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Effects of Functional Feed Based on Fermented Fishery Waste on Growth Performance and Protein Digestibility of Nile Tilapia *(Oreochromis niloticus)*

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ABSTRACT

Feed is the largest cost component in Nile tilapia culture, which calls for feed innovations that are both cost-efficient and functionally valuable. This study evaluated the effects of a functional feed based on fermented fishery waste on the growth performance, feed efficiency, and protein digestibility of Nile tilapia (Oreochromis niloticus). A completely randomised design was applied with four doses of a fermented biostarter (0, 5, 10, and 20 mL kg⁻¹ feed) and three replicates. Fish with an initial weight of approximately 0.5 g were reared for 35 days. The parameters measured were weight and length gain, feed conversion ratio (FCR), survival rate, proximate composition of the feed, and the apparent digestibility coefficient (ADC) of protein determined with chromic oxide as an inert marker. Increasing the biostarter dose raised the crude protein content of the feed and reduced its crude fibre content. The 20 mL kg⁻¹ treatment produced the highest weight gain (144.14 ± 0.40 g), the lowest FCR (0.65 ± 0.03), and the highest protein ADC (82.48%), whereas the highest survival was recorded at intermediate doses (5–10 mL kg⁻¹). Functional feed based on fermented fishery waste effectively improves nutrient utilisation and growth performance of Nile tilapia, and the optimal dose depends on whether growth or survival is the primary production objective.

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INTRODUCTION

Nile tilapia (*Oreochromis niloticus*) is one of the most widely farmed freshwater fish in the world and a key commodity in Indonesian aquaculture. Its relatively fast growth, wide environmental tolerance, omnivorous feeding habit, and acceptance by consumers have made it a cornerstone of affordable animal protein supply in developing countries (El-Sayed, 1999; Ng & Romano, 2013). Global aquaculture production has continued to expand and now supplies a growing share of aquatic animal food, with tilapia among the leading finfish groups by volume (FAO, 2024). In Indonesia, production has risen in line with demand from households seeking inexpensive protein, and intensification of pond, cage, and tank culture has made the species increasingly dependent on compounded feeds. The success of tilapia farming is therefore determined to a large extent by the quality, cost, and efficiency of the feed used.

Feed typically accounts for the largest share of operating costs in semi-intensive and intensive fish culture, commonly estimated at 50–70% of total production costs (Ng & Romano, 2013). Dependence on commercial feeds formulated with conventional ingredients, particularly fishmeal, raises production costs and exposes farmers to volatile ingredient prices. Fishmeal remains a preferred protein source because of its balanced amino acid profile, high digestibility, and palatability, yet its supply is constrained by the finite capacity of reduction fisheries and by competing demand from livestock and other aquaculture sectors (Tacon & Metian, 2008; Naylor et al., 2009). The

growing gap between demand and supply has stimulated extensive research on alternative protein sources, including plant proteins, terrestrial animal by-products, insects, and microbial biomass (Hardy, 2010; Hua et al., 2019). Most plant proteins, however, contain antinutritional factors such as protease inhibitors, phytate, non-starch polysaccharides, and gossypol, and they have unbalanced amino acid profiles that limit their inclusion in diets for many fish species (Francis et al., 2001). Innovations that lower feed costs while maintaining or improving nutrient utilisation and growth are therefore needed.

Using fishery waste as a feed resource is one strategy with the potential to reduce costs while supporting the principles of the circular economy. Fish processing generates large quantities of by-products, such as viscera, heads, frames, skin, and shellfish shells, which together can represent a substantial fraction of the landed weight and are often discarded or underused (Olsen et al., 2014). These materials contain appreciable amounts of protein, lipids, and minerals, and their valorisation as aquafeed ingredients can increase food production while reducing environmental pollution from waste disposal (Stevens et al., 2018). In coastal regions of Indonesia, residues from fish markets and processing, crab shells from swimming crab (*Portunus pelagicus*) processing, and seaweed residues are readily available at low cost. Direct use of such materials, however, is often constrained by low digestibility, variable composition, rapid spoilage, and the presence of indigestible or antinutritional components such as chitin and complex polysaccharides. Feed modification approaches, including bioencapsulation and fermentation, have therefore been developed to improve nutrient quality and enhance nutrient retention during digestion. Mulyani et al. (2023) showed that modifying natural feed through vitamin C bioencapsulation increased protein and energy retention in milkfish larvae, illustrating that changes in feed formulation and processing can markedly affect the bioavailability of nutrients to fish.

Fermentation is one of the oldest and most versatile processing technologies for improving the nutritional value of feed ingredients. During fermentation, microorganisms such as lactic acid bacteria, *Bacillus* spp., yeasts, and filamentous fungi secrete enzymes that hydrolyse proteins, carbohydrates, and lipids into simpler molecules that are more readily digested and absorbed (Dawood & Koshio, 2020). The process can degrade antinutritional factors, reduce crude fibre, increase the proportion of soluble protein, free amino acids, and small peptides, and generate organic acids and other bioactive metabolites that improve palatability and intestinal health (Dawood & Koshio, 2020). The hydrolysis of fish proteins into peptides and free amino acids has been shown to enhance their functional and nutritional properties, which explains the interest in hydrolysed and fermented fish by-products as feed ingredients (Kristinsson & Rasco, 2000). In Nile tilapia, solid-state fermentation of soybean meal with *Saccharomyces cerevisiae* increased its crude protein and hydrolysed amino acid content while lowering phytic acid and trypsin inhibitor levels, thereby improving its nutritive value in practical diets (Hassaan et al., 2015). Similarly, fermentation of soybean and cottonseed meals with *Aspergillus oryzae* increased antioxidant activity and detoxified gossypol in tilapia diets (Lim & Lee, 2011).

Beyond improving ingredient quality, fermented feeds can act as functional feeds, delivering live or inactivated microorganisms and their metabolites that confer health benefits beyond basic nutrition. Functional feed additives such as probiotics, prebiotics, organic acids, and enzymes are increasingly used to enhance growth, feed efficiency, and disease resistance in aquaculture (Encarnação, 2016). Probiotic bacteria can improve

digestion by producing exogenous enzymes, competitively excluding pathogens, and modulating the gut microbiota and immune responses (Hai, 2015). Early work on Nile tilapia demonstrated that dietary supplementation with *Streptococcus faecium*, *Lactobacillus acidophilus*, and *S. cerevisiae* improved growth and feed utilisation (Lara-Flores et al., 2003). Fermented feeds thus combine two potentially complementary effects: an improvement in the intrinsic nutritional quality of the ingredients and the introduction of beneficial microbes and metabolites into the digestive tract.

Despite these advances, several knowledge gaps remain. Most studies on fermented aquafeeds have focused on plant protein sources, particularly soybean, cottonseed, and other oilseed meals, whereas fermentation of locally available fishery waste into a biostarter for tilapia feed has received comparatively little attention. In addition, relatively few studies have simultaneously examined the changes in proximate composition of the feed, the apparent digestibility of protein, and the resulting growth and feed conversion responses of the fish within the same experiment. Linking these levels is important because improvements in growth can only be attributed to better nutrient availability if they are accompanied by measurable changes in feed composition and digestibility (Glencross et al., 2007). Information on the dose–response relationship of fermented biostarters is also limited, even though an excessive dose could affect palatability, gut microbial balance, or fish health and thereby offset the nutritional benefit. Determining an appropriate dose is therefore essential for translating fermentation technology into practical farm feeds.

This study therefore aimed to evaluate the effects of functional feed based on fermented fishery waste, applied at different biostarter doses, on the growth performance, feed efficiency, survival, feed proximate composition, and protein digestibility of Nile tilapia (*Oreochromis niloticus*). It was hypothesised that increasing the dose of fermented biostarter would improve feed protein content and protein digestibility, leading to higher growth and lower feed conversion ratios. The findings are expected to provide a scientific basis for developing low-cost functional feeds from local fishery waste and to support more efficient and sustainable tilapia culture.

METHODS

The experiment followed a completely randomised design with four treatments and three replicates. The treatments were doses of a biostarter produced by fermenting fishery waste, applied to the feed at 0 mL kg^{-1} (control), 5, 10, and 20 mL kg^{-1}. Nile tilapia juveniles with a mean initial body weight of approximately 0.5 g were stocked at 50 fish per rearing unit and cultured for 35 days. Fish were fed at 5% of biomass per day, divided into three daily rations. Water quality was maintained within the optimal range for Nile tilapia throughout the trial (Boyd & Tucker, 1998). Body weight and total length were measured at the start and end of the experiment, and the number of surviving fish was recorded. Growth performance and feed utilisation were calculated using standard formulas (Hopkins, 1992): weight gain (g) = final mean weight − initial mean weight; length gain (cm) = final mean length − initial mean length; feed conversion ratio (FCR) = total feed consumed (g) ÷ total biomass gain (g); and survival rate (%) = (final number of fish ÷ initial number of fish) × 100.

The proximate composition of each experimental feed, comprising crude protein, crude fibre, and crude lipid, was determined following standard AOAC (2005) procedures. Apparent protein digestibility was assessed with chromic oxide (Cr_2O_3) as an inert marker incorporated into the diets; chromic oxide in the feed and faeces was

quantified using the method of Furukawa and Tsukahara (1966), and faecal collection followed the principles described by Austreng (1978). The apparent digestibility coefficient (ADC) of protein was calculated according to Cho et al. (1982) as ADC (%) = 100 × [1 − (% Cr_2O_3 in feed ÷ % Cr_2O_3 in faeces) × (% protein in faeces ÷ % protein in feed)]. Data on growth, FCR, survival, proximate composition, and ADC were tested for normality and homogeneity of variance and then analysed by one-way analysis of variance (ANOVA); when significant differences were detected, means were separated using Tukey's honestly significant difference test at a 95% confidence level (Zar, 2010).

RESULTS AND DISCUSSION

Growth performance

Functional feed based on fermented fishery waste significantly affected the growth of Nile tilapia ($p < 0.05$). Both weight gain and length gain increased consistently with increasing biostarter dose, and the 20 mL kg^{-1} treatment produced the highest weight gain (144.14 ± 0.40 g). The growth trajectories presented in Figure 1 show that the separation between treatments widened progressively over the rearing period, with the 20 mL kg^{-1} group consistently above the lower doses and the control. Figure 2 shows a parallel increase in body length, indicating that the additional nutrients supplied by the fermented feed supported proportional structural growth rather than mass accumulation alone. A dose-dependent improvement in growth of this kind is consistent with reports that fermented ingredients enhance growth in fish by improving nutrient availability, palatability, and gut function (Dawood & Koshio, 2020). Comparable growth responses have been observed in Nile tilapia fed yeast-fermented soybean meal (Hassaan et al., 2015) and *Aspergillus*-fermented soybean and cottonseed meals (Lim & Lee, 2011).

Physiologically, the improvement in weight and length gain can be explained by the increased availability of free amino acids and low-molecular-weight peptides generated by microbial proteolysis during fermentation. Hydrolysed fish proteins are absorbed more rapidly than intact proteins and can be used directly for body protein synthesis, which improves protein retention and growth efficiency (Kristinsson & Rasco, 2000; Chalamaiah et al., 2012). Microbial fermentation also degrades complex structural carbohydrates, and the resulting reduction in crude fibre lessens the physical barrier to enzymatic digestion in the gut. Because tilapia have a limited capacity to digest fibre, a lower fibre fraction allows a greater proportion of the ingested dry matter to be digested and absorbed (NRC, 2011). In addition, live microorganisms and their metabolites introduced with the fermented feed may stimulate digestive enzyme activity and favour a beneficial gut microbiota, as reported for probiotic supplementation in tilapia (Lara-Flores et al., 2003; Hai, 2015). The combination of higher nutrient density, improved digestibility, and probiotic effects is therefore likely to underlie the superior growth observed at the highest biostarter dose.

Feed conversion ratio

Feed utilisation efficiency improved significantly in the fermented-feed treatments, as reflected by lower FCR values compared with the control ($p < 0.05$; Figure 3). The 20 mL kg^{-1} treatment produced the lowest FCR (0.65 ± 0.03), meaning that a larger proportion of the feed supplied was converted into fish biomass as feed quality improved. The reduction in FCR at higher doses indicates that fermentation lowered the crude fibre fraction and the content of compounds that impair digestion, thereby

enhancing digestive and absorptive efficiency. This finding agrees with the principle that diet quality, particularly protein digestibility and amino acid availability, is a key determinant of feed conversion efficiency in omnivorous species such as tilapia (Ng & Romano, 2013; El-Sayed, 1999). Degradation of antinutritional factors is a well-documented benefit of fermentation, and its removal improves protein utilisation in fish fed alternative ingredients (Francis et al., 2001; Dawood & Koshio, 2020). From a practical standpoint, a lower FCR translates directly into reduced feed costs per unit of fish produced, which is the main economic justification for adopting fermented functional feeds on farms (Hua et al., 2019).

Survival rate

Survival of Nile tilapia remained high in all treatments throughout the rearing period, indicating that the fermented feed had no adverse effect on fish viability (Figure 4). The intermediate doses (5–10 mL kg^{-1}) produced the highest survival, whereas survival at the highest dose was comparable to that of the control. This pattern suggests that, although the highest dose maximised growth, intermediate doses provided a better balance between growth promotion and the physiological tolerance of the fish to the feed. Moderate levels of beneficial microorganisms and their metabolites can strengthen intestinal barrier function and non-specific immunity, improving resistance to stress and opportunistic infections (Ringø et al., 2020). At higher inclusion levels, however, a greater microbial load, accumulation of organic acids, or shifts in feed palatability may impose a physiological burden or alter gut microbial balance, so that the health benefit no longer increases with dose (Wang et al., 2008; Encarnação, 2016). Because the survival differences among treatments were small and were not accompanied by measurements of immune or health indicators, this interpretation should be confirmed in longer trials that include physiological and immunological endpoints.

Feed proximate composition and protein digestibility

Proximate analysis showed that fermentation significantly increased the crude protein content of the feed and reduced its crude fibre content. These changes were accompanied by an increase in the apparent digestibility coefficient of protein, which rose with increasing biostarter dose ($p < 0.05$) and reached its highest value, 82.48%, in the 20 mL kg^{-1} treatment. The relative increase in crude protein can be attributed partly to the degradation of carbohydrates and fibre by microbial activity, which concentrates the remaining nitrogenous fraction, and partly to the contribution of microbial biomass protein itself (Dawood & Koshio, 2020). A similar enrichment of crude protein and hydrolysed amino acids, together with a reduction in antinutritional factors, was reported for soybean meal fermented with *S. cerevisiae* for Nile tilapia (Hassaan et al., 2015).

The positive association between feed protein quality and protein digestibility confirms that improving the nutritional quality of the feed directly enhances protein utilisation by the fish. Biologically, fermentation lowers digestive barriers and increases the fraction of soluble protein and small peptides, so that nutrients are used more efficiently and growth performance improves (Kristinsson & Rasco, 2000; Chalamaiah et al., 2012). Linking composition, digestibility, and growth in this way is important because digestibility data provide the mechanistic link that allows growth improvements to be attributed to changes in the feed rather than to uncontrolled factors (Glencross et al., 2007). The consistent upward trend in weight gain with increasing

biostarter dose, together with the parallel increases in feed protein content and protein ADC, indicates that nutrients in the fermented feeds were more readily digested and used for tissue synthesis. These findings support the valorisation of fishery waste as a functional feed resource, which offers both economic and environmental benefits by converting low-value by-products into a feed input that improves production efficiency (Olsen et al., 2014; Stevens et al., 2018).

CONCLUSION

Functional feed based on fermented fishery waste effectively improved the nutritional quality of the feed, the apparent digestibility of protein, and the growth and feed efficiency of Nile tilapia (*Oreochromis niloticus*). Increasing the biostarter dose raised the crude protein content and lowered the crude fibre content of the feed, and these compositional changes were accompanied by progressively higher protein digestibility, greater weight and length gain, and lower feed conversion ratios. The 20 mL kg^{-1} dose produced the best overall performance, with the highest weight gain (144.14 ± 0.40 g), the lowest FCR (0.65 ± 0.03), and the highest protein ADC (82.48%), whereas the intermediate doses of 5–10 mL kg^{-1} yielded the highest survival. The optimal dose therefore depends on the production objective: 20 mL kg^{-1} is recommended where growth and feed efficiency are the priority, and 5–10 mL kg^{-1} where survival and stock stability are paramount. Converting locally available fishery waste into a fermented functional feed offers a practical route to lowering feed costs, reducing waste, and improving the sustainability of tilapia culture. Further studies should evaluate longer culture periods, larger fish sizes, economic returns, and the effects of fermented feeds on gut microbiota, immune status, and disease resistance under farm conditions.

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