

PROTEIN REQUIREMENTS OF GIFT TILAPIA FINGERLINGS REARED IN SEAWATER

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INTRODUCTION

Aquaculture is one of the fastest growing food production sectors in the world and has been growing steadily. Over the last three decades, the global aquaculture production has tripled, growing at an average annual rate of 8.8 %. Asia accounts for 88 % of aquaculture production worldwide (FAO, 2014). The lesser contribution of marine fisheries than inland fisheries and global scarcity of freshwater signify the opportunity for the culture of promising species like tilapia in seawater. Tilapia believed to have been evolved from its marine ancestor (Kirk, 1972). Tilapias are among the most important commercial freshwater fish species in the tropics. (Ahmed H. Alharbi, 2011). Tilapia is otherwise known as "Aquatic Chicken" due to its adaptability to a wide range of conditions (EI-Sayed, 2006). Tilapia culture is gaining popularity because of its white muscle with mild flavor with no intra-muscular bones (Hasan et al., 2014). Suresh and Lin (1992) reported that Food Conversion Ratio (FCR) in some tilapia species reared in saline water are better than in freshwater. Requirement of nutrients and protein particularly, may differ in freshwater and saltwater culture systems (Altinok and Gizzle, 2001). Tilapia reared in seawater and brackishwater environments may reguire low protein requirements than fish reared in freshwater environments, which are species specific (EI-Saved, 2006 b). Hence, the present study was undertaken to find out protein requirements of Genetically Improved Farmed Tilapia (GIFT) fingerlings in seawater

MATERIALS AND METHODS

The experiment was carried out for 75 days in cement tanks at

ABSTRACT GIFT tilapia fingerlings bio growth experiment was carried out in seawater. Seven diets were fed to tilapia fingerlings with different crude protein from 15 - 45 %. The mean weight gain of GIFT tilapia fingerlings reared in seawater showed highly significant positive correlation with different inclusion level of crude protein in the

fingerlings with different crude protein from 15 - 45 %. The mean weight gain of GIFT tilapia fingerlings reared in seawater showed highly significant positive correlation with different inclusion level of crude protein in the experimental diet. The mean growth value of GIFT tilapia fingerlings reared in seawater fed with different experimental diets showed significance differences in C/T2 (p<0.01), T2/T5 (p<0.05) and T6/T7 (p<0.01). GIFT tilapia fingerlings reared in seawater showed better growth (0.22 g / day) diet containing 20 % crude protein than other experimental diets. The present investigation showed that GIFT tilapia fingerlings reared in seawater require 20% crude protein than other experimental diets.

> Coastal Aquaculture Farm Unit at Fisheries College and Research Institute, Tharuvaikulam, Thoothukudi. GIFT tilapia fingerlings were procured from a freshwater rearing facility of this Institute and transported to the Tharuvaikulam (located between latitude 8°53È53.31 N and longitude 78°10È34.97 E) Tamil Nadu, India. All the tanks were cleaned, disinfected using Potassium permanganate (KMnO.) solution and provided with coarse sand in the bottom of the tank (5 cm) in order to mimic the natural bottom environment. Seven hundred twenty healthy fingerlings ranged between (05.10 \pm 0.20 to 6.00 \pm 0.40g) were selected and randomly distributed into 24 experimental tanks (30 tilapia fingerlings × 8 treatments \times 3 replicates) supplied with aeration. The principle feed ingredients were procured from local markets in Thoothukudi, dried in sunlight for two days and pulverized. The hand pellet feed were collected, dried in the shade for 2 h, labeled and stored in air-tight plastic containers. The proximate composition of the feed ingredients (Table 1) and test diets (Table 2) were analyzed using standard procedures (AOAC, 1984). The Nitrogen Free Extract (NFE) was calculated by the following formula:

> NFE = 100 - (% crude protein + % crude fat + % ash + % crude fibre + % Moisture)

> Seven diets were formulated with different inclusion level of protein 15 %, 20 %, 25 %, 30 %, 35 %, 40 %, 45 % using Pearson Double Square method as given by De Silva and Anderson (1995). (Table 3).

Diets were hand fed to the fish twice daily (10 am and 3.30 pm) for 75 days. Uneaten feed and fecal matter were siphoned out of the tanks daily using a plastic hose and feed intake was

determined. Routine water exchange (50%) (Once in 7 days) were carried out and water quality parameters were assessed using standard method (APHA, 1995). Fingerlings were sampled at 25 days interval upto 75 days to detemine growth performance and survival using the following standard formula.

Mean weight gain (g)	=	mean final weight- mean initial weight
Average daily growth (g/day)	=	mean final weight- mean initial weight/ day of culture.
Feed conversion ration	-	total dry feed intake/wet weight gain
Feed conversion efficiency(%)	=	100 (wet weight gain/ dry feed fed)
Specific growth rate (%/day)	-	100 (In final weight- initial weight) / no.of days
Survival rate(%)	-	100(initial number of fish- final number of fish).

Based on the information derived from biological data, individual statistical relationship (Student's't' test and linear correlation) of experimental tilapia fingerlings maintained on different dietary regimes were analyzed following the Biostatistical Method of Christenson (1996).

RESULTS AND DISCUSSION

By gradual increase of salinity in freshwater, (an average of 7 ppt/ day) the tilapia fingerlings became acclimatized to live in almost near seawater condition by the 5th day of experiment. El-Sayed (2006 b) also recorded that the tilapia fingerlings required 04 days to acclimatize from freshwater (0 ppt) to seawater (35 ppt). Athithan *et al.* (2011) reported the acclimation of tilapia fingerlings in almost near seawater condition by the 5th day upon the gradual increase of salt concentration in freshwater on an average of 5 ppt per day. In the present study also similar result was affirmed. Lende *et al.*

(2015) reported that cotton seed meal replacing 30% of fish meal and soybean meal replacing 40% of fish meal in the diet of *O. mossambicus* advance fry gave better growth.

The calculated different bio-growth parameter for different experimental diets for GIFT tilapia fingerlings are given in Table 4. GIFT tilapia fingerlings reared in seawater showed highest growth in T2 (20% CP). The mean growth value of GIFT tilapia fingerlings reared in seawater showed highly significant positive relationship with different inclusion level of protein in the diet (Table 5). Student's 't' test analysis confirmed that mean growth value of GIFT tilapia fingerlings reared in seawater fed with different experimental diets showed significance differences in C/T2 (p<0.01), T2/T5 (p<0.05) and T6/T7 (p<0.01) (Table 6).

Ahmad *et al.*, (2004) reported that the weight gain in *O. niloticus* by using varying (25, 35 and 35 %) level of dietary protein in 70 days experimental period to be 20.8, 24.9 and 23.9 g respectively. Ridha (2008) also reported an average weight gain of 10.1 g in 34 days experimental periods in the GIFT tilapia strain by using 50 % dietary protein.

The average daily growth rate of GIFT tilapia fingerlings reared in seawater showed range between 0.12 to 0.22 g/ day after 75 days of feeding experiments. Highest ADG rate observed in experimental fish fed with 20% CP test diets (0.22g/ day), whereas lowest ADG rate observed in experimental fish fed with 45% CP test diets (0.12g/ day). Mohamed (2013) reported the increase in weight gain of *O. mossambicus* increased with increasing level of protein from 22.40% to 32.70%. Tawwab (2012) opined that the feed intake, FCR and PER were affected by protein level and rearing density (P<0.05), but not by their interaction. Growth performance increased with increasing dietary protein level up to a certain level (optimum level) was also reported for *O. niloticus* (Siddiqui *et al.*, 1988).

The SGR value in GIFT tilapia fingerlings reared in seawater ranged between 1.41 to 1.92 %/ day. Highest SGR was

Proximate composition	Fish meal	GNOC	Rice bran	Wheat flour
Moisture (%)	09.61 ± 0.01	06.87 ± 0.02	06.34 ± 0.03	05.64 ± 0.02
Crude Protein (%)	40.33 ± 0.34	49.74 ± 0.27	06.32 ± 0.61	11.55 ± 0.43
Crude fat (%)	08.42 ± 0.01	06.55 ± 0.03	09.11 ± 0.04	01.73 ± 0.03
Ash (%)	32.05 ± 0.29	05.04 ± 0.03	19.13 ± 0.32	01.37 ± 0.12
Crude Fibre (%)	03.70 ± 0.05	01.96 ± 0.17	30.91 ± 0.24	00.65 ± 0.19
NFE (%)	5.89 ± 0.22	29.84 ± 0.41	28.19 ± 0.32	79.06 ± 0.29
GE (K cal / Kg)	3458	4701	3582	4004

Table 2: Proximate compositions of experimental diet (g/100g dry diet)

Proximate composition	Control C	Treatments T_1	Τ,	Τ.	T,	T,	T ₆	T ₇
Moisture (%)	06.87+0.03	07.19+0.01	06.88+0.01	08.09+0.02	07.73+0.01	06.89+0.01	07.53+0.01	08.19+0,02
Dry matter (%)	93.13 ± 0.21	92.81 ± 0.22	93.12 + 0.28	91.91 + 0.27	92.27 ± 0.29	93.11 + 0.23	92.47 + 0.31	91.81 + 0.29
Protein (%)	35.00 ± 0.07	14.94 ± 0.02	19.99 ± 0.05	24.99 ± 0.08	29.99 ± 0.10	35.00 ± 0.03	39.99 ± 0.01	44.99 ± 0.04
Fat (%)	07.53 ± 0.01	06.14 + 0.05	06.22 + 0.07	06.37 + 0.01	06.39 ± 0.07	06.32 ± 0.12	06.31 + 0.09	06.13 ± 0.02
Fiber (%)	07.00 ± 0.02	05.43 ± 0.04	05.74 ± 0.01	13.59 ± 0.06	13.00 ± 0.03	11.00 ± 0.05	07.76 ± 0.02	09.00 ± 0.12
Ash (%)	02.62 ± 0.04	02.54 ± 0.03	02.58 ± 0.01	02.57 ± 0.07	02.55 ± 0.03	02.57 ± 0.05	02.44 ± 0.01	02.47 ± 0.01
NFE	40.98 ± 1.17	63.76 ± 1.21	58.59 ± 1.16	44.39 ± 1.20	40.34 ± 1.17	38.22 ± 1.11	64.03 ± 1.23	70.78 ± 1.21
GE (kcal/100g)	435.54	403.69	411.25	382.45	394.04	412.70	546.40	600.37

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	15 %	20 %	25 %	30 %	35 %	40 %	45 %
Fish meal	8.40	15.32	22.25	29.18	36.10	43.03	49.95
GNOC	8.40	15.32	22.25	29.18	36.10	43.03	49.95
Rice bran	41.60	34.68	27.75	20.80	13.90	6.97	0.05
Wheat Flour	41.60	34.68	27.75	20.80	13.90	6.97	0.05
	100	100	100	100	100	100	100

Table 4: Growth performance of GIFT tilapia fingerlings reared in seawater fed with different experimental diets

Parameters	Control	Treatments						
	С	T1	T2	Т3	T4	T5	T6	T7
	35%	15%	20%	25%	30%	35%	40%	45%
Mean Initial Weight (g)	04.62 ± 0.13	05.00 ± 0.5	05.40 ± 0.22	05.50 ± 0.38	05.20 ± 0.24	05.60 ± 0.22	05.20 ± 0.35	04.80 ± 0.20
Mean Final Weight (g)	19.62 ± 0.26	16.25 ± 0.26	21.90 ± 0.28	19.00 ± 0.41	17.20 ± 0.22	16.10 ± 0.34	15.70 ± 0.26	13.80 ± 1.02
Mean Weight Gain (g)	15.00 ± 0.27	11.25 ± 0.45	16.50 ± 0.26	13.50 ± 0.29	12.00 ± 0.27	10.50 ± 0.40	10.50 ± 0.51	09.00 ± 0.36
Average Daily Growth Rate (g/day)-ADG	0.20	0.15	0.22	0.18	0.16	0.14	0.14	0.12
Specific Growth Rate (% / day)	1.92	1.57	1.86	1.65	1.60	1.40	1.48	1.41
Feed Conversion Ratio -(FCR)	1:1.15	1:1.66	1:1.22	1:1.52	1:1.62	1:2.00	1:1.85	1:2.00
Feed Conversion Efficiency - (FCE) (%)	86.53	60.27	81.55	65.43	61.53	50.00	53.84	50.00
Feed Efficiency Ratio – (FER) (%)	0.81	0.56	0.76	0.60	0.57	0.46	0.50	0.46
Protein Efficiency Ratio (PER)	2.30	3.73	3.80	2.40	1.91	1.33	1.24	1.02
Apparent Net protein Utilization-ANPU (%)	4.94	7.61	8.23	9.69	9.77	8.52	9.95	10.24

Table 5: Correlation between different experimental diets and mean body weight gain of GIFT tilapia fingerlings under seawater with days of culture

Sl.No	Days of culture(X)	Mean weight gain(Y)	df	Intercept Value (a)	Slope (b)	Correlation Coefficientr	P - value	LS
Control	37.75	15.94	2	4.654	0.1977	0.99	P<0.001	S
Treatment 1	37.75	12.62	2	1.30	0.1408	0.87	P<0.002	S
Treatment 2	37.75	15.94	2	6.32	0.21	0.98	P<0.009	S
Treatment 3	37.75	12.34	2	5.108	0.19	0.99	P<0.001	S
Treatment 4	37.75	12.44	2	6.76	0.15	0.94	P<0.002	S
Treatment 5	37.75	11.35	2	5.88	0.14	0.99	P<0.001	S
Treatment 6	37.75	12.44	2	7.59	0.13	0.84	P<0.004	S
Treatment 7	37.75	10.97	2	6.80	0.11	0.85	P<0.003	S
Treatment 7	37.75	10.97	2	6.80	0.11	0.85	P<0.003	S

LS-Level of significance / S-Significance / df- Degree of freedom

Table 6: Carcass composition of GIFT Tilapia fingerlings (g/100g) reared in seawater with different experimental diets on dry weight basis

Parameters	At initial	Control	Treatments At the end of ex	periment					
		С	T1 15%	T2 20%	T3 25%	T4 30%	T5 35%	T6 40%	T7 45%
Moisture %)	74.05 ± 0.12	74.47±0.21	74.35 ± 0.23	74.63 ± 0.19	74.65 ± 0.15	75.21 ± 0.23	75.11±0.29	73.44 ± 0.20	74.90 ± 0.27
Dry matter	25.95 ± 0.23	25.53 ± 0.22	25.65 ± 0.12	25.37 ± 0.22	25.35 ± 0.12	24.79 ± 0.29	24.89 ± 0.17	26.56 ± 0.18	25.10 ± 0.20
Protein (%)	15.49 ± 0.19	16.39 ± 0.27	16.13 ± 0.23	16.49 ± 0.17	16.99 ± 0.31	17.19 ± 0.26	17.37 ± 0.18	17.82 ± 0.23	18.22 ± 0.26
Fat (%)	04.00 ± 0.15	04.28 ± 0.19	04.79 ± 0.10	04.57 ± 0.13	03.81 ± 0.12	03.61 ± 0.17	03.39 ± 0.16	03.14 ± 0.20	03.02 ± 0.19
Fiber (%)	01.73 ± 0.17	00.87 ± 0.17	00.74 ± 18	00.32 ± 0.21	00.83 ± 0.15	00.53 ± 0.23	00.64 ± 0.29	00.93 ± 0.26	00.36 ± 0.17
Ash (%)	04.11 ± 0.18	03.32 ± 0.26	03.1 ± 0.23	003.11 ± 20	03.29 ± 0.27	03.17 ± 0.29	02.64 ± 0.19	03.84 ± 0.17	02.69 ± 0.26
NFE	00.62 ± 0.04	00.67 ± 0.01	00.89 ± 0.02	00.88 ± 0.07	00.43 ± 0.03	00.29 ± 0.06	00.85 ± 0.04	00.83 ± 0.01	00.81 ± 0.03

observed in T2 (20% CP) and lowest in T7 (45% CP). El-Dahhar et al. (2000) recorded SGR of 1.37 to 2.16 g/ day in O. *niloticus* juvenile using varying level of protein in diets. Moran et al. (2010) reported highest SGR in fish fed with 40% CP based diet in O. *niloticus* fry. Loum et al. (2013) reported highest SGR at 25% CP in O. *niloticus* and the lowest SGR was recorded in fish fed with 35% (T5) in experimental diets.

PER value in GIFT tilapia reared in seawater were ranged between 0.81 to 2.39. Lowest value observed in T5 and highest value in T1. No significant difference between T1 and T2 group. The present results of PER value were decreased

with increased protein level in experimental diets. Similar results were reported with *O. niloticus* (El- Dahhar *et al.*, 2000; Bahnasawy, 2009; Tawwab *et al.*, 2012). According to Bahnasawy (2009), PER value ranged between 1.36 to 2.43 in *O. niloticus*, where it is maximum at 17% CP diet. However, Tawwab *et al.* (2012) reported maximum PER value when *O. niloticus* fed on the 25% CP diet at lower density. Mohamed (2013) reported highest PER (4.22) value when *O. mossambicus* fed with 10% LPD. Increasing the dietary energy level at lower dietary protein levels. The best PER as compared to higher dietary protein levels.

SI. No.	Parameters	X ₁	X ₂	DF	Student's 't' value	P - Value	LS
1	Control / T1	12.1175	12.6175	3	- 0.34	P < 0.38	NS
2	Control / T2	12.1175	14.345	3	- 4.2049	P < 0.01	S
3	Control / T3	12.1175	12.3475	3	- 0.33	P < 0.38	NS
4	Control / T4	12.1175	12.445	3	- 0.30	P < 0.39	NS
5	Control / T5	12.1175	11.3475	3	0.79	P < 0.24	NS
6	Control / T6	12.1175	12.4475	3	- 0.20	P < 0.43	NS
7	Control / T7	12.1175	10.9725	3	0.67	P < 0.27	NS
8	T1 / T2	12.6175	14.345	3	- 1.2688	P < 0.15	NS
9	T1 / T3	12.6175	12.3475	3	0.16	P < 0.44	NS
10	T1 / T4	12.6175	12.445	3	0.37	P < 0.37	NS
11	T1 / T5	12.6175	11.3475	3	1.17	P < 0.16	NS
12	T1 / T6	12.6175	12.4475	3	0.78	P < 0.24	NS
13	T1 / T7	12.6175	10.9725	3	3.17	P < 0.02	NS
14	T2 / T3	14.345	12.3475	3	1.92	P < 0.07	NS
15	T2 / T4	14.345	12.445	3	1.83	P < 0.08	NS
16	T2 / T5	14.345	11.3475	3	2.39	P < 0.05	S
17	T2 / T6	14.345	12.4475	3	1.24	P < 0.15	NS
18	T2 / T7	14.345	10.9725	3	1.96	P < 0.07	NS
19	T3 / T4	12.1175	12.445	3	- 0.07	P < 0.47	NS
20	T3 / T5	12.1175	11.3475	3	1.19	P < 0.16	NS
21	T3 / T6	12.1175	12.4475	3	- 0.05	P < 0.48	NS
22	T3 / T7	12.1175	10.9725	3	0.75	P < 0.25	NS
23	T4 / T5	12.445	11.3475	3	1.36	P< 0.133	NS
24	T4 / T6	12.445	12.4475	3	- 0.004	P < 0.50	NS
25	T4 / T7	12.445	10.9725	3	2.15	P < 0.06	NS
26	T5 / T6	11.3475	12.4475	3	- 0.94	P < 0.21	NS
27	T5 / T7	11.3475	10.9725	3	0.35	P < 0.38	NS
28	T6 / T7	12.4475	10.9725	3	4.10	P < 0.01	S

Table 7: Student's 't' test analysis of the data relating to GIFT tilapia fingerlings reared in seawater fed with different experimental diets

DF – Degrees of Freedom / LS – Levels of Significance ; NS – Non-significance / S – Significance

Table 8: The salient physico-chemical characteristic of water recorded for GIFT tilapia fingerlings reared in seawater

Parameter	Range of variation
Temperature (°C)	30.36 ± 0.08 to 31.91 ± 0.02
Dissolved Oxygen (ppm)	06.48 \pm 0.03 to 06.98 \pm 0.06
Salinity (ppt)	36.00 to 37.00
pH	07.56 ± 0.04 to 07.75 ± 0.08
Alkalinity (ppm)	108.73 \pm 0.06 to 112.41 \pm 0.07
Hardness(ppm)	5789.8 \pm 0.19 to 6023 \pm 0.23
$NH_3-N(\mu g \text{ at. } / \text{ lit.})$	0.23 ± 0.02 to 0.38 ± 0.04
NO ₂ -N (µg at. / lit.)	0.02 ± 0.01 to 0.04 ± 0.03
NO ₃ -N (µg at. / lit.)	0.11 ± 0.06 to 0.22 ± 0.01
PO4 P(µg at. / lit.)	05.33 \pm 0.06 to 07.81 \pm 0.05

value was also observed at 26% dietary protein at an energy level of 22 MJ kg⁻¹ (Ali and Jauncey, 2005).

The final carcass proximate analysis showed an increased level of protein at the end of experiment from the initial value. Lipid content was decreased with increased protein level in all the experimental groups. Protein utilization was significantly affected by protein level and fish weight but not by their interaction. Total ash content was significantly affected by protein levels. Thus, different inclusion level of protein in experiential diets showed significant effect the proximate composition of muscle (moisture, protein, lipid, ash and fibre) when compared to the test group and control in GIFT tilapia fingerlings reared in seawater (Table 7).

The salient physico-chemical water quality characteristics recorded in the present study for all the GIFT tilapia fingerlings were within the range for growth and survival rate. None of the water quality parameters showed difference upon the inclusion of different level of crude protein in the experimental diet of the tilapia fingerlings (Table 8).

Thus, in the present study, inclusion of 20 % crude protein containing experimental diet showed higher optimum growth in GIFT tilapia fingerlings reared in seawater.

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