Global Scientific JOURNALS

GSJ: Volume 8, Issue 6, June 2020, Online: ISSN 2320-9186
www.globalscientificjournal.com

THE EFFECT OF MIXED FEEDING ON THE CONTENT OF PROTEIN TRANSGENIC HYBRID MUTIARA CATFISH G3 AGE 4 MONTHS

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ABSTRACT

High body protein storage can cause protein retention values to increase so that fish growth becomes fast. The growth of catfish will slow down in the prospective broodstock candidate period. The growth of transgenic hybrid mutiara catfish is enhanced by inserts *Cg*GH which can increase insulin-like growth factor-I (IGF-I) so that the response and appetite of fish become high. Efforts that can be made to maintain the appetite so that fish growth does not slow down in the prospective parent period is to provide a mixture of pindang tongkol and pellet. The purpose of this study was to determine the use of the best mix of pindang tongkol and pellet that provided the highest body protein content, protein retention and growth and feed conversion ratio in transgenic hybrid mutiara catfish third generation aged 4 months. The research was conducted at the Faculty of Fisheries and Marine Sciences Hatchery of Universitas Padjadjaran and the Ruminant Nutrition and Animal Nutrition Laboratory of Animal Husbandry Faculty of Animal Husbandry of Universitas Padjadjaran from September to October 2019. The study used an experimental method with a completely randomized design with four treatment mix ratio of pindang tongkol and pellets namely A (20:80), B (30:70), C (40:60), D (50:50) in transgenic hybrid fish and E (50:50) in non-transgenic fish repeated three times. Based on the results of the study, transgenic hybrid mutiara catfish G3 aged 4-6 months with 50% pindang tongkol and 50% pellet feed giving the highest yield with the protein content of 24,47%, protein retention 37,82% and growth rate 2,87%. However the treatment with mixed feed 20% pindang tongkol and 80% pellets gave the lowest FCR value of 0,78.

Keywords: *Growth, Pellets, Pindang Tongkol, Retention Protein, Transgenic Hybrid Mutiara Catfish*

INTRODUCTION

Catfish is one of the leading commodities in freshwater aquaculture because it is popular with many people, the price is affordable and the growth is fast. One strain of catfish that has a good quality is sangkuriang catfish, but currently has decreased quality due to inbreeding to uncontrolled. The effort to overcome this is to produce strains of catfish with superior quality in growth.

Balai Penelitian Pemuliaan Ikan (BPPI) Sukamandi in 2014 produced fast-growing catfish named mutiara catfish. Mutiara catfish is the result of a combination of four strains of catfish, namely mesir, paiton, sangkuriang and dumbo catfish (Iswanto *et al*., 2014). Mutiara catfish has improved the nature of growth by inserting growth hormone dumbo catfish (*Clarias gariepinus* Growth Hormone/*Cg*GH) through transgenesis technology conducted by Buwono *et al*. (2016) produced transgenic mutiara catfish. The crossing of transgenic mutiara catfish with sangkuriang catfish produces transgenic hybrid mutiara catfish of good quality and has been produced in the third generation.

Transgenic fish are effective in converting fats and carbohydrates into metabolic energy sources and can convert fats into proteins compared to non-transgenic fish known as protein sparing action (Huang *et al*., 2017). Excess protein stored by fish is used optimally for the growth process. High body protein storage can cause the value of protein retention to increase so that fish growth becomes fast.

The rapid growth of fish depends on the amount of protein that can be absorbed and utilized by the body as a builder (Purba, 2004).

The growth of sangkuriang catfish (non-transgenic) will slow down when they are 5-6 months old (prospective broodstock candidate period) (Jauhari *et al*., 2012). The somatic growth of the prospective broodstock candidate will slow down because feed energy is only used for routine metabolism and some other energy is used for the growth of reproductive organs (Roff, 1983). The growth of transgenic hybrid mutiara catfish is enhanced by inserts *Cg*GH that can increase insulin-like growth factor-I (IGF-I) in the liver which plays a role in the refinement of carbohydrates into energy so that feed protein can be overhauled into body protein, and increase response and appetite in fish (Henning *et al*., 2013; Higgs *et al*., 2009; Permana *et al*., 2012). One of the factors that influence fish growth is feed.

Transgenic hybrid mutiara catfish have a good response and appetite for artificial and fresh feed. Efforts that can be made to maintain the appetite of fish is to provide a mixture of strong-scented food such as pindang tongkol. Pindang tongkol has a strong enough aroma so that it can stimulate the appetite of fish and increase the attractiveness of fish to the feed given. A fish appetite is maintained will produce optimal fish growth.

Therefore, to prove the value of protein retention can affect the growth of transgenic hybrid mutiara catfish at 4-6 months of age, individual fish studies are needed for approximately 2 months (prospective broodstock candidate period) by feeding a mixture of pindang tongkol and pellet. The best feed mix will provide the highest body protein content, protein retention and growth, and the lowest feed conversion ratio.

MATERIALS AND METHODS

TOOLS AND MATERIALS

Tools and materials used in the study are aquariums as a container for the maintenance of test fish, aerators, stone and aeration hose as oxygen supply devices, a heaters to control the temperature of maintenance media, a thermometers to measure water temperature, oxygen-test kits brand Sera to measure dissolved oxygen, lakmus pH to measure the degree of acidity of water, digital scales to weight fish feed and body weights, drain to pick up and move fish, label paper to give identity to each treatment, trash bag to aquarium cover, sipon tools to clean aquarium, plastic as container fillet fish, cutter for separating meat and fish bones, container styrofoam for meat storage container fillet, cameras to document research activities, stationery to record research results, transgenic hybrid mutiara catfish G3 aged 4 months resulting from crosses between trangenic mutiara catfish G2 and sangkuriang as a test animal, pindang tongkol as fresh feed, pellets as artificial feed, and methylene blue to prevent parasites and diseases in fish.

METHODS

The research method used was an experimental method with a completely randomized design (CRD) with five treatments of mixed feed and three replications. Each treatment has different protein levels (Table 1). Feed A is a mixture of 20% pindang tongkol and 80% pellet, feed B is a mixture of 30% pindang tongkol and 70% pellet, feed C consists of a mixture of 40% pindang tongkol and 60% pellet, feed D is a mixture of 50% pindang tongkol and 50 % pellets, and feed E is a mixture of 50% pindang tongkol and 50% pellets. Feed A, B, C, D are given to transgenic hybrid mutiara catfish and E feed is only given to sangkuriang catfish (control).

Table 1. Mixed Protein Feed Each Treatment

Treatment	Comparison(%)		Mixed Protein (%)
	Pindang Tongkol (PT)	Pellets (P)	
A	20	80	38,36
B	30	70	38,04
C	40	60	37,72
D	50	50	37,40
E (Control)	50	50	37,40

OBSERVATION PARAMETERS AND DATA ANALYSIS

Observation parameters in the research conducted included protein retention, growth, feed conversion ratio and water quality. Growth data and feed conversion ratio were analyzed quantitatively by analysis of variance (*One Way Anova*) to determine differences between treatments. If inter-treatment has significant effect, then it is continued with Duncan's Multiple Range Test (Sigma plot 12) with a confidence level of 95%. Data on protein retention and water quality were analyzed in a comparative descriptive.

Protein retention is the percentage of feed protein that is converted to protein stored in a fish's body. Protein retention can be calculated using the Buwono (2000), namely:

$$\frac{\text{Amount of body protein at the end of the study (g)- Amount of body protein at the beginning of the study (g)}}{\text{Amount of protein given (g)}} \text{ x } 100\%$$

The specific growth rate of test fish is calculated using the Bell *et al*. (2010), namely:

$$\frac{\text{Ln average weight of fish at the end of the study (g)- Ln average weight of fish at the beginning of the study (g)}}{\text{Length of maintenance time (days)}} \text{ x } 100\%$$

The feed conversion ratio is a value that show how much feed is needed to produce certain fish weights. FCR is calculated using the formula Bell *et al*. (2010), namely:

$$\frac{\text{Amount of feed consumed by fish (g)}}{\text{(Average weight of end of study fish + Average weight of dead fish) - Average weight of initial fish of study (g)}}$$

Water quality measured includes dissolved oxygen, degree of acidity (pH) and temperature.

RESULTS AND DISSCUSSION

PROTEIN RETENTION OF TRANSGENIC HYBRID MUTIARA CATFISH G3

Based on the proximate test results of fish meat, the initial body protein levels of the study were obtained, namely 16,93% (transgenic hybrid mutiara catfish G3) and 14,2% (sangkuriang catfish). The highest level of final body protein was found in treatment D with a value of 24,74% and the lowest protein content was in treatment E (control) with a value of 18,23% (Table 2). Similar results also occur in transgenic carp, tilapia and coho salmon that have higher body protein (Fu *et al*., 1998; Rahman *et al*., 2001; Raven *et al*., 2006). This can be caused by decreased use of protein as a source of routine metabolic energy in fish so that the body's protein content will increase. Increased body protein content can also be caused due to the presence of *Cg*GH. The growth hormone is directly proportional to IGF-I hormone concentration (Marnis *et al*., 2016). According to Higgs *et al*. (2009), growth hormone and IGF-I can accelerate the overhaul of carbohydrates and fats into energy. High or low protein content in the body of a fish can affect the value of protein retention.

Table 2. Analysis Research Results

Treatment	Initial Body Protein (%)	Final Body Protein (%)	Feed Protein (%)	Protein Retention (%)	LPS (%)	FCR
A	16.93	21.62	38.66	23.01	2.81 ± 0.28	0.78 ± 0.07
B	16.93	19.04	38.04	18.46	2.59 ± 0.54	0.86 ± 0.22
C	16.93	18.26	37.72	13.75	2.11 ± 0.12	1.01 ± 0.06
D	16.93	24.74	37.40	37.82	2.87 ± 0.58	0.79 ± 0.17
E	14.20	18.23	37.40	10, 07	1.59 ± 0.17	1.31 ± 0.17

The addition of pindang tongkol meat can affect the retention value of fish body protein. The highest protein retention value was in treatment D of 37,82% and the lowest protein retention value was in treatment E of 10,07% (Figure 1). Even though the value of feed protein content (Table 2) given was lower, the results obtained higher than other treatments.

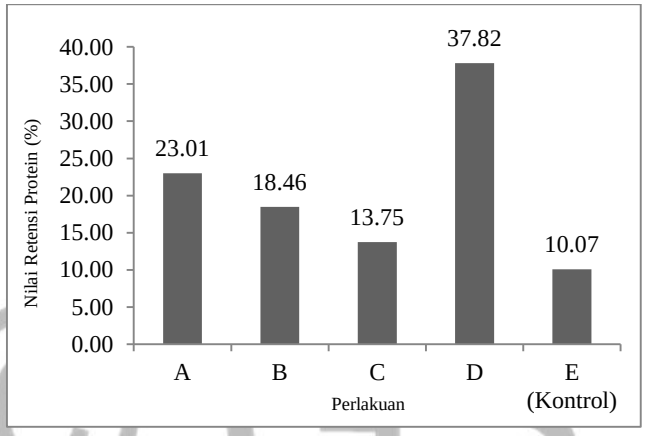

Figure 1. Protein Retention of Transgenic Hybrid Mutiara Catfish tested with feed mixed (A, B, C, D and E)

Feed mixture ratio 50:50 can affect the work of GH exogenous and increase IGF-I which plays a role in synthesizing feed protein into body protein, then stored in the body and later will be used in the growth process of the fish's body. The inserted exogenous growth hormone (GH) can increase the essential amino acids threonine, phenylalanine and lysine. Amino acids are the main constituents of proteins. When amino acids increase, protein deposits increase. High protein deposits stored in fish bodies at the end of the study can increase protein retention (Farmanfarmaian and Sun, 1999).

The protein retention value of transgenic hybrid fish is higher than non-transgenic fish. Following research conducted on transgenic goldfish (Chatakondi *et al*., 1995; Kurdianto *et al*., 2016) and Siam catfish with rGH enriched feed (Lubis *et al*., 2019). High protein retention in transgenic hybrid fish occurs due to GH exogenous insertions. Fish that have only one growth hormone (non-transgenic) will have less protein retention and absorption values compared to fish that have two growth hormones (transgenic) (Kobayashi *et al*., 2007). Besides, the retention value of protein is also determined by the source of the protein used in the feed so that it can know the quality of the protein determined by amino acids and the fish's need for these amino acids (Webster and Lim, 2002).

GROWTH OF TRANSGENIC HYBRID MUTIARA CATFISH G3

Based on observations for 8 weeks, transgenic hybrid mutiara catfish G3 fed a mixture of 50% of pindang tongkol and 50% of pellets (D) gave the highest weight gain of 213,66 g with a final weight of

262,7 g (Figure 2). By the results of research conducted by Henditama *et al*. (2015) and Trisnawati *et al*. (2014).

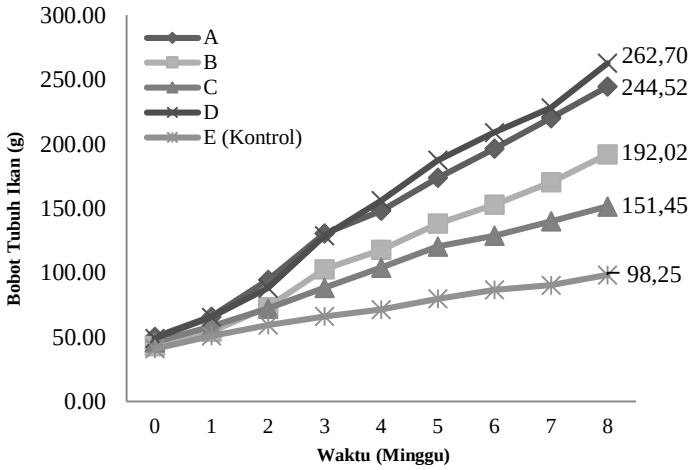

Figure 2. Growth of Transgenic Hybrid Mutiara Catfish Every Week After the Feeding of Test Feed (A, B, C, D and E)

The growth of the broodstock catfish production stage had slowed when they were 5 to 6 months old (Jauhari *et al*., 2012). However, the results of research on transgenic hybrid mutiara catfish G3 show that at the age of 5 to 6 months, fish growth has not slowed down, weight gain is increasing every week which is still quite high (Figure 2). According to SNI 6484.3 (2014), an 8 month old African catfish can reach a size of 300 g. However, in transgenic hybrid mutiara catfish that are given a mixture of meat mixed with pindang tongkol and pellets, at the age of 8 months the size is estimated to reach 500 g. Catfish which are fed with meat of pindang tongkol will increase appetite and response to feed because the pindang tongkol crumbs have a strong enough aroma to stimulate fish appetite (Garling *et al*., 1976).

The highest average growth rate was found in transgenic hybrid mutiara catfish G3 treatment D of 2,87% weight/day, while the lowest average growth rate was in sangkuriang catfish with E treatment of 1,59% weight/day (Figure 3; Table 2).

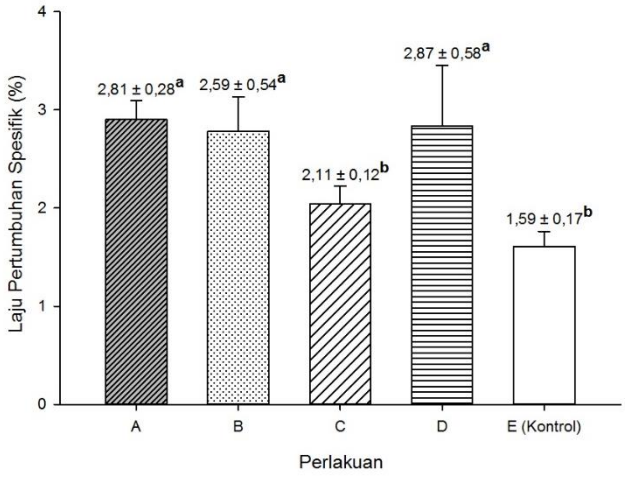

Figure 3. Specific Growth Rate of Transgenic Hybrid Mutiara Catfish given test feed (A, B, C, D and E)

The growth rate of transgenic hybrid mutiara catfish G3 is higher than that of sangkuriang (non-transgenic) catfish. Following research conducted by Buwono *et al*. (2016), Kobayashi *et al*. (2007) and Li *et al*. (2007). Growth with a ratio of 50% pellet feed mix and 50% pindang tongkol booster can increase growth and protein retention in fish. High protein retention will produce high growth (Farmanfarmaian and Sun, 1999). Fish growth can also be influenced by the feed conversion ratio so that it can be seen how much the fish feed efficiency can produce high growth (Kim *et al*., 2005; Martinez *et al*., 2000; Melzer, 2011).

FEED CONVERSION RATIO OF TRANSGENIC HYBRID MUTIARA CATFISH G3

Value feed conversion ratio (FCR) were obtained, namely the treatment of A (0,78), B (0,86) C (1,01), and D (0,79) not significantly different, but significantly different from treatment E (1,31) (Table 2). The lowest FCR value was found in treatment A, namely the feeding of a mixture of 20% pindang tongkol and 80% pellets (Figure 4).

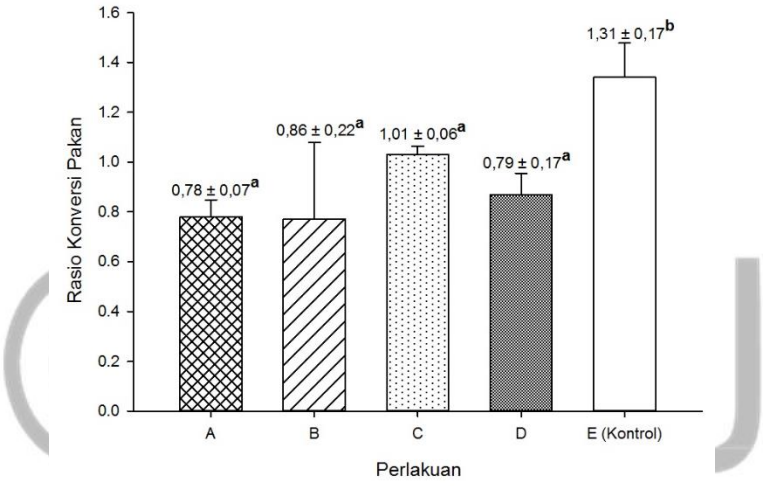

Figure 4. Feed Conversion Ratio of Transgenic Hybrid Mutiara Catfish Feed Feed Test (A, B, C, D and E)

A low FCR values occur because there is an increase in the conversion of feed into body protein. It can also be caused by the expression of growth genes (*Cg*GH) which can convert fat and feed carbohydrates into routine metabolic energy sources in transgenic fish (Buwono *et al*., 2019). The role of GH exogenous in reducing FCR of transgenic fish has also been reported by Kurdianto *et al*. (2016) and Martinez *et al*. (2000). Feeding rates for transgenic fish are lower, but growth is higher than non-transgenic fish (Martinez *et al*., 2000). The low FCR and high fish growth characterize a good level of feed efficiency. The sfficiency of transgenic fish feed is related to GH exogenous and IGF-I inserts which are closely related to the efficiency of converting feed into the meat (Kim *et al*., 2005).

WATER QUALITY

Quality measured during the study is temperature, pH and dissolved oxygen (DO). The temperature during the study ranged from 29^{o}-30^{o} C, pH 7 and DO 4 mg/L (Table 3). The results of the water quality measurements show that the water quality used during the study is suitable for the maintenance of transgenic hybrid mutiara catfish as it is still following the standards recommended by SNI 6484.3 (2014) regarding the production of african catfish.

Table3. Water Quality Analysis Research

Parameter	Results	Standards	
Temperature(°C)	29 - 31	25 - 30	SNI 6484.3 (2014)
pH	7	6,5 - 8	
DO (mg / L)	4	> 3	

CONCLUSION AND SUGGESTION

CONCLUSION

Based on the results of research that have been done, the highest body protein content, protein retention and growth rate in transgenic hybrid mutiara catfish G3 aged 4-6 months (prospective broodstock candidate period) are found in the treatment with a mixture of 50% pindang tongkol and 50% pellet feed. Whereas the lowest FCR was found in the treatment with mixed feed 20% pindang tongkol and 80% pellets.

SUGGESTION

Feeding a mixture of 50% pindang tongkol and 50% pellets can be used in the production process of prospective broodstock candidate transgenic hybrid mutiara catfish G3 as it provides the highest yield. Besides, it is necessary to conduct further research on the comparison of these mixed feeds to the reproduction of prospective broodstock candidate transgenic hybrid mutiara catfish G3.

ACKNOWLEDGEMENTS

The author would like to thank the lecturers who provided their advice and guidance to the writers, parents and relatives who have provided encouragement and material prayers, friends who always encourage and motivate writers, and all those who have helped in this research.

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