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A preliminary study on the effect of enriching feed with fish oil on the growth and survival rate of climbing perch *Anabas testudineus*

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ABSTRACT

The climbing perch (Anabas testudineus) is an economically valuable freshwater fish. Relatively slow growth has been a challenge in the domestication of this species in Indonesia. Nutrition, including feed lipid content, is one factor affecting growth. This study examined the effect of enriching feed with fish oil on the growth and survival of climbing perch (A. testudineus) fingerlings. The research was carried out at the Water Quality and Aquatic Biology Laboratory, Faculty of Animal Husbandry and Fisheries, Tadulako University, Palu, Central Sulawesi, Indonesia from 17 December to 18 January 2020. A completely randomized design (CRD) was used with 4 treatments and 5 replicates. The fish oil feed enrichment treatments were: A (control, 0%); B (1%); C (2%) and D (3%). Water quality remained within the optimum range throughout the research period. Over the month, climbing perch absolute weight gain ranged from 2.4±0.981 g (A) to 3.4 ± 0.836 g (D), while growth in length ranged from 0.404±0.092 cm (A) to 0.504±0.071 cm (D); however, the differences were not statistically significant (P>0.05). The survival rate of climbing perch over the one month study period was 100% under all treatments, indicating that basic nutritional needs were met. Enrichment of a commercial feed with fish oil (Scott's emulsion) at rates of 1-3% did not provide a significant benefit in terns of climbing perch fingerling growth.

Introduction

The climbing perch (*Anabas testudineus*) is an economically valuable fish that lives in swamps and rivers. This fish is relatively expensive compared to introduced fisheries commodities such as carp, catfishes, tilapia and pacu. According to Kottelat *et al.* (1993), the climbing perch can inhabit inland waters such as lakes, rivers and marshes, including fresh and brackish water bodies. The climbing perch is widely distributed in Indonesia, including the islands of Sumatra (Ahmad and Fauzi, 2010; Jamsari *et al.*, 2010; Putra *et al.*, 2016), Java (Roberts, 1993), Kalimantan (Ahmadi, 2019; Mustakim *et al.*, 2018; Slamat *et al.*, 2012) and Sulawesi (Ndobe *et al.*, 2019 & 2020; Parenti *et al.*, 2014).

Wild climbing perch populations are increasingly under pressure from habitat degradation as well as fishing pressure (Ndobe *et al.*, 2014 and 2019; Slamat *et al.*, 2012). The domestication of this species has begun in several regions of Indonesia (Ahmad and Fauzi, 2010; Bijaksana and Balantek, 2012). In Sigi District, Central Sulawesi, freshwater aquaculture is a local government priority and a domestication program has been underway since 2014 (Ndobe *et al.*, 2019). Research to date under this program has included basic life history parameters (Ndobe *et al.*, 2019 and 2020) as well as various aspects of climbing perch husbandry. Captive breeding to produce climbing perch seed has been successful in experimental, government and community-based hatchery settings in Sigi District, as in various other regions and countries, especially Bangladesh and India (Sarkar *et al.*, 2005; Sarkar *et al.*, 2015). Many factors can influence climbing perch growth and survival; these include stocking density (Khatune-Jannat *et al.* 2012; Uddin *et al.*, 2016); feed (Mahmood

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et al., 2004); genetic factors (e.g. strains) (Alam *et al*., 2014; Chakraborty and Haque, 2014); other internal factors including individual and group behaviour (Zworykin, 2018); and environmental conditions (Mondal *et al*., 2010). However, one challenge to the development of profitable climbing perch culture, especially in the Indonesian socio-economic context, is the relatively long grow-out period (Akbar, 2012).

According to Yulintine (2012), the tendency of climbing perch to exhibit slow growth and low survival in comparison to other swamp and river-dwelling fish commodities is thought to be related to inappropriate feed that fails to meet climbing perch nutritional needs. Providing feed appropriate to meet these needs should stimulate optimal growth and development, thereby improving productivity. Inappropriate feed is also one of the factors which can result in poor fish quality. For example, in pelleted feed the pellets may be too large, the nutritional content may be inadequate, or the feed type may be inappropriate for the fish being cultured (Hossain *et al*., 2012; Sarker *et al*., 2016).

The addition of fish oil to the feed could potentially play an important role in supplying nutrients required for fish growth in sufficient concentrations, because fish oil contains beneficial fatty acids known to promote fish growth as well as human health (Fard *et al*., 2019; Shepherd and Bachis, 2014). Fish oil contains about 25% saturated fatty acids and 75% unsaturated fatty acids, as well as high amounts of vitamin A and vitamin D, two fat-soluble vitamins (Komariyah, 2009 in Munisa *et al.,* 2015). One way to increase the beneficial fatty acid content of fish feed is enrichment with emulsions containing fish oil such as Selco emulsion or Scott's emulsion (Koldewey, 2005; Wati and Imanto, 2009).

In research conducted on the enrichment of eel (*Anguilla bicolor*) feed with fish oil, the highest increase in growth rate has been reported from the addition of fish oil at rates of 2% (Perdana *et al*., 2016), 3% (Ismayanti *et al*., 2019) and 5% (Mukti *et al*., 2014). This study aimed to evaluate the effect of feed enrichment with fish oil, specifically in the form of Scott's emulsion (a product readily available to fish farmers in the Sigi region), on the growth and survival rate of climbing perch (*Anabas testudineus*) fingerlings.

Materials and Methods

Site and time

This research was carried out in the Aquaculture Laboratory, Faculty of Animal Husbandry and Fisheries, Tadulako University in Palu, Central Sulawesi, Indonesia. The research lasted for one month from 18th December 2019 to 17th January 2020.

Experimental animals and feed

The experimental fish, climbing perch (*Anabas testudineus*) with a total length of 6-10 cm and body weight of 6-14 g, were obtained from the wild population in Maranata Village, Sigi District, Central Sulawesi with the assistance of local fish farmers. The fish were transported to the laboratory in plastic bags partially filled with water and then filled with oxygen before being sealed. The bags were placed in styrofoam boxes with ice to maintain a suitable temperature during transport. These boxes were transported by motorcycle from Maranata to the Tadulako University Aquaculture Laboratory.

The commercial feed (HI-PRO-VITE 781-1) was used in this study; this floating feed has an advertised formulation with a minimum crude protein content of 31%, minimum crude lipid content of 5% and maximum content of 8% crude fibre, 13% ash and 12% water (CP-Prima 2020). Scott's emulsion, with a fish oil content of 200g/L, was used as a source of lipid (fish oil) enrichment. The oil was added to the feed in the appropriate ratio for each treatment, thoroughly mixed to ensure even coating and absorption, and then sun-dried.

Experimental design and procedures

A completely randomized design (CRD) with 4 treatments and 5 replicates (20 experimental units) were used in this study. The fish oil (Scott's emulsion) feed enrichment treatments were: A (control, 0%); B (1%); C (2%) and D (3%). The experimental fish (N = 100) were acclimated for 7 days before stocking in 15L round black plastic basins, each filled with 10L of freshwater and equipped with aeration. The climbing perch were stocked at one fish per 2L water (5 fish in each basin). The fish were fed on experimental diets twice a day (8 a.m. and 5 p.m.) for 30 days, at a feeding rate of10% of total fish body weight. Water quality was monitored at each feeding time.

Measured parameters

Absolute length gain, weight gain and survival rate were calculated for each experimental unit using the following equations:

Length gain = mean total length at day 30 – mean total length at day 0 (in cm)

Weight gain = mean body weight at day 30 – body weight at day 0 (in g)

Survival rate = 100 · number of live fish on day 30 · (number of live fish on day 0)$^{-1}$ (in %)

Data analysis

Analysis of variance (ANOVA) was applied to evaluate the effect of the treatment, when the effect is significant, a post-hoc least significant difference (LSD) test was applied. Survival rate and water quality were analysed descriptively.

Results

The analysis of variance (ANOVA) at the 95% confidence level did not show any significant effect of fish oil feed enrichment on the weight gain or total length gain of climbing perch (P>0.05). Climbing perch (*A. testudineus*) fingerling mean weight gain over the one month study period was in the range of 2.4-3.481 g (Figure 1a) while mean length was in the range of 0.404-0.504 (Figure 1b). The relatively high standard deviation values of weight gain (0.347-0.981, equivalent to 11-40%) and total length gain (0.048-0.092, equivalent to 10-23%) reflect considerable between-replicate variation.

The survival rate of the climbing perch fingerlings over the study period was 100% in all treatments. The overall water quality parameter ranges were: temperature 26.7-29.3^{o}C, pH 6.13-8.43 and dissolved oxygen (DO) 5.1-7.3 mg/L (Table 1).

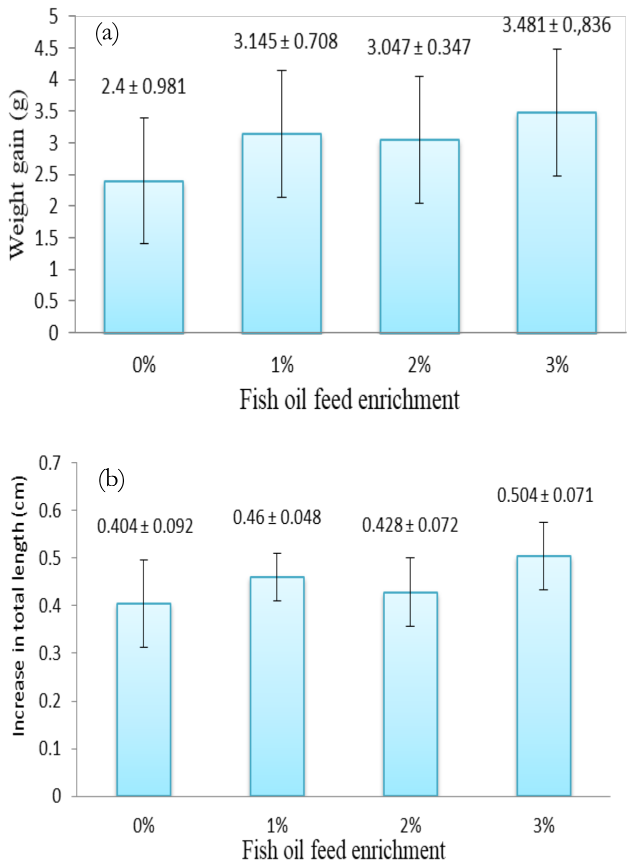

Figure 1. Net growth of climbing perch (*Anabas testudineus*) over the one month study period: (a) Weight gain; (b) Total length gain.

Table 1. Water quality parameters in the climbing perch (*Anabas testudineus*) experimental culture units.

No.	Treatment*	Water quality parameter range		
		Temperature (°C)	pH	DO (mg/L)
1.	A	26.7-29	6.33-8.13	5.2-6.4
2.	B	26.7-29.1	6.13-7.99	5.2-7.2
3.	C	26.5-29.3	6.40-8.08	5.1-7.0
4.	D	26.7-29.1	6.15-8.43	5.1-7.3

* Fish oil feed enrichment: A = control, 0%; B = 1%; C = 2% and D = 3%

Discussion

Climbing perch can take a year or more for the fish stocked to reach marketable size (75 to 100), and relatively slow growth can be one of the main challenges in climbing perch aquaculture (Akbar, 2012). Feed formulation can affect the growth rate of *Anabas testudineus*, including protein content (Hossain *et al.*, 2012) and the lipid/protein ratio (Ali *et al.*, 2012). The mean climbing perch length gain and weight gain were higher for all treatments (B, C and D) with fish oil enrichment compared to treatment A (control, no fish oil enrichment), even though there was high between-replicate variability and the differences were not statistically significant (P>0.05). Both length and weight gains considerably higher than in this study have been reported using formulations with lipid content of 8-12% (Alam *et al.*, 2010; Hossain *et al.*, 2012). In eels (*Anguilla bicolor*), Ismayanti *et al.* (2019) found that seed reared on feed enriched with 3% fish oil performed significantly better than the control formulation in terms of specific growth and survival rate, as well as two parameters not measured in this study (feed efficiency and fat retention). In the same species, Mukti *et al.* (2014) trialled feed enrichment at 0%, 5%, 10% and 15% and found that eels grew fastest with the addition of 5% fish oil, but at higher enrichment levels eel growth actually decreased, indicating that, above the optimum range, higher proportion of fish oil may be ineffective or even counter-productive.

There are growing concerns over the use of fishmeal and fish oil in fish feed (Misund *et al.*, 2017; Shepherd and Bachis, 2014), especially with respect to the sustainability of current exploitation rates of target fish stocks and competition with their use for human consumption (Hua *et al.*, 2019). There is a growing body of research on alternatives to fishmeal and fish oil (Oliva-Teles *et al.*, 2015). Potential fish oil replacements in fish feed formulations include the use of fatty acids derived from plants, including oil-rich algae (Norambuena *et al.*, 2015) and other organisms such as microbes (Sprague *et al.*, 2017). Research on the use of such feed ingredients as well as other potential ways to enrich the nutritional quality and/or digestibility of climbing perch feed seems worth exploring in future research on the domestication of this valuable food fish.

We speculate that nutritional limitations other than the lipid content of the commercial feed were not supplied by the enrichment medium used (fish oil in the form of Scott's emulsion). In particular, the protein level recommended for *A. testudineus* fry is 40%, with 30% as the minimum effective protein content (Hossain *et al*., 2012). Proximate analysis of a different batch of the "781" feed used in this study gave lipid values within the specified range but crude protein around 20% (Rantedongi, 2019), well below the 31% minimum specified by the manufacturers (CP Prima 2020). While the lack of statistical significance coupled with 100% survival rate indicates that the commercial (control) feed and the feed enriched with fish oil were all able to supply proteins and other nutrients needed by the fish in order to survive, growth was not as uniform or as rapid as could be desired. It is possible that the commercial feed was already close to the desired protein content, which could explain why the enrichment with fish oil did not have a statistically significant effect, even though the mean values for climbing perch growth in length and weight were higher for the treatments than for the control feed. Conversely, it is also possible that the protein content of all feed formulations in this study may have been below the optimal range for climbing perch, thereby limiting potential positive effects of enrichment.

In addition to the formulation of the feed treatments, this high survival rate is thought to be related to the low stocking density used during the research, the design of the experimental units, including the maintenance of appropriate water quality parameters. Despite fluctuations, throughout the study period the three water quality parameters measured (pH, dissolved oxygen and temperature) remained well within the ranges tolerated by climbing perch, and indeed within the ranges recorded at the site of origin of the fingerlings in Sigi District (Ndobe, unpublished data 2013). Climbing perch can tolerate dissolved oxygen levels as low as 3.5 mg/L (Chowdhury *et al*., 2014), while in this study the lowest recorded value was 5.1 mg/L. The observed temperature range (26.7-29.3^{o}C),was influenced mostly by the diel cycle in ambient temperature, and remained well within the wide thermal niche (around 13-40^{o}C) reported for climbing perch (Sarma *et al*., 2010), while the pH range (6.13-8.43) was within the range recorded for wild populations in Indonesia (Ernawati *et al*., 2009).

The results are consonant with the view that fish growth and survival are influenced by many internal factors such as the fish species, genetic traits, ability to use food and resistance to disease, but also by environmental factors such as water quality, feed, and room to move or stocking density (Chakraborty and Nur, 2012; Jonsson and Jonsson, 2014; Uddin et al., 2016). One possible confounding factor could be individual and group behavioural differences. Individual climbing perch can display markedly different behaviours, including with regard to feeding and movement (energy expenditure), and the interactions between individuals and group dynamics can also differ (Zworykin, 2018). In view of the relatively small number of fish (5) per replicate, such factors could have a considerable effect, and could be one reason for the high inter-replicate variability observed. However, the survival of all experimental fish indicates that not only were all feed formulations acceptable but also that the culture conditions were adequate.

Conclusions

Enrichment of a commercial feed with fish oil (Scott's emulsion) at rates of 0-3% did not produce a significant effect (P>0.05) on weight gain, total length gain and survival rate of climbing perch (*Anabas testudineus*) fingerlings over a one month culture period.

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