating both have strong individual effects. However, their interaction was not significant (F = 1.853, p = 0.211), meaning their combined use does not add additional benefits. The low MSE (0.010) confirms the model's accuracy.

However, the interaction between probiotic and paddlewheel does not have a meaningful impact on RGR (f = 1.853, p = 0.211), indicating their combined effect is not significant. The model's low error value (mse = 0.010) reflects its high accuracy in predicting RGR. Overall, the findings highlight the strong individual effects of probiotic and paddlewheel on growth, but their interaction does not add significant value.

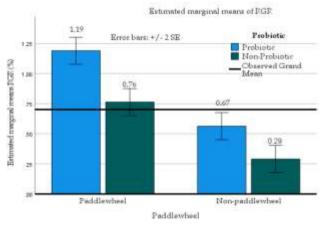


Figure 5. RGR (estimated marginal means) of Anguilla bicollor reared over 45 days

The bar chart shows how paddlewheels and probiotics influence RGR across four groups: with or without paddlewheels and with or without probiotics. The combination of paddlewheels and probiotics produced the highest RGR, exceeding 1.2, demonstrating their strong combined effect. On the other hand, the group without either treatment had the lowest RGR, below 0.5. Paddlewheels consistently improved RGR, whether probiotics were used or not, while probiotics provided an additional boost in both paddlewheel and non-paddlewheel groups.

The error bars (±2 SE) show some variability, but the differences between groups are clear. The overall average RGR, marked by the black line, highlights these contrasts. Probiotics and paddlewheels both significantly improve RGR, with the combination treatment achieving the highest RGR at 0.98%. This combined treatment outperformed probiotics-only, paddlewheels-only, and the control group. However, there was no significant difference between the probiotics-only and paddlewheels-only treatments.

Previous studies have shown that *Lactobacillus* probiotics in fish feed can enhance digestive enzyme activity. For example, Van Doan et al. (2021) reported increased lipase and protease activity in Nile tilapia fed with probiotics. Similarly, *Lactobacillus plantarum* significantly improved the RGR of Japanese eels compared to the control group. This probiotic enhances the activity of amylase, protease, and lipase, increases intestinal villi length (Kusumawaty et al., 2020; X. Wang et al., 2024; Mordenti et al., 2016).

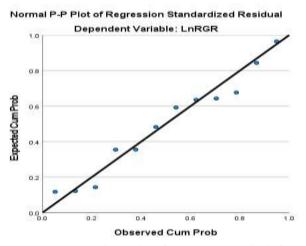


Figure 6. Normal p-p plot of regression residuals for probiotics and paddlewheel feeding effects on *Anguilla bicollor* growth rate

Probiotics enhance digestive enzyme activity by producing exogenous enzymes like lipase and protease. This aligns with Zheng et al. (2019), who highlighted the critical roles of proteases, lipases, and amylases in digestion and nutrient absorption in fish. Increased enzyme activity can boost overall metabolism. Similarly, Conforto et al. (2021) emphasized that probiotics and their exoenzymes enhance intestinal enzyme activity and stimulate the production of endoenzymes, improving nutrient digestibility and utilization (Harun et al., 2023; Y. J. Lin & Tzeng, 2018; Y. T. Lin et al., 2023). Exogenous enzymes, with their broad pH activity range, further aid digestion by improving substrate hydrolysis.

The Normal P-P Plot shows that the residuals from the regression model are close to the diagonal line, indicating the data meets the normality assumption. Minor deviations are expected, especially with a small sample size, and do not affect the model's reliability.

The regression analysis revealed a very strong relationship (R = 0.975) between the independent variables (paddlewheel and probiotic) and the dependent variable (LnRGR). The model explains 95.1% of the variance in LnRGR ($R^2 = 0.951$) with high statistical significance (p < 0.001). The adjusted R^2 of 0.940 confirms the model's robustness, and the standard error of 0.13351 reflects accurate predictions. Overall, the model is highly effective and reliable in explaining the variation in LnRGR.

Feed digestion in fish is influenced by three key factors: the presence of digestive enzymes, the level of enzyme activity, and the time feed interacts with these enzymes. Enzymatic digestion starts in the stomach, where pepsin, produced by gastric glands, breaks down proteins. Other enzymes, such as chitinase and lipase, are also involved. In the intestine, enzymes are produced by both the pancreas (the primary source) and secretory cells in the intestinal wall (Brett et al., 1979). Probiotics are often added to feed to enhance digestion. According to Fuller (1992) probiotics are live bacteria that improve intestinal health by balancing gut microbiota, inhibiting pathogens, and releasing enzymes that aid digestion. Some probiotics, being aerobics, require oxygen to function effectively

Feed Convert Ratio

The conversion ratio of eels reared in treatment ponds (J1, J3, J5 and J7) was 1.09, 1.48, 1.635 and 3.11 respectively.

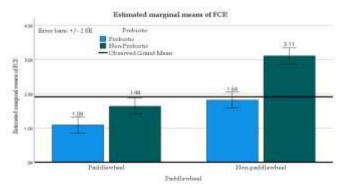


Figure 7. FCR (estimated marginal means) of Anguilla bicollor reared over 45 days



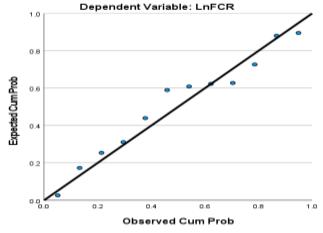


Figure 8. Normal p-p plot of regression residuals for probiotics and paddlewheel feeding effects on FCR of *Anguilla bicollor*

The analysis of the "tests of between-subjects effects" table, with log-transformed FCR (LnFCR) as the dependent variable, shows the model is highly effective, explaining 97.6% of the variation ($R^2 = 0.976$, Adjusted $R^2 = 0.964$). Probiotics significantly influence LnFCR (F = 38.533, p < 0.001), accounting for 82.8% of the variation, while paddlewheels have an even greater impact (F = 59.746, p < 0.001), contributing 88.2%. However, their interaction is not statistically significant (F = 0.911, p = 0.368), indicating no added advantage when combining probiotics with paddlewheels. The model's reliability is supported by a small error term (0.137) and mean square error (MSE = 0.017).

Treatment comparisons reveal that the probiotic + paddlewheels combination differs significantly from the probiotic-only and control treatments, but not from the paddlewheels-only treatment. Similarly, paddlewheels differ significantly from the control but not from the probiotic or probiotic + paddlewheels groups. Probiotic treatment differs significantly from the probiotic + paddlewheels and control groups but not from paddlewheels. Meanwhile, the control group differs significantly from all other treatments. These findings highlight distinct effects across treatments, with some overlap involving paddlewheels.

Regression analysis further confirms the significant impact of paddlewheel aeration and probiotics on LnFCR. With a strong R value of 0.958, 91.7% of the variation in LnFCR is explained by these factors, leaving only 8.3% to other variables. The model is highly reliable, supported by an adjusted R² of 89.8%, a low standard error of 0.13024, and strong statistical significance (F change = 49.630, p < 0.001). Additionally, the Normal P-P Plot shows that residuals are normally distributed, as most points align closely with the diagonal, confirming the model's validity. Overall, this robust model demonstrates that paddlewheels and probiotics independently improve feed efficiency, though their combination does not provide additional benefits.

The findings of this study are consistent with previous research by De Schrijver et al. (2000), which demonstrated that probiotics enhance an organism's ability to digest feed, resulting in improved growth performance. Studies on Japanese eels (J. S. Lee et al., 2015; S. Lee et al., 2018; Y. J. Lin & Tzeng, 2018) highlighted that supplementation with Bacillus subtilis WB60 (0.5)× 10^{7} CFU/g) combined with mannooligosaccharide (MOS) at 5 g/kg in feed significantly increased key growth parameters, such as final body weight, RGR, and FCR, when compared to the control group. These studies emphasize the role of probiotics in improving nutrient absorption, optimizing digestive enzyme activity, and ensuring more efficient utilization of feed.

The results suggest that eels supplemented with probiotics can absorb nutrients more effectively, allowing the feed to be converted into body mass rather than being wasted. This improvement in feed efficiency is reflected in better growth and lower FCR values. According to Al-Shorbagy et al. (2024), Cibuntu et al. (2023), Luo et al. (2013), and Yanti et al. (2022) FCR values are strongly linked to feed quality-lower FCR values indicate that the feed is of high quality and that the fish are efficiently utilizing it for growth. Moreover, the combination of probiotics and dietary additives helps to further enhance the gut environment, promoting a balance of beneficial gut bacteria and stimulating digestive enzyme production. This synergy between probiotics and dietary supplements ensures that the nutrients in the feed are utilized to their fullest potential, ultimately improving the growth performance and sustainability of aquaculture practices.

Interaction between Probiotics and Paddlewheels in Research

Although the study showed that combining probiotics and paddlewheels resulted in the best growth for eels, statistical analysis (ANOVA) found their interaction was not statistically significant (F = 0.606, p = 0.459). This means that while both factors independently contribute to growth, they don't directly enhance each other's effects. In simpler terms, their contributions are additive rather than synergistic, with each playing a unique role in supporting eel cultivation.

This research looked at how probiotics and paddlewheels work together to improve the growth of *Anguilla bicollor*. Probiotics help by improving feed efficiency—balancing gut microbiota, suppressing harmful pathogens, and producing digestive enzymes (Dewanti et al., 2022; Hatakeyama et al., 2023; Zheng et al., 2019). However, many probiotics, like *Lactobacillus sp. Bio7*, rely on oxygen to function effectively, which is where paddlewheels come in. Paddlewheels ensure adequate dissolved oxygen levels, which not only support the eels' metabolism but also enhance the activity of probiotics.

Together, these two factors create an effective system: probiotics improve digestion and feed efficiency, while paddlewheels maintain oxygen levels to support metabolism, probiotic function, and water quality. This combination makes them valuable tools for sustainable eel farming.

Probiotics, especially those based on aerobic bacteria like Lactobacillus sp., play a crucial role in aquaculture by offering several key benefits. They help regulate gut microbiota by reducing pathogenic microbial populations, creating a healthier intestinal environment (Y. Guo et al., 2025; Kusumawaty et al., 2023). Probiotics also improve feed efficiency by producing exogenous enzymes such as lipase, protease, and amylase, which enhance nutrient digestion and absorption (Van Doan et al., 2021). In addition to improving digestion, probiotics boost fish immunity, influencing both local defenses (intestinal mucosa, skin, and gills) and systemic immune responses (Grandi et al., 2003; He et al., 2021; Zhang et al., 2024). They also produce short-chain fatty acids, which promote the proliferation of epithelial cells in the gut, further enhancing nutrient absorption (Furuita et al., 2007; Gómez-Limia et al., 2021). However, it is important to note that probiotics, particularly aerobic strains like Lactobacillus, require sufficient oxygen to maintain their metabolic activity and function effectively.

The Role of Paddlewheels in Aquaculture Paddlewheels play a vital role in increasing dissolved oxygen (DO) levels in cultivation ponds, which is essential for eel metabolism. Maintaining optimal DO levels (>3 mg/L) supports aerobic respiration and organic matter oxidation, both of which are critical for eel growth and survival (Holden et al., 2022; Setyono et al., 2023). Additionally, paddlewheels provide the oxygen necessary to activate aerobic probiotics like Lactobacillus sp., enabling these beneficial bacteria to function effectively (Garnawansah et al., 2017) Aeration also helps improve water quality by preventing the buildup of harmful compounds like ammonia and nitrite, which can be toxic to eels (Haenen et al., 2010; Taufik et al., 2023; Yulia Sugeha & Suharti, 2009). Thus, while probiotics and paddlewheels individually contribute significantly to eel cultivation, their interaction does not amplify their effects. Instead, they complement each other in ensuring the overall health and growth of eels while maintaining an optimal aquatic environment.

Water Quality

The attached graph shows the observed changes in dissolved oxygen (DO) levels in ponds influenced by probiotics and paddlewheels. In pond J1, DO fluctuated between 3.6 and 7.74 mg/L, with the lowest level recorded at 19:00 WIB and the highest at 11:00 WIB. For pond J3, DO ranged from 3.9 to 7.5 mg/L, with the lowest at 07:00 WIB and the highest at 11:00 WIB.

Meanwhile, pond J5 showed a wider range, with DO values fluctuating from 0.7 to 7.1 mg/L, reaching the lowest at 03:00 WIB and the highest at 11:00 WIB. Similarly, in pond J7, DO ranged from 0.6 to 7.2 mg/L, with the lowest recorded at 03:00 WIB and the highest at 11:00 WIB.

| Parameters | Probiotics + Paddlewheels | Paddlewheels | Probiotics | Control | Standard |
|-------------|---------------------------|--------------|--------------|-------------|--------------------------------------|
| pН | 6.69 - 7.9 | 6.65-8.06 | 6.97 – 7.7 | 7.16 - 7.67 | 6.5 – 8.5 (Taufiq-Spj et al., 2020) |
| Nitrite | 0,06 - 0.41 | 0.13-2.59 | 0.034 - 0.24 | 0.03 - 0.29 | 0.10-0.78 (Scabra et al., 2016) |
| Ammonia | 0 - 0.14 | 0.02-0.44 | 0.02 - 1.48 | 0.04 - 0.84 | Max 1-2 mg/l (Engin & Carter, 2001) |
| Temperature | 28.6 - 30.8 | 28.7-31.9 | 29.8 - 32.8 | 29.6 - 33 | 28-33 (Luo et al., 2013a) |
| DO | 3 - 7.74 | 3.9 – 7.5 | 0.7 – 7.1 | 0.6 - 7.2 | Hipoxia < 2 (Le Moigne et al., 1986) |

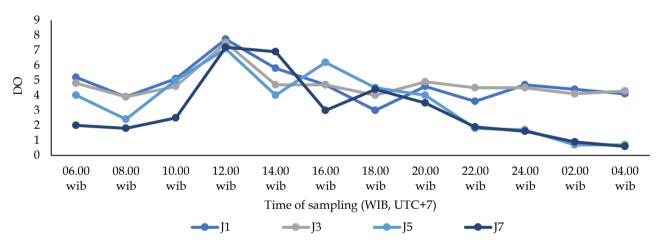


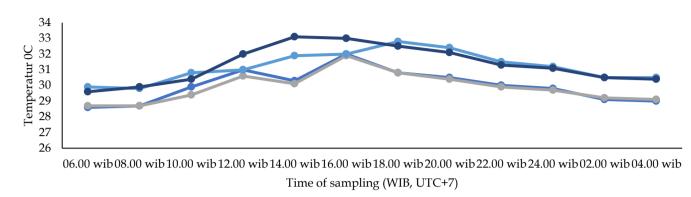
Figure 9. Graph of DO in Anguilla bicollor pond for 24 hours

During eel rearing, dissolved oxygen (DO) levels ranged from 3.4 to 5.7 mg/L. Adequate DO levels are essential for oxidizing waste, supporting food metabolism, and generating energy needed for eel growth and survival. Aeration systems play a critical role in maintaining these levels, reducing fish stress, and supporting metabolic processes (Fauzana et al., 2023). The observed DO levels remained within the normal range for eel cultivation, which is >3 mg/L (Budiardi et al., 2022; Saputra et al., 2024; Scabra et al., 2016).

Eels rely on their gills for respiration, which are in the branchial cavity and consist of four pairs of gills. Each gill is made up of filaments and lamellae, containing a network of blood vessels for oxygen exchange (Degani et al., 1985; Ellis & Smith, 1983). Uniquely, eels can also absorb oxygen through their skin, allowing them to survive temporarily in humid open-air environments (Taufiq-Spj et al., 2020; Toro et al., 2024). Their gill flaps, located as small slits behind the head, help retain moisture in the branchial cavity, ensuring survival outside water for short periods (Tesch, 1977). DO levels in biofloc systems are typically lower than in non-biofloc systems due to the activity of heterotrophic bacteria, which utilize oxygen to decompose organic matter. This decomposition process, along with nitrification and microbial respiration, leads to higher carbon dioxide levels and reduced DO in biofloc systems (Bossier & Ekasari, 2017; Chethurajupalli & Tambireddy, 2022; Kurniaji et al., 2023; Vinatea et al., 2023), While ammonia, nitrite, nitrate, and phosphate levels are not significantly different between biofloc and control systems, ammonia levels are generally lower when using probiotics early in the observation period. This is because heterotrophic bacteria in biofloc systems decompose ammonia more effectively, especially with a carbon-tonitrogen (C:N) ratio of 12-15, as noted by Tong et al. (2020). Nitrifying bacteria also help maintain low ammonia levels (Anand et al., 2014), which has been observed in several studies on biofloc systems (Anand et al., 2014; Rajkumar et al., 2016). However, in hypoxic environments (DO < 2 ppm), the prevalence of Shewanella putrefaciens in Anguilla significantly increases, potentially leading to disease outbreaks (Esteve et al., 2017). This highlights the importance of maintaining adequate oxygen levels to ensure eel health and reduce the risk of infections.

the paddlewheels in the pond are shown in the attached graph. The fluctuating range of temperature in pond J1 with a temperature value of 28.6^o- 32^o C (lowest at 05.00 WIB and highest at 17.00 WIB).

The results of observations of changes in water temperature in the pond with the effect of probiotics and



→ J1 → J3 → J5 → J7

Figure 10. Temperature graph of the Anguilla bicollor pond for 24 hours

In pond J3, temperatures ranged from 28.7°C to 31.9°C (lowest at 05:00 WIB, highest at 17:00 WIB), while J5 ranged from 29.9°C to 32.8°C (lowest at 05:00 WIB, highest at 19:00 WIB). In J7, temperatures varied between 29.6°C and 33.1°C, with the lowest recorded at 05:00 WIB and the highest at 13:00 WIB.

Fish growth depends on the balance between energy intake and expenditure, both of which are influenced by temperature (Elmi et al., 2023) As temperatures rise, metabolic rates increase, but growth rates decrease when temperatures exceed the species' optimum due to higher energy demands for maintenance metabolism (Luo et al., 2013b).

Eels have adapted to thrive at high optimum growth temperatures, allowing them to disperse widely and successfully. Newly hatched larvae from the Sargasso Sea are physiologically equipped to inhabit diverse geographic regions. However, exceeding the optimum temperature comes at a higher cost than undershooting it, as performance declines rapidly above the threshold. This makes maintaining a high optimum temperature advantageous for eels. High phenotypic plasticity has been proposed as an adaptive trait in species with broad ranges and panmictic populations, enabling them to cope with environmental variability (Ahn et al., 2012; Fukuda et al., 2009; Luo et al., 2013; Sato et al., 2006). For American eels, the optimum temperature for somatic growth is 27–28°C, which is significantly higher than average water temperatures in the northern part of their range. As a result, rising water temperatures due to climate change may enhance their growth potential (Holden et al. 2022).

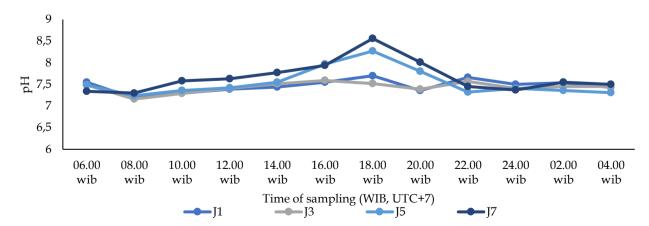


Figure 11. Graph of pH in Anguilla bicollor ponds for 24 hours

The results of observations of changes in the pH of water in the pond with the influence of probiotics and water wheels in the pond are shown in Figure 11. The pH fluctuations in the ponds varied across treatments. In J1, pH ranged from 7.18 to 7.7, with the lowest value recorded at 07:00 WIB and the highest at 17:00 WIB. In J3, pH values ranged from 7.16 to 7.59, with the lowest at 07:00 WIB and the highest at 15:00 WIB. For J5, the pH ranged from 7.24 to 8.27, and in J7, the range was 7.3 to 8.56, with the lowest and highest values in both ponds observed at 07:00 WIB and 17:00 WIB, respectively.

The pH of pond water fluctuates due to photosynthesis and respiration activities and is closely tied to water quality factors like ammonia toxicity, hydrogen sulfide (H₂S), alkalinity, and metabolic processes. High pH increases ammonia toxicity, while low pH raises H_2S toxicity, both of which can directly and indirectly affect fish health.

According to Amrullah et al. (2024) maintaining optimal water quality parameters—such as DO (5–6 ppm), pH (7.2–8.3), and ammonia (<0.05 ppm)—is crucial for fish health and the success of probiotics in feed. Probiotics reduce the buildup of organic waste in the water, stabilizing key parameters like DO, pH, and ammonia concentration (Fatmawati et al., 2023). Probiotic strains like *Lactobacillus plantarum* and *Lactobacillus bulgaricus* produce lactic acid and other organic compounds that lower pH and inhibit the growth of pathogenic bacteria (Susilo et al., 2023).

Conclusion

The effect of dietary probiotics and paddlewheels on the growth of eels reared for 45 days had a positive impact. The combination treatment resulted in the highest RGR: 1.19%) and the most efficient FCR: 1.09), with a 100% SR across all treatments. These findings highlight the additive, rather than synergistic, effects combination providing a comprehensive solution to challenges related to suboptimal water quality and feed inefficiency in eel farming. To optimize production in total aquaculture technology, it is necessary to conduct further studies on the use of various probiotics strain or combination and paddlewheels with intensification treatment of eels cultivation to obtain high production values and to find the ideal amount that boosts eel growth. Applying these techniques to more intensive systems like biofloc or recirculating aquaculture systems (RAS) might also reveal further benefits. These efforts aim to promote more sustainable and efficient eel farming practices.

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Investigation and experimental, G.G, (B.H) and (D.W); formal analysis, G.G, B.H and (D.H); resources, G.G, B.H and D.W; data curation, G.G, B.H and D.W: writing—original draft preparation, G.G, B.H and D.W; writing—review and editing, G.G, D.H, M.Z, D.P, I.R, and R.I: visualization, G.G, D.H, M.Z, D.P, I.R, and R.I: visualization, G.G, D.H, M.Z, D.P, I.R, and R.I; supervision, G.G, B.H, M.Z, D.H and D.W; project administration, W.H, R.K and B.H; funding acquisition, G.G, D.P, D.H, M.Z, D.W, B.H, I.R and R.I. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest

There are no conflicts of interest among any of the authors.(Rafiq et al., 2024)

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