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GROWTH, PROTEIN EFFICIENCY RATIO AND PROTEIN RETENTION OF G6 TRANSGENIC MUTIARA CATFISH WITH MIXED FEEDING AT DIFFERENT RATIO OF COMMERCIAL FEED AND REBON SHRIMP FLOUR

Pertumbuhan, Rasio Efisiensi Protein dan Retensi Protein Ikan Lele Mutiara Transgenik G6 dengan Pakan Campuran Pada Rasio Pakan Komersil dan Tepung Udang Rebon yang Berbeda

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ABSTRACT

This study aims to evaluate the growth of transgenic G6 mutiara catfish using a mixture of commercial feed and rebon shrimp meal with different ratios between treatments. This research was carried out in May-August 2023, in the Lab. Aquaculture Gd.4 Faculty of Fisheries and Marine Sciences, Padjadjaran University. This research used the experimental method Completely Randomized Design (CRD) with four treatments and three replications. Treatment in the form of a mixture of commercial feed and rebon shrimp meal with a ratio of 50:50 (A), a ratio of 65:35 (B) and a ratio of 80:20 (C) was given to G6 transgenic mutiara catfish and the control treatment (65:35 ratio) given to sangkuriang catfish (treatment B*). Maintenance was carried out for 56 days with feeding three times a day. The parameters observed included growth in absolute weight (Wg), feed conversion ratio (FCR), protein efficiency ratio (PER), and protein retention (RP) which were analyzed using Analysis of Varience with 95% confidence level if significantly different followed by Duncan's test using SigmaPlot 15.0 software. The research results showed that the addition of commercial feed and rebon shrimp flour with a ratio of (80:20) had an influence on growth performance as indicated by an increase in absolute weight gain (687,53 g), FCR (0,64), protein efficiency ratio (18,7) and protein retention (46,43%.) Water quality values during rearing were still within tolerable limits for the growth of G6 transgenic mutiara catfish.

Keywords: Commercial Feed, Growth, Protein Efficiency, Rebon Shrimp Meal, Transgenic Catfish

ABSTRAK

Penelitian ini bertujuan untuk mengevaluasi pertumbuhan ikan lele mutiara transgenik G6 menggunakan pencampuran pakan komersil dan tepung udang rebon dengan rasio yang berbeda antar perlakuan. Penelitian ini dilaksanakan pada bulan Mei-Agustus 2023, di Lab.

Akuakultur Gd.4 Fakultas Perikanan dan Ilmu Kelautan Universitas Padjadjaran. Penelitian ini menggunakan metode eksperimental Rancangan Acak Lengkap (RAL) dengan empat perlakuan tiga kali ulangan. Perlakuan berupa pakan campuran pakan komersil dan tepung udang rebon dengan rasio 50:50 (A), rasio 65:35 (B) dan rasio 80:20 (C) diberikan pada ikan lele Mutiara transgenic G6 serta perlakuan kontrol (rasio 65:35) diberikan pada ikan lele Sangkuriang (perlakuan B*). Pemeliharaan dilakukan selama 56 hari dengan pemberian pakan tiga kali sehari. Parameter yang diamati meliputi pertumbuhan bobot mutlak (Wg), rasio konversi pakan/feed convertion ratio (FCR), rasio efisiensi protein (PER), serta retensi protein (RP) yang dianalisis menggunakan ANOVA/Analysis of Varience dengan tingkat kepercayaan 95% apabila berbeda nyata dilanjutkan dengan uji Duncan menggunakan software SigmaPlot 15.0. Hasil penelitian menunjukan penambahan pakan komersil dan tepung udang rebon dengan rasio (80:20) memberikan pengaruh terhadap performa pertumbuhan ditunjukkan oleh peningkatan nilai pertambahan bobot mutlak (687,53 g), FCR (0,64), rasio efisiensi protein (18,7) dan retensi protein (46,43%.) Nilai kualitas air selama pemeliharaan masih dalam batas ditoleransi untuk pertumbuhan ikan lele mutiara transgenik G6.

Kata Kunci: Pakan Komersil, Pertumbuhan, Efisiensi Protein, Tepung Udang Rebon, Lele Transgenik

INTRODUCTION

Mutiara catfish is a fish resulting from breeding research carried out by the Sukamandi Fish Breeding Research Institute (BPPI) in 2014. This fish showed an increase in weight gain of up to 21% compared to other types of catfish (Iswanto, 2014). However, this high growth rate cannot be maintained in the next generation, to increase growth performance again, this is done by utilizing the transgenesis process (Buwono *et al*., 2016). The transgenesis technology process basically transfers certain superior genes to related fish which have the advantage of a faster growth rate compared to non-transgenic catfish.

Apart from improving genetics, one of the factors to accelerate fish growth performance is by changing the feed formula needed by fish as a source of energy and growth. Baihaqi *et al*. (2020) stated that feed is the largest variable cost in the production process, namely around 60-70% of production costs. To reduce the production period and feed costs, alternative ingredients are needed which are substituted to replace some of the protein from commercial feed. Rebon shrimp flour is the ingredient of choice for commercial feed mixtures because it has complete nutritional content to support fish growth. The protein content of rebon shrimp flour is 52.35%, close to fish meal protein of 55% (Satyani & Sugito, 1997). Rebon shrimp flour protein contains complete essential amino acids (methionine, arginine, threonine, tryptophan, histidine, isoleucine, leucine, lysine, valine, phenylalanine) which are needed for fish growth, and has attractant properties that stimulate fish appetite.

The treatment ratio of adding rebon shrimp flour to commercial feed has not been carried out for the growth of transgenic catfish and non-transgenic catfish (Sangkuriang catfish). It is necessary to test the use of rebon shrimp flour on growth, feed conversion ratio, protein efficiency ratio and protein retention for both types of catfish.

METHODS

Time and Place

The research was carried out from May to August 2023, in the Aquaculture Laboratory Building 4, Faculty of Fisheries and Marine Sciences, Padjadjaran University. Proximate analysis of test fish was carried out at the Ruminant Animal Nutrition and Animal Food Chemistry Laboratory, Faculty of Animal Husbandry, Padjadjaran University.

Tools and Materials

The materials used consisted of 40 transgenic G6 mutiara catfish and 15 sangkuriang catfish measuring 11 cm long with a weight of around 13 g (1.5 months old), rebon shrimp flour, commercial feed Prima Feed (PF)-1000 and Carboxymethyl Cellulose. The tools used are digital scales with an accuracy of 0.01, 12 aquariums measuring 40x25x25 cm3, fiber tub, aeration, heater (Atman), mercury thermometer with an accuracy of 0.1°C, DO Meter (Lutron), pH meter (DrGray), Millimeter block, and ruler with an accuracy of 0.1 cm, grinder, pelletizer, jar, spoon, oven, fine sieve, plastic zipper, styrofoam, and waring machine.

Test Feed Manufacturing Stage

During maintenance, the feed given is test feed in the form of commercial feed with added rebon shrimp flour. Rebon shrimp and commercial pelleted feed are previously finely floured, then mixed according to the treatment ratio, then mixed according to the formulation, warm water and Carboxymethyl Cellulose are added, then mixed until evenly distributed, after that the feed mixture is molded by a pelleting machine, then dried using an oven, it takes time. 1 hour using a temperature of 50-60°C, or using the sun for 1-2 days (Khairuman, 2002). Then, store it and close it tightly in a ziplock bag, add silica gel to keep the food from getting damp or moldy.

Preparation for Implementation

Each aquarium was filled with ±15 L of water, followed by the installation of aeration and heater installations, then 5 fish were put into the aquarium and first acclimatized for 7 days in a fiber tub to adjust to the new environment and rearing media. During the acclimatization process, the fish are fed (without the addition of rebon shrimp flour) with a feeding frequency of 3 times a day, then the fish's body weight is weighed to determine the initial weight and dose of feed that will be given at the rearing stage.

Research Implementation

The research was carried out for 56 days and observations were carried out every 7 days, including measuring water quality, weighing and measuring body length along with weighing the feed that would be given to adjust the amount of feed in the next rearing period. Feed is given 3 times a day, namely at 07.00 WIB, 12.00 WIB and 16.00 WIB. The amount of feed given is 5% of the fish biomass. Based on Desilva et al., (1995) that the optimal feeding frequency for catfish is 3 times a day according to their natural eating habits.

Research Methods

The research used a Completely Randomized Design (CRD) with 4 treatments and 3 replications. The treatment given is as follows:

A : 50% commercial feed : 50% rebon shrimp flour (mutiara catfish G6 transgenic)
B : 65% commercial feed and 35% rebon shrimp flour (mutiara catfish G6transgenic)
B* : 65% commercial feed and 35% rebon shrimp flour (catfish sangkuriang as control)
C : 80% commercial feed and 20% rebon shrimp flour (mutiara catfish G6 transgenic)

Research Parameters

Absolute weight growth (W)

Based on Hidayat *et al*. (2013) absolute weight gain can be calculated using the formula:

$$W = Wt - Wo$$

Information:
W = Absolute weight gain (g)
Wt = Final rearing fish weight (g)
Wo = Initial fish weight of rearing (g)
T = Duration of research (days)

Feed Conversion Ratio (FCR)

This parameter aims to compare the amount of feed consumed with fish biomass, showing the increase in fish biomass weight after being fed during maintenance and feed efficiency. Calculated using the Effendie (1997) formula below:

$$\mathrm{FCR} = \frac{\mathrm{F}}{(\mathrm{Wt} + \mathrm{D}) - \mathrm{WO}}$$

Information:
FCR = Feed conversion ratio
Wo = Biomass weight of test fish at the start of the study (g)
Wt = Biomass weight of test fish at the end of the study (g)
D = Weight of dead fish (g)
F = Weight of Feed given (g)

Protein Efficiency Ratio

The protein efficiency ratio can be determined by comparing the weight gain with the amount of feed protein consumed during maintenance (Erfanullah & Jafri, 1999). The protein efficiency ratio formula is as follows:

$$\mathrm{PER} = \frac{\mathrm{Wg}}{\mathrm{P}}$$

Information:
PER = Protein efficiency ratio
Wg = Total absolute weight (g)
P = Protein intake (g)

Protein Retention

Protein retention can be determined by carrying out proximate analysis of fish body protein at the beginning and end of rearing. The protein retention formula is as follows (Takeuchi, 1988):

$$\mathrm{PR} = \frac{(\mathrm{Fp} - \mathrm{Lp})}{\mathrm{P}} \times 100\%$$

Information:
PR = Protein retention
Fp = Amount of fish body protein at the beginning of rearing (g)
Lp = Amount of body protein at the end of maintenance (g)
P = Amount of protein consumed during maintenance

Water Quality

Observations of water quality parameters are carried out every 10 days, used as supporting data in determining the optimum conditions for raising test fish, including water temperature, dissolved oxygen (DO), and pH.

Data Analysis

Data on growth parameters, feed conversion ratio, protein efficiency ratio and protein retention were analyzed using ANOVA statistical analysis at a confidence level of 95%. To see the effect between treatments, Duncan's advanced test was carried out ($p<0.05$) using SigmaPlot 15.0 software. Data on survival rates and water quality were analyzed descriptively.

RESULT

Absolute Weight

The use of rebon shrimp meal in commercial feed affects the absolute weight gain performance of G6 transgenic mutiara catfish. The results of statistical analysis for each growth parameter showed a value (F count > F table) which stated that the addition of rebon shrimp flour to the feed had a real influence on the growth of G6 transgenic mutiara catfish (Figure 1).

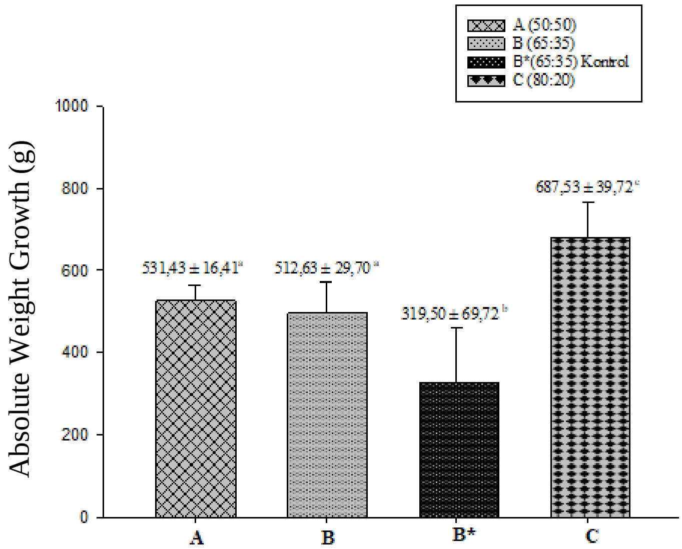

Figure 1. Absolute Weight Growth of G6 Transgenic Mutiara Catfish and Sangkuriang Catfish During 56 Days of Research. The Mean Value Followed by SD With Different Letter Notation Shows Significance ($p<0.05$)

The highest absolute weight growth was obtained in treatment C (80:20) with an absolute weight growth of 687.53 g, followed by treatment A of 531.43 g, treatment B of 512.63 g and treatment B* as a control with absolute weight the lowest was 319.50 g (Figure 1).

The addition of rebon shrimp flour to commercial feed (Figure 2) shows differences in feed conversion values. The lowest FCR value was shown by treatment C (ratio 80:20) with a feed conversion of 0.64, and the highest FCR value (1.67) was in treatment B* (control) which indicated that the fish were increasingly inefficient in utilizing feed (wasting feed). Specifically, the FCR value among transgenic fish (treatments A, B and C) ranges between (0.64-0.78), indicating that fish can use mixed feed efficiently for fish growth and transgenic fish are more feed efficient than non-transgenic fish.

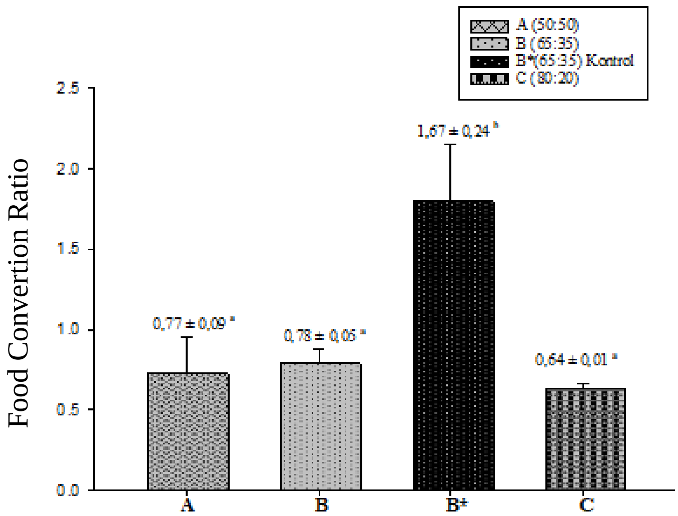

Figure 2. Feed Conversion Ratio Value of G6 Transgenic Mutiara Catfish and Sangkuriang Catfish During The 56 Days ff The Study. The Average Value Followed by SD With Different Letter Notations Indicates Significance ($p<0.05$)

Protein Efficiency and Protein Retention Ratio

The use of rebon shrimp flour in feed is able to increase the protein efficiency ratio and protein retention value significantly compared to non-transgenic fish with a protein efficiency ratio value reaching 18.75 ± 0.35% and a protein retention value of 46.43 ± 1.16% (Figure 3 and 4).

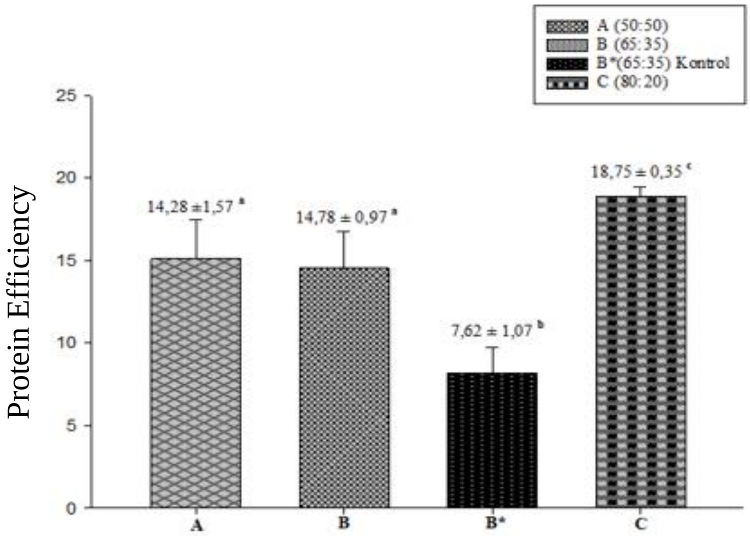

Figure 3. Protein Efficiency Ratio Values of G6 Transgenic Mutiara Catfish and Sangkuriang Catfish During 56 Days of Research. The Mean Value Followed by SD With Different Letter Notation Shows Significance ($p<0.05$)

Overall, Figures 3 and 4 show that in transgenic catfish (treatments A, B and C) the treatment with a mixture of commercial feed and rebon shrimp flour was able to increase both the protein efficiency ratio (range 14.28-18.75) and the protein retention value. (27.40-46.43%) than non-transgenic fish (treatment B*). The increase in the ratio of protein efficiency and protein retention in transgenic catfish is an effect of GH-transgenesis which stimulates increased protein synthesis. In contrast, in non-transgenic fish, both protein efficiency and protein retention values were lower (7.62 and 15.13%).

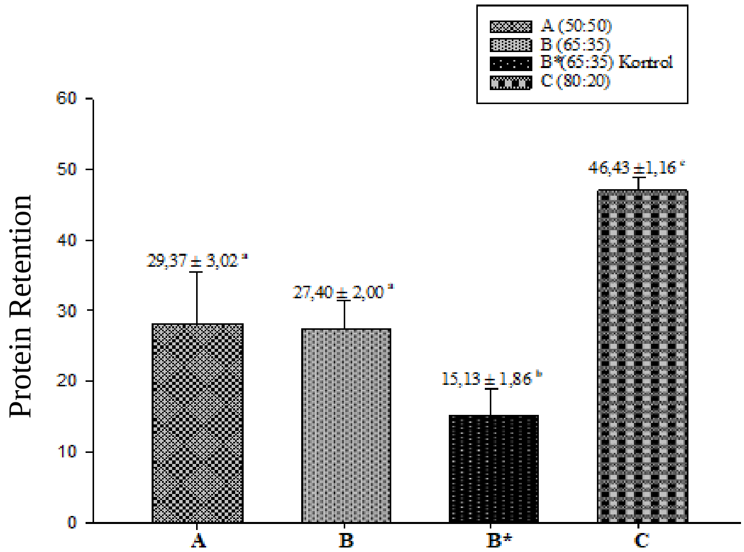

Figure 4. Protein Retention Value of G6 Transgenic Mutiara Catfish and Sangkuriang Catfish During 56 Days of Research. The Mean Value Followed by SD With Different Letter Notation Shows Significance ($p<0.05$)

Water Quality

The results of measuring water quality parameters during the research are presented in Table 1 below.

Table 1. Water Quality Range During The Study

Parameter	Treatment				Optimum Range (SNI 01-4087-2006)
	A	B	B*	C	
Temperature (°C)	27,9 – 30	27 - 30	26,5 - 29	26 – 28	25 – 30
pH	4 –7,5	6,6 – 8	4,8 – 7,6	5,2 – 8	6,5 – 8,5
DO (mg/L)	4,3 – 6,9	4 -7,8	4,6 – 7,4	5,3 - 7	>4

DISCUSSION

The increase in absolute weight growth in treatment C with the addition of 20% rebon shrimp flour was caused by the high protein content to support the fish's needs. Apart from that, it had a nutrient balance and formulation in accordance with the opinion of Isnawati *et al*. (2015) that feed has balanced protein and amino acids will influence fish growth. Rebon shrimp flour has antractant properties that encourage fish appetite. The use of rebon shrimp flour in fish feed formulas supports fish growth because it is rich in essential amino acids (methionine, arginine, threonine, tryptophan, histidine, isoleucine, leucine, lysine, valine and phenylalanine) which can speed up the metabolic process of converting feed into protein for growth. According to Guilaume *et al*. (2001) the amino acid arginine plays a role in growth because it can stimulate insulin secretion to increase growth hormone. This is in accordance with research by Indira *et al*. (2023) that the addition of rebon shrimp flour to commercial feed can increase the growth of goldfish (*Cyprinus carpio*) fry such as increasing weight and length, as well as daily growth rate. Based on research conducted by Rapida *et al*. (2022), giving dried rebon shrimp (*Acetes* sp.) increases the growth and survival of fish (*Colossoma macropomum*) which shows the benefits of adding rebon shrimp flour because it contains essential amino acids. Amino acids are generally needed to support growth and balance the body's metabolic processes (maintenance) in fish which can trigger fish growth. The maximum use limit for rebon shrimp flour as a commercial feed mixture is only 20%, considering that the addition of more than 20% increases the fluoride content which disrupts the digestion and absorption of feed that is metabolized by the fish's body (Abrar *et al*., 2019).

The increase in absolute growth cannot be separated from the activity of growth hormone (GH) in transgenic mutiara catfish which is higher than in sangkuriang catfish. Apart from having endogenous GH, transgenic mutiara catfish has the addition of exogenous GH in the form of CgGH (as a gene insert) which causes more GH production to be synthesized and makes the fish grow more than double (over-growth) compared to non-transgenic fish. This molecular study shows that growth hormone can be used as a growth indicator in transgenic fish. The activity of the CgGH hormone is closely related to the high activity of the hormone (IGF-1) in transgenic mutiara catfish. As a consequence, the absolute growth of transgenic fish is higher than that of non-transgenic fish, both in terms of body weight and absolute length. The results of research conducted by Buwono *et al*. (2015) stated that transgenic mutiara catfish had a growth speed of 2.76 to 3.53 times the size of non-transgenic catfish in G1 and 3 to 4 times in G2 (Buwono *et al*., 2019).

The results of the analysis of the feed conversion ratio values for transgenic mutiara catfish and sangkuriang catfish in (Figure 2) show that treatment C with a ratio of (80:20) had a low feed conversion ratio (FCR), namely 0.64, due to The nutrients in the feed can be digested well so that the feed is more efficient to apply and use in cultivation activities. This is in line with the statement by Robinson & Li (2015) which states that a low feed conversion value indicates that the feed is becoming more efficient and can reduce production costs. Feed conversion is influenced by the protein content of the feed, fish genetics and the ability of the fish to conserve protein (protein sparing effect) which can show the best feed conversion value. The smaller the feed conversion ratio, the food consumed is good for showing fish growth during maintenance and conversely, the greater the feed conversion ratio indicates that the feed given is not effective in supporting fish growth. The low feed conversion value is also caused by the effect of exogenous GH in transgenic fish which can reduce the feed conversion ratio. In line with the statement (Cook *et al*., 2000) which states that exogenous GH can increase the absorption of feed in the intestine such as protein, thereby increasing feed efficiency. This shows that the feed efficiency of G6 transgenic mutiara catfish is higher than that of non-transgenic catfish. Supported by research by Habibullah *et al*. (2015) and Buwono *et al*. (2016) which states that transgenic catfish have a better level of feed efficiency so that the FCR produced will be lower. The effect of GH gene transfer in fish has also been tested on several other farmed fish species and has been proven to increase growth.

The value of the protein efficiency ratio with a mixture of commercial feed flour and rebon shrimp flour with a ratio of (80:20) to transgenic G6 mutiara catfish which is the highest treatment so that it is able to utilize protein in feed well and efficiently (Figure 3), increasing the breakdown of protein into peptides to amino acids. The more protein that is broken down causes an increase in the amount of amino acids that can be absorbed and used by the body for growth. According to Hepher (1988), the protein efficiency ratio value is also influenced by the fish's ability to digest feed and the quality of the feed which can influence the protein efficiency value. The protein efficiency ratio and fish growth are positively correlated with feed digestibility, where the lower the feed digestibility, the lower the protein efficiency ratio. Treatment B* gave the lowest result, namely 7.61 when compared to other treatments, it is suspected that treatment B* was not able to utilize feed properly, low digestibility of feed in non-GMO fish and also more protein was used as an energy source without maximum protein storage. (Kim *et al*., 2018)

The high protein efficiency ratio in G6 transgenic mutiara catfish is also thought to be because it has a higher essential amino acid content than non-transgenic catfish. The effect of the presence of GH which influences the breakdown of carbohydrates and lipids through complex enzymatic pathways into pyruvic acid and then converted into amino acids, both of which contribute to increasing the amino acid content (Melzer, 2011). The protein content in feed can influence the feed's ability to provide essential amino acids. The amino acid content in feed protein determines the balance between essential and non-essential amino acids. An imbalance of essential and non-essential can lead to amino acid deficiencies, which will limit weight gain (Zhang *et al*., 2017).

The results of protein retention (Figure 4) show that feeding with the addition of rebon shrimp flour mixed with commercial feed has a significantly different effect on the protein retention value of trangsenic mutiara catfish and sangkuriang fish. Protein retention is a description of the amount of protein provided, which can be absorbed and used to build or repair damaged body cells, as well as being used by the fish's body for daily metabolism (Buwono, 2000). The high protein retention value in treatment C with the addition ratio of 20% rebon shrimp flour is because in this treatment the percentage of dry weight at the end of the study increased, the proximate test results produced high protein and the water content at a lower percentage compared to treatment A, B and B*. and there is a balance of nutrients in mixing commercial feed flour and rebon shrimp flour in feed with a ratio of (80:20) which really supports the needs of the fish. Protein retention is influenced by protein content, protein quality, and the amount stored for fish growth. The retention value in treatment B* with a ratio of 65% commercial flour and 35% rebon shrimp flour using sangkuriang catfish has a low value which can be caused by feed that does not support normal fish growth, growth, so that protein is changed a lot to meet metabolic needs. other. The high percentage of feed protein consumption in Sangkuriang catfish is caused by feed protein being used not only for growth, but also as digestible energy, so that the protein stored in the body of Sangkuriang catfish is lower. This is also because the G6 transgenic mutiara catfish consumes a lot of non-protein energy sources (fats and carbohydrates). The results of this research are proven by research by Muhamad (2020) which states that Sangkuriang catfish only has one GH and has a protein retention of 10.8% with feed containing 37.4% protein, which is different from transgenic mutiara catfish which has a protein retention of 42.9%. and in research by Buwono *et al*. (2023) which states that sangkuring fish has a protein retention of 8.49% which has a feed content of 39% while in transgenic mutiara catfish it is 35.8%. If protein retention is below 20%, indicating slow growth like treatment B*, this is the impact of the fish not having exogenous GH gene insertions in non-transgenic fish.

The comparison of high and low protein retention between G6 transgenic mutiara catfish and sangkuriang catfish cannot be separated from the presence of GH activity in the fish. GH functions in transporting amino acids in feed to skeletal muscle, heart, adipose tissue, liver and other visceral organs both for storage and use for growth. GH exerts a protein sparing effect by mobilizing the body's energy substrates, such as glucose, free fatty acids, and ketones in the same tissues where protein synthesis is stimulated. Therefore, protein retention is high in G6 transgenic mutiara catfish because there are two GHs working, namely endogenous and exogenous GH (CgGH), compared to sangkuriang catfish which only has endogenous GH. Transgenic fish can effectively convert fat and carbohydrates into metabolic energy sources and convert fat into protein compared to non-GMO fish, which is known as protein sparing action.

Based on data from measurements of water quality parameters (Table 1), the water quality range values between treatments A, B, B* and C did not experience significant differences because the four treatments were carried out in the same place and environmental conditions (homogeneous). The water temperature in the maintenance medium ranges from 26-30°C. According to Khairumam & Amri (2002) the temperature range for catfish is 20 -30°C, but the optimal temperature is 27°C. Referring to SNI, this temperature range is still within the normal threshold value recommended for catfish. The pH value obtained during the research ranged from 4-8. Kordi (2011) study shows that a good pH for maintaining fish seeds is 6.5-9. The range of pH values during the study was still within the normal range for catfish growth. During the maintenance period dissolved oxygen ranges from DO 4.3 – 7.8 mg/L which is still tolerable for the life of catfish. The results of these measurements indicate that the water quality used during the research was within the range recommended by SNI (2006).

CONCLUSION

The conclusion of this research is that the addition of rebon shrimp flour to commercial feed has a significant effect on increasing growth performance, feed conversion ratio, protein efficiency ratio and protein retention in G6 transgenic mutiara catfish. The use of 20% rebon shrimp flour is the optimal treatment shown by the highest weight gain (687.53 g), lowest FCR (0.64), protein efficiency ratio (18.75), best protein retention (46.43%) compared to other treatment.

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