



The optimum level of dietary protein and feeding for improving the growth performance and feed efficiency of juveniles Hybrid tilapia (*Oreochromis niloticus* × *Oreochromis aurea*) reared in brackish water

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ABSTRACT

A 6-weeks factorial trial (2×2) was conducted to determine the optimum level of dietary protein and the optimum feeding rate for the best growth and feed efficiency of hybrid tilapia, *Oreochromis niloticus* × *Oreochromis aurea*. Two dietary protein levels were examined; 27% and 34% CP with 5% and 7% of biomass, and each treatment was performed in triplicate. Twelve plastic tanks ($54 \times 38 \times 28$ cm: L × W × H) were used to accomplish this work. The average initial weight was determined (7.5 ± 0.16 g), and fish samples were stocked (12 juveniles/tank). Fishes were fed three times daily and 6 days weekly. The water was daily exchanged at a rate of about 20% of water volume/tank. Results showed that dietary protein levels (from 27% to 34%) did not significantly affect either growth or feed usage parameters. However, 7% feeding rate had significantly higher growth parameters than 5%. Statistical analysis of the interaction between dietary protein and feeding levels showed that fishes fed at 7% as the feeding level with dietary protein at 27% or 34% was significantly higher in growth than those fed at 5% with the same diets. Additionally, results confirmed the significant difference in specific growth rate and survival rate of fishes fed at 7% as feeding rate with any diet protein level. Thus, this study suggests that 27% CP with 7% as feeding rate is more for the best growth of hybrid tilapia juvenile under the same conditions of this trial.

INTRODUCTION

The aquaculture sector in Egypt suffers from many problems, such as increased land costs and decreased freshwater supplies that hinder its development, which are the main reasons for the intensification of fish farming in Egypt. Moreover, increasing the production cost is considered the main challenge that negatively affects aquaculture projects' investment. **El-Sayed (1999)** reported that feeding is considered the main factor in aquaculture; it represents 50%– 60% of the total cost of aquaculture investments.

Additionally, protein represents about 50% of the feed cost in intensive culture. Dietary proteins are one of the essential nutrients and the component of animal tissue essential for growth. Furthermore, animal protein is the primary source of essential and nonessential amino acids (Larumbe-Morán *et al.*, 2010). Decreasing the dietary protein level increases pressure on body protein because the dietary nutrients should meet the demand of tissue protein synthesis, maintenance, and metabolism. Consequently, a reduction of fish growth is observed together with an increasing fish mortality rate. In contrast, the excess protein in fish diet leads to new protein build-up; the remainder of which would be directed to the deamination of amino acids to produce energy consumption. Thus, the excretion rate of ammonia increases in the reared ponds (Wilson, 2002; Yang *et al.*, 2002; Yue *et al.*, 2020). The feed quality can be measured with diet content of protein level, and feed cost can be greatly reduced if optimal dietary protein levels could be fed to fishes. The primary goal of every researcher is to determine the optimum protein level of fishes, the tilapia for instance, and increase farms' profits. Many studies were conducted to determine the optimum dietary levels of the tilapia fish, such as the Nile tilapia, *Oreochromis niloticus* (Abdel-Hakim *et al.*, 2001; Soltan *et al.*, 2002; El-Saidy & Gaber, 2005; Subandiyono & Hastuti, 2020; Yang *et al.*, 2021), *Sarotherodon galilaeus* (Sweilum, 2005). Other researchers worked on the hybrid tilapia, *Oreochromis niloticus* × *Oreochromis aurea* for the same target (Twibell & Brown, 1998; Wafa, 2002). Remarkably, the feeding rate is the amount of a specific feed per fish per day. The determination of the optimum feeding rate is essential for success in marine and fresh fish culture. Knowledge of feeding levels is important for achieving the best growth and feed efficiency and preventing water quality deterioration due to overfeeding. Thus, the optimum feeding level contributes to tremendous savings of feed cost (Davies *et al.*, 2006; Türker & Yildirim, 2011; Mohammed *et al.*, 2017). The hybrid tilapia, *Oreochromis niloticus* × *Oreochromis aurea*, is the most important commercial aquaculture species. Among the main reasons for its popularity is its ease of adaptation to culture and confinement conditions, its tolerance to salinity, and its high densities that enhance its retail value (He *et al.*, 2017; Nakphet *et al.*, 2017). Egypt's tilapia production represents about 66% and 63% of the total production (GAFRD, 2017). Thus, it is considered the primary fish in Egypt. In this context, this study was conducted to optimize the level of dietary protein and feeding to support the growth of the hybrid juvenile tilapia in intensive culture in brackish water.

MATERIALS AND METHODS

A factorial experiment (2×2) was conducted to inspect two dietary protein levels with two feeding rates on performance and feed use of hybrid juvenile tilapia (*Oreochromis niloticus* × *Oreochromis aurea*). This trial was conducted in a fish feeding laboratory, Faculty of Aquaculture and Marine Fisheries, Arish University. Experimental fishes were obtained from the fish research centre, Arish University, North Sinai, Egypt. Fishes were transported to the experimental site in a plastic tank (m^3) filled with oxygen. Fishes were acclimatized to laboratory conditions for a week.

Experimental treatments and feeds

This experiment was designed to be a factorial 2×2 trial for testing two dietary protein levels, 27% and 34%, with two feeding rates, 5% and 7%, of fish biomass. Treatments were performed in triplicate and formulated as the following: T1, fishes fed a diet containing 27% at a 5% feeding level; T2, fishes fed a diet containing 27% CP at a 7% feeding level; T3, fishes fed a diet containing 34% at a 5% feeding level; T4, fishes fed 34% dietary protein with a 7% feeding rate (Table (1)). The two commercial diets, 27% and 34% crude protein, were obtained from SKRETTING Egypt Company Table (2).

Table 1: The experimental design

Tank Number	Diet protein	Feeding rate %
T 1	27%	5%
T 2		7%
T 3	34%	5%
T 4		7%

Table 2: A proximate chemical analysis of experimental diets (% Dry matter basis)

Chemical analysis	Diet 1	Diet 2
Crude protein (CP)	27%	34%
Crude fiber (CF)	4.3%	4,78%
Crude fat	5.1%	7%
Gross energy (GE, Kcal/g)*	3,900	4,078
Pellet size	2 mm	2mm
Type pellets	floating	floating
Price of kg diet, L.E**	8.9	11.,2

Notice: - Chemical analysis was determined according to (A.O.A.C, 2010).

*, was calculated according to (NRC, 1993)

**, liver Egyptian.

Fish experimental and rearing conditions

The hybrid tilapia juveniles had an average weight of 7.5 ± 0.16 g and length of 8 ± 0.35 cm. Twelve plastic tanks were used; each tank had dimensions of 54 x 38 x 28 cm (L × W × H) with a volume of 57 l supplemented with two air pumps (220 v, 50 Hz, 5 w) to produce around 4 litres of oxygen/min. The fishes were reared in brackish water with a 3-ppt salinity. Fishes were randomly distributed at a stocking rate of 12 juveniles/tank. Fishes were fed three times daily at 8 a.m., 1 p.m., and 5 p.m. for 6 days weekly; the

feeding was done manually. The water exchange rate was 20% of water volume daily after the second meal.

Sampling collection

Fishes were weighed every 2 weeks to adjust the feeding amount according to changes in the body weight, and the remains of uneaten feed and fish wastes were removed from the tank bottom using a small suction pump. All tanks were drained and cleaned every 2 weeks during sampling to adjust feed intake. At the end of the trial, all fishes in each tank were weighed (g), measured to determine total length (cm) of every individual and then killed and dried to evaluate the chemical composition of the whole-body fish. The biochemical analysis of feed and whole-body fish were conducted following the methods described in **AOAC (2006)**. The gross energy was estimated for diets using the factors 5.64, 9.44, and 4.22 Kcal for CP, EE, and carbohydrates, respectively (**NRC, 1993**).

Water physicochemical analysis

Water temperature, the dissolved oxygen, and the pH were recorded daily and measured using a multi-parametre water quality analyzer (MULP-8C); values of total ammonia and nitrite were measured every 2 weeks by chemical methods (**APHA, 1992**).

Calculation indicates

Weight gain (WG, g)

FW-IW where; FW is final body weight and (g) IW is initial body weight(g).

Average daily gain (ADG, g)

WG/t where; t is the experimental period (day)

Relative growth rate (RGR, %)

$(WG/IW) \times 100$

Specific growth rate (SGR, %)

$(\ln FW - \ln IW) / t \times 100$

Condition factor (CF, %)

$(FW/FL^3) \times 100$ where; FL is final total length (cm)

Survival rate (SR, %)

$(\text{Number of fish at the end} / \text{Number of fish at the beginning of trial period}) \times 100$

Feed intake (FI, g/fish)

Feed intake per tank/number of fish at the end for this tank

Feed conversion ratio (FCR)

FI, g per fish/WG, g

Feed conversion efficiency (FCE, %)

$(WG/FI) \times 100$

Protein efficiency ratio (PER)

WG/protein intake (g)

Energy efficiency ratio (EER)

WG/energy intake

Statistical analysis

Data were analyzed by one- and two-way analysis of variance. The differences among groups were determined using Waller–Duncan test at $P \leq 0.05$ as the significance level using SPSS Statistical Package Program v.17 (SPSS, 2008).

RESULTS

Table (3) presents a detailed statistical description of water quality parameters; temperature degrees ranged between 25.5 and 29°C with a mean of 28.22 ± 0.32 . The pH was between 7.2 and 8.8, with a mean of 8.12 ± 0.16 . The DO value fell between 5.8 and 7.4 mg/l with a mean of 6.61 ± 0.187 mg/l. The total ammonia nitrogen also ranged between 0.03 and 0.15 mg/l with an average of 0.085 ± 0.01 mg/l.

Table 3: Means of water physiochemical parameters of all treatments

Item	Range	minimum	maximum	Means	SE	variance
Temperature	3.50	25.5	29.00	28.22	0.329	1.08
pH	1.60	7.20	8.80	8.21	0.164	0.27
DO, mg/l	1.60	5.8	7.40	6.61	0.187	0.35
Total ammonia, mg/l	0.12	0.03	0.15	0.08	0.011	0.001

Effect of different dietary protein levels on growth rate

Table (4) shows the effect of different protein levels regardless of feeding levels on juvenile hybrid tilapia's growth and survival rate. Statistical analysis indicated no significant differences between fishes fed a diet containing 27% CP and those fed a diet of 34% CP in FW, FL, WG, ADG, RGR, SGR, and SR. In contrast, CF was significantly ($P \leq 0.05$) higher with the diet containing 34% CP.

Table 4: Effect of dietary protein on growth, regardless of the feeding level

Item	Dietary protein		Sig.	SED*
	27% CP	34% CP		
FW, g	15.69	17.95	0.214	1.70
FL, cm	10.86	10.84	0.969	0.37
WG, g	8.19	10.48	0.207	1.69
ADG, g	0.196	0.250	0.211	0.04
RGR, %	109.24	139.77	0.207	22.60
SGR, %/day	1.72	2.05	0.207	0.24
CF, %	1.22b	1.39a	0.032	0.07
SR, %	79.14	87.51	0.184	5.86

(a, b) Average in the same row having different superscripts differ significantly ($P \leq 0.05$)

*SED is standard error of difference

Effect of different dietary protein levels on feed efficiency

Table (5) shows the impact of dietary protein levels regardless of feeding rates on feed efficiency parameters of the juvenile hybrid tilapia. All indicators of FI, FCR, FCE, PER, and EER did not significantly affect the level ($P \leq 0.05$) with increasing dietary protein level from 27% to 34% CP.

Table 5: Effect of dietary protein on feed utilization parameters, regardless of the feeding level

Item	Dietary protein		Sig.	SED*
	27% CP	34% CP		
FI, g/fish	19.92	22.12	0.49	3.07
FCR	2.50	2.14	0.07	0.17
FCE, %	40.40	47.42	0.091	3.75
PER,	1.49	1.39	0.42	0.12
EER	0.103	0.115	0.22	0.014

*SED is standard error of differences

Effect of different feeding levels on growth rate

Table (6) shows the influence of different feeding levels regardless of dietary protein levels on growth and survival rate. The statistical analysis showed significant variations at a feeding level ($P \leq 0.05$) between 5% and 7%. Fishes fed at a high feeding rate had the highest FW (19.31 g), FL (11.23 cm), WG (11.891 g), ADG (0.283 g), RGR (157.55%), SGR (2.24%), CF (1.40%), and SR (91.61%). In contrast, fishes fed with 5% of their biomass were recorded the lowest of these indicators, which were 14.33 g; 10.47 cm; 6.86 g; 0.163 g; 91.46%; 1.53%; 1.22%; 75%.

Table 6: Effect of feeding level on growth, regardless of the dietary protein

Item	Feeding level		Sig.	SED*
	5% of biomass	7% of biomass		
FW, g	14.33 ^b	19.31 ^a	0.001	0.96
FL, cm	10.47 ^b	11.23 ^a	0.023	0.28
WG, g	6.86 ^b	11.81 ^a	0.001	0.96
ADG, g	0.163 ^b	0.283 ^a	0.001	0.024
RGR, %	91.46 ^b	157.55 ^a	0.002	12.94
SGR, %/day	1.53 ^b	2.24 ^a	0.001	0.137
CF, %	1.22 ^b	1.40 ^a	0.041	0.07
SR, %	75.04 ^b	91.61 ^a	0.003	3.73

(a, b) Average in the same row having different superscripts differ significantly ($P \leq 0.05$)

* SED is the standard error of difference

Effect of different dietary protein levels on feed efficiency

Feed utilization parameters were not significantly influenced by different feeding rates (Table, 7). However, fishes fed with a high feeding rate of 7% of their biomass recorded better FCR (2.20), FCE (45.63%), PER (1.50), and EER (0.114) than those fed with 5% of their body weight, which had FCR (2.44), FCE (42.19%), PER (1.38) and EER (0.105). Naturally, FI significantly differs between the two treatments; fishes fed with 7% as feeding rate ingested 25.8 g feed/fish, and those fed at 5% of their biomass took 16.21 g feed/fish.

Table 7: Effect of feeding level on feed utilization parameters, regardless of the dietary protein

Items	Feeding level		Sig.	SED*
	5% of biomass	7% of biomass		
FI, g/fish	16.21 ^b	25.8 ^a	0.001	0.84
FCR	2.44	2.20	0.250	0.19
FCE, %	42.19	45.63	0.434	4.22
PER	1.38	1.50	0.304	0.11
EER	0.105	0.114	0.403	0.009

(a, b) Average in the same row having different superscripts are differ significantly ($P \leq 0.05$)

* SED is the standard error of difference

The effect of the interaction between dietary protein and feeding levels on the growth rate

Results of interaction between dietary protein and feeding levels are presented in Table (8). Data showed significant differences among treatments in all growth parameters and survival rates. The fishes of T1, which were fed a diet containing 34% CP with a feeding rate of 7% of their biomass, had the highest FW (20.58 g), WG (13.08 g), ADG (0.31 g), RGR (174.39%), and SR (91.62%). They were followed by fishes of T2, which were fed on a diet containing 27% CP at a 7% feeding rate. In contrast, T3 and T1 did not significantly differ in these parameters. The highest SGR was obtained by T4 (2.40%) and T2 (2.08%), and there was no significant difference among them; the lowest SGR was achieved for T3 (1.70%) and T1 (1.37%), and there was no statistically significant difference among them ($P \leq 0.05$). CF did not significantly differ among T4 (1.45%), T3 (1.35%), and T2 (1.34%), and they were significantly higher than T1, which recorded the lowest CF (1.09%). This confirmed that the increase in dietary protein level from 27% to 34% did not significantly affect growth performance.

The effect of the interaction between dietary protein and feeding levels on feed efficiency

Table (9) shows that the interaction between dietary protein level and feeding rate did not significantly appear among groups in FCR, FCE, PER, and EER. However, the worst FCR and the lowest FCE, PER, and EER were obtained by fishes of T1, which were fed a low dietary protein at a low feeding level. Alternatively, FI was significantly higher in groups fed at a high feeding level than those fed at 5% of their biomass.

Table 8: The interaction between dietary protein and feeding level on growth indicators

Item	Treatment				Sig.	SED*
	T1 27% CP with 5% of biomass	T2 27% CP with 7% of biomass	T3 34% CP with 5% of biomass	T4 34% CP with 7% of biomass		
FW, g	13.33 ^c	18.05 ^b	15.33 ^c	20.58 ^a	0.001	1.02
FL, cm	10.5	11.22	10.44	11.24	0.203	0.44
WG, g	5.83 ^c	10.55 ^b	7.88 ^c	13.08 ^a	0.001	1.01
ADG, g	0.139 ^c	0.253 ^b	0.188 ^c	0.31 ^a	0.001	0.024
RGR, %	77.77 ^c	140.71 ^b	105.15 ^c	174.39 ^a	0.001	13.54
SGR, %/day	1.37 ^b	2.08 ^a	1.70 ^b	2.40 ^a	0.002	0.144
CF, %	1.096 ^b	1.34 ^a	1.35 ^a	1.45 ^a	0.004	0.063
SR, %	66.68 ^c	91.60 ^a	83.40 ^b	91.62 ^a	0.001	0.11

(a, b, c) Average in the same row having different superscripts differ significantly ($P \leq 0.05$)

* SED is the standard error of difference

Table 9: The interaction between dietary protein and feeding level on feed utilization parameters

Item	Treatment				Sig.	SED*
	T1 27% CP with 5% of biomass	T2 27% CP with 7% of biomass	T3 34% CP with 5% of biomass	T4 34% CP with 7% of biomass		
FL, g/fish	15.86 ^d	23.98 ^b	16.57 ^c	27.58 ^a	0.001	0.01
FCR	2.72	2.28	2.17	2.12	0.120	0.24
FCE, %	36.67	44.00	47.59	47.23	0.233	5.35
PER	1.36	1.62	1.39	1.39	0.414	0.167
EER	0.094	0.112	0.116	0.115	0.381	0.014

(a, b, c, d) Average in the same row having different superscripts differ significantly ($P \leq 0.05$)

* SED is the standard error of difference

Chemical composition of whole body fish at the end of the experimental period

As presented in (10) body contents of dry matter (DM), fat (EE) and ash were significantly changed with the different treatments. Body content of DM and ash did not differ between T1 (29.15, %, 25.33%) and T4 (30%, 25.30 %) but these treatments were significantly higher in these contents than T2 (26.95%, 23.66%) and T3 (26.2%, 22.66%). Fat content significantly differed among treatments, T2 had the highest fat content which recorded 22.60% followed by T1 (20.55%) and T3 (20.10%), while T4 was the lowest which recorded (17.90%). On the other hand, the body fish in all treatments did not significantly vary in their crude protein (CP) content, but fish fed a diet containing 34% CP were higher in their protein content than those fed a diet 27% CP.

Table 10: Effect of dietary protein level and feeding level on whole body composition of hybrid tilapia juvenile (on dry matter basis)

Item	Treatment				Sig.	SED*
	T1 27% CP with 5% of biomass	T2 27% CP with 7% of biomass	T3 34% CP with 5% of biomass	T4 34% C with 7% of biomass		
DM, %	29.15 ^a	26.95 ^b	26.20 ^b	30.00 ^a	0.04	0.49
CP, %	54.25	53.75	57.25	57.05	0.43	2.43
EE, %	20.55 ^b	22.60 ^a	20.10 ^b	17.90 ^c	0.00	0.13
Ash, %	25.33 ^a	23.66 ^b	22.66 ^b	25.30 ^a	0.08	0.42

(a, b, c,) Average in the same row having different superscripts differ significantly ($P \leq 0.05$)

* SED is the standard error of difference

DISCUSSION

Water quality parameters within the optimum limit for rearing the tilapia are reported by **FAO (2006)**; the temperature for the optimum growth of African fish is between 25 and 30. **Davis (1993)** recommended that the optimum pH is between 6.5 and 9. **Riche and Garling (2003)** showed that the optimum DO mg/l for the growth of the tilapia is more than 5 mg/l. In addition, **Stone and Thomforde (2004)** reported that the desirable range of total ammonia is between 0 and 2 mg/l.

The dietary protein is one of the most important components in aquaculture feeds. It is the main element of animal macules and body fluids. Moreover, weight gain is a result of protein accumulation in tissues, growth reflection, survival rate (SR), and production costs (**Kim & Lee, 2005**). Tables (4) and (5) show the effect of dietary protein level on the hybrid tilapia performance, regardless of the feeding rates. The statistical analysis did not show significant differences between the fishes fed a diet containing 27% and those fed a 34% dietary protein in growth or feed usage parameters. The current results agree with those of **Abdel-Hakim and Mustafa (2000)** who found that the protein requirement of the Nile tilapia fed with artificial feeds ranged between 28%–30% CP. Additionally, **Abdel-Hakim et al. (2001)** reported that increasing dietary protein from 25% to 30% improved the SGR of the Nile tilapia reared in fiberglass tanks. Similarly, **Wafa (2002)** demonstrated that the shift of dietary protein levels from 25% to 30% significantly increased the WG of the tilapia. Furthermore, the results in Tables (4) and (5) agree with **Clark et al. (1990)**; they affirmed that SGR of Florida red tilapia was not significantly affected when fishes were fed diets containing 20%, 25%, and 30% CP. **Soltan et al. (2002)** found that the protein requirements of the Nile tilapia reared in tanks are more than 25%, which coincides with the results in Tables (4) and (5). Although the feed efficiency parameters did not significantly differ, the PER increased with fishes fed a diet of 27% CP compared with those fed a diet containing 34% CP. This result concurs with that of **Twibell and Brown (1998)**, who reported that PER was not significantly affected by increasing dietary protein levels from 23%–25%. To illustrate, **Wafa (2002)** stated that

a higher PER was recorded with the hybrid tilapia fed a diet of 25% CP than those fed a diet of 30% CP. The opposite trend showed that the growth rate of the hybrid tilapia significantly increased as the dietary protein level increased from 25% to 30% and 35% CP (Viola & Zohar, 1984). An increase in dietary protein level enhances and improves fish performance, but there is a limit for protein level in the diet. After that, the increase in growth is negligible. This may be due to the increase in protein level in the tilapia diet, which led to decreased dietary energy availability. As a result, more energy was required to deaminate and excrete excess absorbed AA, and the body protein gain is not commensurate with a higher eaten protein (Jayant *et al.*, 2018). Additionally, dietary with 27% CP had a higher level of carbohydrates than dietary with 34% CP; hence an increase in non-protein digestible energy in the diet increases nitrogen retention by decreasing nitrogen loss (Engin & Carter, 2001). Furthermore, NRC (2011) reported that an increase in dietary protein level above the fish requirement results in reduced deposition of amino acids in muscles and boosts deamination and catabolism rate of amino acids. Moreover, the ammonia excretion increases with an increase in the protein level, whereas excess amino acid decomposes to produce energy resulting in ammonia production (Wilson, 2002). In the same context, Ahmad *et al.* (2019) suggested that an enhancement in dietary protein level results in a decline in FW, WG, SGR, and lower FCE. Additionally, Balito-Liboon *et al.* (2018) assessed that any amount above the optimum dietary protein level tends to reduce growth. Therefore, a pronounced effect may play a role in decreasing the growth with an increased protein level in the diet, which is the low stability of diet due to reduced starchy plant compared to the low dietary level. The optimum protein requirements depend on many factors, such as fish behavior, the viability of natural food, fish age and size, rearing conditions and water salinity, the proportion of protein to energy in the diet, stocking rate, feeding rate, and diet protein source. For example, the protein requirement increased at a young age than the older age in the same species; this was shown in the study of EL-Sayed *et al.* (2003). Larumbe-Morán *et al.* (2010) reported that the tilapia reared under a high density required more protein in their diet than those reared in freshwater. Abdel-Aziz *et al.* (2019) found that replacing fish meal with soybean meal from diets of the red tilapia fry resulted in decreased SGR for the same protein level. Additionally, the diet content of amino acids and their balance is an important factor that affects the determination of the optimum dietary protein level (Abdelghany, 2006). From Tables (4) and (5), for juvenile hybrid tilapia with initial weight 7.5 g and reared at a stocking rate of one juvenile/5 l in good water quality, their parameters of growth and feed usage did not significantly vary when fed two dietary protein levels of 27% and 34% CP.

Feeding control and the optimum feeding rate determination are essential for success in marine and freshwater fish culture. Knowledge about feeding levels is important to achieve the best growth, promote feed efficiency and prevent water quality deterioration resulting from overfeeding. In addition, the optimum feeding level

contributes to tremendous savings of feed cost (Davies *et al.*, 2006). Considering the results presented in Table (6), growth and feed efficiency was significant with feeding level regardless of dietary protein levels. The feeding rate showed significantly higher SGR, WG, and the best FCR and PER. Juvenile fed at a feeding rate of 7% of their biomass was better in all indications of growth and feed efficiency than those fed at 5% as feeding rate. These results agree with that of Ng *et al.* (2000) and Adebayo and Quadri (2005); they observed that, with increased feeding rate, the growth rate enhanced at a higher rate, instead of decreasing at a lower rate. Similarly, Riche *et al.* (2004) and Yuan *et al.* (2010) showed increased growth performance by protein and lipid contents with increased feeding rate. However, these results disagree with those of Deyab and Hussein (2015) who found that 5% is the optimum feeding rate for the growth of the Nile tilapia with an initial weight of 9 g and fed a diet containing 33.8% crude protein. Similarly, feed consumption and growth curve increase with increasing feeding level up to a given limit. Wang *et al.* (1998) and Singh *et al.* (2003) reported that the enhancement in feeding level results in increased fish growth rate up to a maximum rate; however, the growth curve decreased. The age of fish species is an important factor that affects the optimum feeding rate; notably, small fishes need a higher feeding rate than larger fishes in the same species (El-Sayed, 2002; Cho *et al.*, 2003).

It is worthy mentioning that, the feed quality is an impressive factor in the optimal feeding level, whereas the varying ingredients in a standard diet greatly affect the feeding rate (Mohammed *et al.*, 2017). Moreover, the rearing conditions significantly affect the optimum feeding rate. In this essence, the optimum feeding rate of juvenile Korean rockfish was changed with different water temperatures (Mizanur *et al.*, 2014). Moreover, the photoperiod or outdoor culture allows blooming phytoplankton in the rearing ponds; consequently, the optimum feeding rate is lower than in indoor culture. The results in Table (7) show that the SR of fishes fed at 7% of their biomass was significantly higher than those fed at 5%; this result disagrees with that of Deyab and Hussein (2015) and Mohammed *et al.* (2017) who noted that the SR of *Siganus rivulatus* fry and *Oreochromis niloticus* juveniles insignificantly differed with different feeding rates. Moreover, this table illustrated an improvement in FEC, FCR, PER, and EER when the feeding level was increased to 7%. However, the statistical analysis did not give any significance in these indications between feeding rates of 5% and 7%. This result agrees with that of Marimuthu *et al.* (2011); they studied the effect of feeding levels of 2, 5, 8, and 12 of African catfish biomass, and reported that the FCR did not differ significantly between treatments.

The interaction between the level of dietary protein and feeding rate confirmed that growth performance and SR were better with fishes fed diets containing 34% (T4) and 27% CP (T2) at a feeding level of 7% than those fed with the same diets at 5% body weight (T3 and T4). The WG of T4 slightly increased compared to the that of T2. This is attributed to fishes of T4 fed at the optimum feeding level with high protein level,

whereas the activity of protease and lipase enzymes increased with increasing dietary protein level causing more feed usage and improvement of growth (**Mohanta *et al.*, 2008; Kumar *et al.*, 2011**). Additionally, fishes suffer from indigestion of feed high in carbohydrates, thus diets containing a high protein level had a high digestion coefficient because their carbohydrate is low. Furthermore, an increase in dietary carbohydrate levels resulted in decreasing feed intake. The statistical analysis also showed that SGR and SR did not differ significantly between T4 and T2 in the same manner as FW, WG, SGR, and SR did not differ significantly between T3 and T1. Thus, an increase in feeding level from 5% to 7% affected the growth and the SR of the hybrid tilapia juveniles with an initial weight of 7.5 g, compared to the increased dietary protein level from 27 to 34% CP. This result disagrees with that previously reported by **El-Saidy and Gaber (2005)**; the growth performance had an insignificant increase with increasing dietary protein levels but showed a significant increase with increasing feeding levels. The improvement in the hybrid tilapia juvenile resulted from increasing feed rate up to the optimum rate attributed to the boosting of feeding level, encouraging the increase in the availability of resources of amino acids, fatty acids, vitamins, and minerals. Hence, there are surplus resources of an absorbed compound stored as glycogen or lipid in the liver or muscles. Furthermore, an increase in feeding levels allows for increased opportunity to eat, causing less aggression and less lost energy because of crowding and competition during feeding operation (**Ashley-dejo *et al.*, 2014**).

Table (9) shows that FI (g/fish) significantly increases with increasing feeding level and dietary protein; this may be due to increased feeding level from 5% to 7% according to the aforementioned results, showing a strong effect of feeding rate, supported by **Cisse (1996)**. A change in dietary protein levels from 20% to 30% CP did not significantly affect the feed consumed by the tilapia. Conversely, **El-Dahhar (2000)** and **Soltan *et al.* (2002)** reported that increased FI would be significantly increased. Interaction between dietary protein level and feeding rate did not differ significantly on FCR, PER, and EER. This finding disagrees with that of **El-Dahhar *et al.* (1994)** and **Soltan *et al.* (2002)**, who suggested that FCR is significantly improved with increasing dietary protein level from 17% to 30% or 25% to 30% CP. Additionally, **Deyab and Hussein (2015)** stated that FCR is significantly affected by different feeding rates. Furthermore, T2 had the highest value of PER (Table, 9) and this agrees with that of **Wang *et al.* (2017)**, who demonstrated that fishes fed a diet containing 24% recorded a higher PER value than those fed a diet containing 36% CP.

Increasing the feeding rate above the optimum limit may result in fish growth; however, the efficiency of this growth is low, and the resulting composition from this growth is increasingly skewed toward storage as glycogen or neutral lipid deposits within hepatic, muscular, and adipose tissues (**Jobling, 1994**). Similarly, **Wang *et al.* (2007)** observed that *Nibeia miichthioides* fed with a high level above the optimum feeding level resulted in increased WG; however, this growth was less efficient, and PER was lower

with increasing carcass content of lipid. Increasing the feeding rate at a level higher than the optimum level results in a reduced FCR and increases the waste feed, decreasing and deteriorating the water quality. All water parameters were within the optimum limits for the tilapia growth, and both FCR and PER were not significantly decreased by the effect of different dietary protein or feeding levels (Table, 3). Accordingly, 7% as feeding rate is the optimum feeding level of the hybrid tilapia juveniles.

Chemical analysis of body fish appeared that fish fed a high level of CP (34% CP) had lower fat with higher protein content and vice versa was with those fed a diet containing 27% CP. These observations are in agreement with the conclusion of **El-Saidy and Gaber (2005)** and **Larumbe-Morán et al. (2010)** who reported that fish fed a low dietary protein had the highest carcass content of fat. This is a result of the increase of the carbohydrate level in low level protein diets to store energy and transform carbohydrates to lipids (**Tacon, 1989**). The opposite trend was reported in the studies of **Abdel-Hakim et al. (2001)** and **Soltan et al. (2002)** who found that, fish body content of CP, EE decreased when the dietary protein level increased. Another theory of **Al-Hafedh (1999)** depicted an insignificant influence of dietary protein level (25 to 45%) on body content of the CP of the Nile tilapia, and added that the lipid content decreased with the increase in protein level, while no clear trends were monitored in ash content. Regarding the relationship between body composition and feeding rates, **Deyab and Hussein (2015)** stated that the body content of CP was not significantly affected by the different feeding rates, and, notably, this result agrees completely with the present findings.

CONCLUSION

Conclusively, increasing the dietary protein level from 27% to 34% did not significantly affect growth and feed efficiency parameters of the hybrid tilapia juveniles; however, increasing the feeding levels from 5% to 7% of body weight significantly improved growth, but the indicators of feeding used did not significantly vary. Interaction between dietary protein and feeding levels was significantly higher in growth for fishes fed at 7% of their biomass with dietary 27% or 34% CP than those fed at 5% of their biomass with the same diets. Results showed insignificant differences in SGR, CF, and SR between juveniles fed at 7% as feeding level with a diet containing 27% CP and those fed at the same rate with a 34% CP. However, the interaction between dietary protein and feeding level did not significantly show in feed for the parameters used. Finally, a 27% dietary protein level with a 7% feeding level is more suitable for the optimal growth of hybrid tilapia juveniles.

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