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Title-Combinatory Effects of Diets with Three Protein Levels and Two Fat Levels on Growth Performance and Fillet Composition of Cage Cultured Genetically Improved Farmed Tilapia (GIFT)

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Abstract

Effect of six diets having two lipid levels and three crude protein levels on growth and fillet composition of Genetically Improved Farmed Tilapia (GIFT) reared in cages was evaluated. Diets having Crude Protein levels (CP) 26%, 30% and 36% and Crude Fat levels (CF) (7% and 12%) were formulated. Experiment was set up according to 3 X 2 factorial designs. GIFT fingerlings (2.94 \pm 1.47g) were stocked at a density of 75 fish /m⁻³ in net cages each having a water volume of 2 m³. Fish in quadruplicated treatments were fed with six diets for 150 days. Fish fed with 36% CP diets had significantly different (P≤0.05) values for all growth indices; Final Mean Weight, Specific Growth Rate, Feed Conversion Ratio and Net Yield while no such difference found between 26% CP and 30% CP diets for the same growth indices. Increasing fat level from 7% to 12% had no significantly affected (P≤0.05) the, CF content in fish fillets while no protein sparing seen in fish fillets. There was no marked difference in protein content in fish fillets of fish and the diet having 36% CP and 7% CF is suitable for the grow out phase and 36/12 for fattening phase of GIFT reared in cages.

Keywords: Protein sparing; Fish feeds; Fish growth; Fillet composition; GIFT; Combinatory diets

Introduction

Fish nutrition and feeding is one of the major requirements for sustainable aquaculture production. Feed is the most expensive component in fish culture and a major part of the total variable cost representing about 30% to 60% is allocated [1,2]. Protein is the principal organic constitute of animal tissues [2,3]. The economical and nutritional aspect of protein utilization in fish is to improve the synthesis of tissues rather than protein use as energy source [4,5]. To balance these two important aspects (nutritional value and cost effectiveness) aquaculture investigators are interested in to reduce protein content in diet without affecting the growth [6].

Protein requirement of tilapia varies with the size/age, protein quality, non-protein energy level, water temperature and salinity, presence of natural food and allowance of feed [7]. If there is insufficient amount of energy in the diet the excess amount of protein will convert in to energy by the fish [8-10].

Dietary lipid is good source of highly digestible energy and only source of essential fatty acids. Lipid also plays a sparing action for protein in fish if the adequate requirement of energy level through diet is provided by lipids [11,12].

Many studies have been conducted on sparing ability of lipid and carbohydrate (energy source) to protein for various fish species in variety of culture conditions [13,14]. Inclusion of higher levels of protein in fish diet also needs high energy levels to maintain desired protein to digestible energy ratio [5].

Increase in dietary lipids in fish feed than the requirement however could increase fat deposition in fish tissues and also negative effect on utilization of other nutrients resulting growth retardation [15,16].

Different dietary protein and oil levels on growth of tilapia had no indication of sparing effect on dietary protein [17]. Feed intake of juvenile Nile tilapia (*Oreochromis niloticus* increased with the increase of dietary proteins, while dietary energy seemed to have no influence on the growth [6]. Kim, et al. [5] demonstrated that the effect of different dietary protein (20% and 40%) and lipid levels (9% and 17%) growth and body composition of juvenile Far Eastern catfish, *Silirusa sotus*. The protein requirement for the maximum growth of juvenile far eastern catfish is more than 40% and increasing the dietary lipid level from 9% to 17% had improved growth and feed utilization.

As such there is a need of proper understanding on nutritional requirements of a particular fish species and interaction of those nutrients on growth of fish when formulating diets for different fish species.

Fish species are cultured in cages in a wide variety of culture environments. Fish in cages is constrained by limited availability of fish feed of required quality [3]. Genetically Improved Farmed Tilapia (GIFT) a developed strain of *O. niloticus* and considered as a suitable culture fish species in cages [18]. With the aim of popularizing the GIFT cage culture in large number of reservoirs and tanks scattered throughout Sri Lanka, present study was conducted to evaluate the effect of two dietary lipid levels and three crude protein levels on growth and body composition of GIFT raised in cages.

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	Treatments (% Protein/% Lipid)							
Ingredients (%)	Formulated diets CP/CF							
	Fish meal	20	20	25	25	35	35	
Soya bean meal	20	20	27	27	35	35		
Coconut seed cake	20	20	20	20	20	20		
Maize	26	3	18	3	7	#		
Rice bran	13	35	9	23	2	8		
Vitamin & Mineral mix	1	1	1	1	1	1		
Vitamin E	#	1	#	1	#	1		
		Nutrient conter	t (dry matter basis)			- ·		
CP (%)	26.14	26.18	30.47	30.25	36.65	36.8		
CF (%)	7.32	12.47	7.7	12.55	7.65	12.25		
Ash (%)	7.06	8.47	7.45	8.55	10.01	10.11		
Cost/Kg (US\$) [*]	0.55	0.5	0.63	0.6	0.77	0.75		

#not included, *Sri Lankan rupees converted in to US\$ (12.09.2015)

Table 1: Ingredient percentage, cost and chemical composition of the experimental diets (mean, n=2).

FMW (g)	SGR (% day ⁻¹)	FCR	PER	NY (Kg/2m ³)	SR (%)
111.12 ± 6.0ª	2.41 ± 0.03ª	1.54 ± 0.04ª	2.48 ± 0.07ª	13.6 ± 3.14ª	90.0 ± 3.54ª
109.58 ± 7.23ª	2.40 ± 0.04ª	1.72 ± 0.09 ^a	2.23 ± 0.12ª	11.9 ± 6.25ª	89.16 ± 3.59ª
117.14 ± 2.93ª	2.45 ± 0.01ª	1.61 ± 0.07 ^a	2.04 ± 0.09 ^a	12.9 ± 6.88 ª	85.5 ± 4.77ª
113.74 ± 10.0 ^a	2.42 ± 0.05 ^a	1.51 ± 0.04 ^a	2.18 ± 0.06 ^a	13.8 ± 4.78ª	89.00 ± 3.77ª
158.26 ± 3.56 ^b	2.65 ± 0.01 ^b	1.22 ± 0.04 ^b	2.23 ± 0.08 ^a	18.0 ± 6.45 ^b	89.16 ± 1.77ª
166.32 ± 6.90 ^b	2.68 ± 0.02 ^b	1.14 ± 0.03 ^b	2.63 ± 0.2ª	19.0 ± 6.45 ^b	86.33 ± 4.33ª
	FMW (g) 111.12 ± 6.0ª 109.58 ± 7.23ª 117.14 ± 2.93ª 113.74 ± 10.0ª 158.26 ± 3.56 ^b 166.32 ± 6.90 ^b	FMW (g) SGR (% day ⁻¹) 111.12 ± 6.0 ^a 2.41 ± 0.03 ^a 109.58 ± 7.23 ^a 2.40 ± 0.04 ^a 117.14 ± 2.93 ^a 2.45 ± 0.01 ^a 113.74 ± 10.0 ^a 2.42 ± 0.05 ^a 158.26 ± 3.56 ^b 2.65 ± 0.01 ^b 166.32 ± 6.90 ^b 2.68 ± 0.02 ^b	FMW (g) SGR (% day ⁻¹) FCR 111.12 ± 6.0 ^a 2.41 ± 0.03 ^a 1.54 ± 0.04 ^a 109.58 ± 7.23 ^a 2.40 ± 0.04 ^a 1.72 ± 0.09 ^a 117.14 ± 2.93 ^a 2.45 ± 0.01 ^a 1.61 ± 0.07 ^a 113.74 ± 10.0 ^a 2.42 ± 0.05 ^a 1.51 ± 0.04 ^a 158.26 ± 3.56 ^b 2.65 ± 0.01 ^b 1.22 ± 0.04 ^b 166.32 ± 6.90 ^b 2.68 ± 0.02 ^b 1.14 ± 0.03 ^b	FMW (g) SGR (% day ⁻¹) FCR PER 111.12 ± 6.0 ^a 2.41 ± 0.03 ^a 1.54 ± 0.04 ^a 2.48 ± 0.07 ^a 109.58 ± 7.23 ^a 2.40 ± 0.04 ^a 1.72 ± 0.09 ^a 2.23 ± 0.12 ^a 117.14 ± 2.93 ^a 2.45 ± 0.01 ^a 1.61 ± 0.07 ^a 2.04 ± 0.09 ^a 113.74 ± 10.0 ^a 2.42 ± 0.05 ^a 1.51 ± 0.04 ^a 2.18 ± 0.06 ^a 158.26 ± 3.56 ^b 2.65 ± 0.01 ^b 1.22 ± 0.04 ^b 2.23 ± 0.08 ^a 166.32 ± 6.90 ^b 2.68 ± 0.02 ^b 1.14 ± 0.03 ^b 2.63 ± 0.2 ^a	FMW (g) SGR (% day ⁻¹) FCR PER NY (Kg/2m ³) 111.12 ± 6.0 ^a 2.41 ± 0.03 ^a 1.54 ± 0.04 ^a 2.48 ± 0.07 ^a 13.6 ± 3.14 ^a 109.58 ± 7.23 ^a 2.40 ± 0.04 ^a 1.72 ± 0.09 ^a 2.23 ± 0.12 ^a 11.9 ± 6.25 ^a 117.14 ± 2.93 ^a 2.45 ± 0.01 ^a 1.61 ± 0.07 ^a 2.04 ± 0.09 ^a 12.9 ± 6.88 ^a 113.74 ± 10.0 ^a 2.42 ± 0.05 ^a 1.51 ± 0.04 ^a 2.18 ± 0.06 ^a 13.8 ± 4.78 ^a 158.26 ± 3.56 ^b 2.65 ± 0.01 ^b 1.22 ± 0.04 ^b 2.23 ± 0.08 ^a 18.0 ± 6.45 ^b 166.32 ± 6.90 ^b 2.68 ± 0.02 ^b 1.14 ± 0.03 ^b 2.63 ± 0.2 ^a 19.0 ± 6.45 ^b

¹Values in the same column having different letters are significantly different (P≤0.05). FMW – Final Mean Weight, SGR – Specific Growth Rate, FCR – Feed Conversion Ratio, PER – Protein Efficiency Ratio, NY – Net Yield, SR – Survival Rate CP- Crude Protein, CF- Crude Fat

Table 2: Growth performance indices of GIFT fed with six experimental diets for 150 days (mean ± SE, n = 4)¹.

Material and Methods

The study was carried out in cages established in Kattakaduwa perennial reservoir (80 ha, N 06° 07' 26.2" E 80° 52' 41.2"), at Hambantota District (Southern Province) in Sri Lanka. Twenty-four cages, each having 2 m³ water capacities (size 1 m W × 2 m L × 1.5 m H), made of 2 mm high density polyethylene nets, were used. Twenty-four cages were randomly divided in to six groups to have four cages for each treatment. A total of 3600 GIFT fingerlings having average initial weights (2.94 ± 1.47 g) were stocked at a density of (75 fish/m⁻³). The diets with three dietary Crude Protein (CP) levels (26%, 30% and 36%) and two dietary Crude Fat (CF) levels (7% and 12%) were used and the experiment was designed according to 3 × 2 factorial designs.

The six diets were prepared by locally available feed ingredients. Ingredient percentage, cost and proximate composition of the six diets are given in Table 1. The experimental fish in each cage fed at 10% biomass of fish at initial month and daily feeding percentage altered accordingly monthly mean weight gain using standards described by Lim and Webster [3].

Fish were fed for 150 days. Monthly samples from each cage (30%) were taken randomly. Individual body weights were recorded to the nearest 0.01 g by using an electronic balance (Digital Scale A& D).

Final Mean Weight (FMW), Net Yield (NY), Specific Growth Rate (SGR), Feed Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) were calculated as described by Abdel-Tawwab [19] and Survival Rate (SR) also calculated at the end of the study.

NY = Gross yield (g) - Initial total fish weight (g)

SGR (% day⁻¹) = $100 \times$ (Ln Final mean weight – Ln Initial mean weight)/ Culture period (days)

FCR = Dry feed given (g)/Wet weight gain (g)

PER = Increment in body weight (g)/Protein intake (g)

 $SR = 100 \times (Number of fish harvested/Number of fish stocked)$

At the end of the study three fish from each sex from each cage were collected and a total of 24 fish per treatment were pooled as a composite sample for proximate analysis. Six fish were randomly collected from this composite sample and subjected to proximate analysis using standard procedures [20].

Normally distributed data were subjected to Two way Analysis of Variance and Student Newman-Keuls test to find how the growth indices vary in fish fed with six different diets. Effect of three protein levels and two fat levels on growth were seperately analysed by One way Analysis of Variance using SPSS 16 statistical software. Data are reported as mean \pm standard error and the significance level was assessed at (P≤0.05).

Results

The growth performance and feed utilization of GIFT fingerling under different levels of dietary protein and fat are given in the Table 2. Changing protein level from 26% to 36% resulted significant differences ($P \le 0.05$) in growth performance indices; final mean weight, specific growth rate, net yield and feed conversion ratio (Table 2). The highest values for FMW, SGR, NY and better FCR were observed in fish fed with 36% CP diets (Table 3). There was no significant difference

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Parameter				Mean values for each fat levels		
	26% CP	30% CP	36% CP	7% CF	12% CF	
FMW	110.35 ± 4.36ª	115.44 ± 4.88ª	162.29 ± 3.9 ^b	128.84 ± 6.71ª	129.88 ± 8.88ª	
SGR (% day-1)	2.40 ± 0.02 ^a	2.44 ± 0.02ª	2.66 ± 0.01 ^b	2.50 ± 0.03ª	2.50 ± 0.04ª	
FCR	1.63 ± 0.05ª	1.58 ± 0.04ª	1.18 ± 0.03 ^b	1.46 ±0.05ª	1.46 ± 0.07ª	
PER	2.36 ± 0.08ª	2.11 ± 0.06 ^a	2.43 ± 0.15ª	2.25 ± 0.07ª	2.35 ± 0.11ª	
NY (Kg)	12.8 ± 4.6ª	13.4 ±4 .21 ª	18.6± 4.62 ^b	14.9 ± 7.44 ^a	14.90 ± 9.59ª	
N – Final Mean Weight, SGR – Specific Growth Rate, FCR – Feed Conversion Ratio, PER – Protein Efficiency Ratio, NY – Net Yield, CP- Crude Protein, CF- Crude						

Table 3: Pooled value (mean ± SE, n = 8 for crude protein levels and n = 12 for crude fat levels)¹ for FMW, SGR, FCR, PER and NY of GIFT during 150 days cage culture period.



Experimental diets CP/ CF	Fillet composition						
	СР	CF	Ash	DM			
26/7	16.66 ± 0.13ª	2.36 ± 0.01 ^a	1.07 ± 0.04 ^{ab}	20.14 ± 0.2^{a}			
26/12	16.16 ± 0.73ª	3.18 ± 0.47 ^b	1.11 ± 0.07 ^{ab}	20.51 ± 0.33ª			
30/7	18.62 ± 0.22 ^{bc}	2.26 ± 0.13 ^a	0.99 ± 0.06^{a}	21.89 ± 0.15 ^b			
30/12	20.15 ± 0.02^{cd}	2.42 ± 0.02^{a}	1.19 ± 0.10 ^{ab}	23.79 ± 0.14°			
36/7	17.89 ± 1.02 ^{ab}	3.44 ± 0.15 ^b	1.15 ± 0.10 ^{ab}	22.33 ± 0.87 ^b			
36/12	20.56 ± 0.16 ^d	4.09 ± 0.10°	1.34 ± 0.01 ^b	26.17 ± 0.20 ^d			
CP – Crude Protein CE – Crude Fat DM – Drv Matter							

¹Values in the same column having different letters are significantly different (P≤0.05).

Table 4: Proximate composition of fish fillet (mean ± SE, n=4)¹ according to diets.

in final mean weight and other growth indices between protein levels, 26% to 30%.

No significant difference (p>0.05) in growth indices between fish fed with 7% and 12% CF having same protein level diets indicates that increasing fat levels have no marked contribution on growth performance indices of GIFT at any protein level (Tables 2 and 3). Fish fed with the diet 36/12 shows marked growth when they reached an average weight above 130 g which is not prominently seen in lower fat diet (36/7) (Figure 1).

Fillet of fish fed with 30% and 36% had higher protein content irrespective of the fat content than fish fed with 26% CP diet. No significant difference was observed in protein content of fish fillets of fish fed with 30% and 36% CP diets (Table 4). High fat deposition in fish fillets was obtained in 12% CF in 26% and 36% protein diets.

Protein efficiency ratio was not significantly different among treatments. Survival rates were 86.33% to 90% among six treatments.

Discussion

Tilapia is known as herbivorous and omnivorous fish species feed on variety of natural food and artificial feeds [21]. Improving growth and feed efficiency with increased protein level is well known in fish [7]. Studies have shown that tilapia attain maximum growth for a range of 20% to 56% CP in diet El-Dahhar, Zeweil [6]. *Oreochromis niloticus* fry having weight of 0.8 g and 40 g need respectively 40% and 30% minimum amount of crude protein levels in diet for maximum growth [22]. The most cost effective dietary protein level lay between 25% to 28% [3]. These values are mostly taken by measuring the growth response of fish to quality graded protein (purified or semi-purified) [3], instead of commercially available cost effective feed ingredients. Present study demonstrates that increasing dietary protein level by using cheap (Table 1), easily available feed ingredients has positive effect on growth of GIFT in cage culture.

Most people prefer to consume larger tilapia while, higher

prizes claimed for large fish have made them not affordable. This has constrained them to eat smaller fish (<150 g). As such cost has overcome the consumer preference [23]. In the present study the GIFT fish attained a maximum size of 160 g within 150 days in cages is more favorable in terms of cost benefit to the consumers.

Dietary protein requirement of young Tilapia (*O. mossambicus, O. niloticus, O. aureus* and *Tilapia zilli*) often range from 34% to 36% in diet for maximum growth potential. Similarly, Bahnasawy [24] observed that mono-sex Nile tilapia reared in fertilized tanks reached significantly higher growth performance by different protein levels (17% CP to 35%) however no significant growth increase was observed between 30% and 35% CP diets. Present study also demonstrated that GIFT fed with 26 and 30% CP diet had no significant difference in growth performance revealing that dietary feed protein level within the optimal protein range makes no significant difference in growth of fish.

Abdel-Tawwab [19], Ahmad [25] reported that FCR decrease when protein level increased in diets. Similar effect was observed in the present study when CP level increased to 36% suggesting that requirement of high protein diet to maintain an efficient FCR. FCR observed between 26% and 30% CP diets was not significantly different. These results are in agreement with Bahnasawy [24], who revealed that there is no significant difference found for FCR when mono-sex tilapia when fed with 25, 30 and 35% CP.

In the present study GIFT has shown reduction in PER when CP level increase from 26% to 30%. Reduction of PER with increasing dietary CP level have been reported in many studies for different tilapia species [25-27]. This is mainly due to conversion of protein to energy when fish fed with high level of protein in diet [28]. Dietary energy supplementation through inclusion of high lipid levels proportionate to protein levels in diets may have promoted the efficient utilization of crude protein. Considering the PER and the cost of feed formulation 26% and 30% CP level diet can be considered as the most cost effective CP levels for the grow out phase of GIFT cage culture.

Diet containing 10-15% CF demonstrated good growth and feed utilization for hybrid tilapia [29]. They suggested that hybrid tilapia needs 5% CF for minimum growth and 12% CF for maximum growth in diet. Lim, Webster [3] has shown that tilapia does not tolerate as high CF in diet as carnivorous fish species and the growth can be depressed. As the growth performance and feed utilization are not significantly different in two fat levels in the present study, 7% lipid containing diet could be used for the growth of GIFT raised in cages.

High lipid diets have shown more beneficial effects for fish growth closer to the harvesting period when compared to low lipid level diets, when proper protein level is provided [30,31]. Increases of growth at the fourth month of the grow out phase in the fish fed with 36% CP and 12% CF (Figure 1) indicate that ability to utilize lipids more effectively, closer to maturity of GIFT.

Synthesis and deposition rate of protein and fat content in fish body could be related to muscle function [32,33] and the composition of diet. High fat level diets (12% CF) have not increased the fat content significantly among fish fillets except in 36/12 CF diet and minimal contribution to the protein sparing of the fish (Table 4).

Conclusion

GIFT in cages fed with elevated protein levels (36%) produce best growth performance. Fat levels (7 and 12%) has no significant difference effect on GIFT growth performance and feed utilization. Increased fat levels (12%) have not contribute to protein sparing of fish too. As such, 36/7 diet is suitable for grow out phase and 36/12 for fattening stage of the GIFT in cages.

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