

GROWTH PERFORMANCE OF NILE TILAPIA (*Oreochromis niloticus*) FINGERLINGS FED DIET SUPPLEMENTED WITH DIFFERENT OF SPIRULINA PLATENSIS LEVELS

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SUMMARY

The present study was designed to evaluate the use of spirulina (*Spirulina platensis*) as a replacement of different protein levels for Nile tilapia (*Oreochromis niloticus*) fingerlings. Spirulina was incorporated into four isonitrogenous and isocaloric diets at 0, 5, 10, 15 and 20% of the protein-based diet. The diets were fed to the fish at satiation twice a day for 56 days. The final weight gain, specific growth rate, protein efficiency ratio and survival rate of fish were significantly ($p < 0.05$) higher for group of fish fed diet supplemented with 20% Spirulina platensis from diet protein. The lowest feed conversion ratio was obtained for group of fish fed 20% Spirulina. There was a significant difference ($p < 0.05$) in protein and lipid in carcass proximate composition of the fish fed on diet replacement with 20% spirulina from diet protein compared to those on the experimental groups.

Moreover, fish fed diets containing different spirulina levels exhibited higher protein, serum creatinine, urea, albumin, globulin, triglycerides HDL-C values. On contrary, significant ($P < 0.05$) decreased was observed for Albumin /Globulin Ratio, LDL-CAST, ALT and AST values compared with the control. Serum protein (g/dl), creatinine (mg/dl), urea (mg/dl) albumin (g/dl), globulin (g/dl) and triglycerides (mg/dl) values were significantly increased ($P < 0.05$) with the increase of Spirulina levels being the highest at T4 (20% Spirulina/kg) diet. This study demonstrates that up to 20% of spirulina replacement was most effective in stimulating fish growth for Nile tilapia (*Oreochromis niloticus*) fingerlings.

Keywords: *Spirulina platensis*, replacement, Nile tilapia, growth performance, feed utilization, carcass composition

INTRODUCTION

Tilapia (*Oreochromis niloticus*) is widely cultured in the world. They tolerate a wide range of water quality conditions and could develop resistant to many diseases. Tilapia is a hardy prolific, fast growing tropical fish, and it can survive on a diversity of food (El-Sayed, 2006). One of the biggest problems facing the utilization of fish nutrition, in many aquaculture operations today, feed accounts more than half of the variable operating cost (NRC, 2011). Therefore, the potential use of unconventional foodstuffs such as algae, for substitution the high cost ingredients such as fishmeal is very important. Algae have attention as a possible alternative protein source for cultured fish, particular in tropical and subtropical developing countries where algae production rates are high and their higher protein, vitamins and essential fatty acids contents (El-Hindawy *et al.*, 2006 and Badawy *et al.*, 2008). Spirulina is a cyanobacterium that has been commercially cultivated for more than 10 years due to its high nutritional content; e.g. protein, amino acids, vitamins, minerals, essential fatty acid and β -carotene (Meng-Umpham, 2009). Spirulina can be considered a nutritional supplement that has various health benefits for humans, and a feed supplement for animals having economic benefits. As an example, it can be a suitable food supplement when fed to trout,

sea bass, fancy carp, red tilapia, shrimp and mollusk. It has been found that the alga can be used as an alternative source of protein and can also be used to improve the color, flavor and quality of meat (Lu *et al.*, 2003). Nowadays, *Spirulina* can be used to establish immune-potentiating functions in carp (Watanuki *et al.*, 2006; Tongsirir *et al.*, 2010; Mu *et al.*, 2000 and Nandeesha *et al.*, 2001) indicated that Spirulina could be used as an effective partially or completely replacement for fishmeal in formulated aqua feeds. Spirulina was reported to replace up to 40% of FM protein in tilapia (*Oreochromis mossambicus*) diet (Olvera-Novoa *et al.*, 1998) and even higher replacement of FM was possible in common carp (*Cyprinus carpio*) (Nandeesha *et al.*, 1998) and on Mekong giant catfish (*Pangasianodon gigas*) (Tongsiri *et al.*, 2010 and Khanzadeh *et al.*, 2016). The available nutritional data on this species are very limited up to date (Lim and Lee, 2009; Bueno Galaz *et al.*, 2010 and Zeinab *et al.*, 2015). The main propose of this study was to investigate effect of supplemented different levels of Spirulina from diet protein on BWG, FRC and feed intake in Nile tilapia (*Oreochromis niloticus*) fingerlings.

MATERIALS AND METHODS

An experiment was conducted at fish Research center in Suez Canal University Ismailia- Egypt to

study the effect of replacement of five levels of *Spirulina platensis* from diet protein on growth performance and body composition of Nile tilapia (*Oreochromis niloticus*) fingerlings. The experiment was lasted for 56 days.

Experimental diets:

Five isonitrogenous and iso-caloric diets were formulated from practical ingredients where the control diet was without *Spirulina platensis* (0%) and the other four diets were replaced by 5% (T2), 10% (T3), 15% (T4) and 20% (T5) *Spirulina platensis* from diet protein, respectively. The

composition and proximate analysis of the experimental diets are presented in Table (1 and 2). The fish were hand-fed to satiation, twice/day (10 am and 4pm) for 56 days. The experimental diets were formulated to contain almost 30% crude protein and gross energy 448 Kcal./100g. The experimental diets were prepared by individually weighing of each component and by thoroughly mixing the minerals, vitamins and additives with corn. This mixture was added to the components together with oil. Water was added until the mixture became suitable for making granules.

Table 1. Composition and proximate analysis of the experimental diets for Nile Tilapia (*Oreochromis niloticus*) fingerlings fed diets supplemented with different levels of *Spirulina plantesis* from diet protein

Ingredients	Treatments				
	Control (%)	Treatment T1 (5%) (%)	Treatment T2 (10%) (%)	Treatment T3 (15%) (%)	Treatment T4 (20%) (%)
Commercial fish meal ^(25%)	5.00	5.00	5.00	5.00	5.00
Soybean meal	60.00	57	55	52	48
Spirulina algae	00.00	2.73	5.45	8.17	10.89
Yellow corn	20.00	20.22	19.55	19.83	21.11
Rice bran	9.0	9.0	9.0	9.0	9.0
Sun flour oil	4.0	4.0	4.0	4.0	4.0
Mineral mixture ¹	1.0	1.0	1.0	1.0	1.0
Vitamin mixture ²	1.0	1.0	1.0	1.0	1.0
Total	100	100	100	100	100
Proximate Analysis					
Protein	30.60	30.18	30.72	30.91	30.75
Lipids	8.10	8.10	8.10	8.20	8.2
Ash	5.43	5.34	5.42	5.42	5.42
Fibers	7.53	7.52	7.52	7.52	7.51
NFE ³	48.34	48.86	48.24	47.95	48.12
Gross energy (Kcal/100g) ⁴	448.11	447.87	448.37	449.21	449.00
Cost/Kg (L.E)	3.65	3.56	3.50	3.41	3.29

1. Each Kg mineral mixture premix contained Mn, 22 g; Zn, 22 g; Fe, 12 g; Cu, 4 g; I, 0.4 g, Selenium, 0.4 g and Co, 4.8 mg.

2. Each Kg vitamin & contained Vitamin A, 4.8 million IU, D3, 0.8 million IU; E, 4 g; K, 0.8 g; B1, 0.4 g; Riboflavin, 1.6 g; B6, 0.6 g, B12, 4mg; Pantothenic acid, 4 g; Nicotinic acid, 8 g; Folic acid, 0.4 g Biotin, 20 mg,

3. Nitrogen Free Extract = 100 - (%Protein + %Fat + %Fiber + %Ash).

4. Gross Energy based on protein (5.65 Kcal/g), fat (9.45 Kcal/g) and carbohydrate (4.11Kcal/g). According to (NRC, 2011)

Chemical composition of *Spirulina* used in the present study was shown in Table (2). Crude protein content 55%, fat 7%, total carbohydrate 21%, fiber 5%, Minerals 8% and moisture were 4%. The wet mixture was passed through CBM granule machine with 2mm diameter. The produced pellets were dried at room temperature and kept frozen until experimental start.

Water temperature and dissolved oxygen was determined by Mettler Toledo, model 128.s/No1242 where the average range of temperature 25.30± 1.7 °C and dissolved oxygen was above 4.7 ± 0.5 mg/l. other water quality including pH and ammonia were measured every two days by pH meter (Orion model 720A, s/no 13062) and ammonia meter by Hanna ammonia meter where the average range of ammonia

was 0.07±0.001 mg/l and pH was ranged in 7.7 ± 0.7 throughout the experimental period (Table3). All water quality parameters within the accepted levels for Nile tilapia (Boyd, 1984).

Table 2. The chemical analysis of *Spirulina Platensis*

Component	Concentration (%w/w)
Crude protein	55
Fat (lipids)	7
Total carbohydrate	21
Fiber	5
Minerals	8
Moisture content	4

Table 3. Water quality parameters (Mean ±SD)

Item	Treatments				
	Control*	T1*	T2*	T3*	T4*
Temperature(°C)	25.30± 1.7	25.30± 1.7	25.30± 1.7	25.30± 1.7	25.30± 1.7
Oxygen(mg/L)	4.7 ± 0.50	4.7 ± 0.50	4.7 ± 0.50	4.7 ± 0.50	4.7 ± 0.50
Ammonia (mg/l)	0.07±0.01	0.07±0.01	0.07±0.01	0.07±0.01	0.07±0.01
pH	7.7 ± 0.72	7.7 ± 0.74	7.7 ± 0.74	7.7 ± 0.72	7.7 ± 0.72

*Note: Control diet was without *Spirulina platensis* (0%) and T1,T2,T3 and T4 were replaced by 5%,10%,15% and 20% *Spirulina platensis* from diet protein, respectively

Experimental unit:

Fish were stocked in 15 rectangle plastic tank (80cm x 60cm x 60cm) and were randomly divided into four equal experimental groups (20 fish each treatment, three replicate/tanks). Tanks supplied with air blowers. Photoperiod was 12h light/ 12h dark. Water temperature was maintained at (25 ±1°C) by using a 250 watt immersion heater with thermostat. The tank water was exchanged with fresh water 2-3 time/week. Tanks were washed and changed with fresh water every four weeks.

Experimental fish:

Nile tilapia fingerlings (*Oreochromis niloticus*) fingerlings were used in this study with mean average initial weight of 9.9±0.02 g were obtained from Fish Research Center, Suez Canal University. Fish were homogenous in body weights and apparently healthy. Fish were acclimated to laboratory conditions for 2 weeks prior before the experiment study.

Experimental Methodology:

Samples of fish:

At the start and the end of the main experimental period (56 day), 5 fish were randomly taken from each experimental group. Fish were used for chemical analysis of the whole body. The tested diets and body were analyzed for crud protein (CP %) ether extract (EE %), crude fiber (CF %), ash (%) and moisture. The whole body composition of fish samples were analyzed except crud fiber (CF %) according to the procedures described by standard A. O. A. C. methods (1995). The nitrogen free-extract (NFE %) was calculated by differences.

Fish were individually weighed every two weeks by UWE MJW-300 balance to the nearest 0.01g. The amount of diet was readjusted according to the new weight of fish.

Growth performance parameters:

The growth performance parameters are calculated according to the following equations:

Average Weight Gain (AWG):

(AWG) = Average final weight (g) – Average initial weight (g)

Average Daily Gain (ADG):

(ADG) = [Average final weight (g) – Average initial weight (g)] / time (days)

Specific Growth Rate (SGR %/day):

(SGR % / day) = 100 [Ln Wt1 – Ln Wt. 0 / t]

Where: - Ln: normal log Wt. 0: initial weight (g).

Wt. 1: final weight (g) T: time of days.

Feed and protein utilization parameters:

Feed and protein utilization parameters are calculated according to the following equations:

Feed Conversion Ratio (FCR):

FCR = Total feed consumption/ weight gain.

Feed Efficiency (FE):

FE = weight gain/ Total feed consumption

Survival (%):

SR=Ni x 100/N0

Where:

Nt = Total number of fish survived in tank at end of experiment.

N0 = Total number of fish survived in tank at beginning of experiment.

Blood parameters:

Blood samples were collected at the end of experiment; fish in each aquarium were weighted and 5 fish were taken randomly for blood sampling. After transferring Eppendorf tubes containing 4 ml blood to laboratory and allowed to clot at room temperature. Separation of serum from blood cells was carried out by centrifuge (Model 200 Labofuge, Heraeussepach Company, made in Germany) in 3000 rpm for 5 minutes. Then using Pastor Pipette, serum was transferred to marked Eppendorf and was maintained in -20 °C until measurement (Pottinger and Carrick, 2001) for total protein, albumin, triglycerides, The activities of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were carried out using Colorimetric method, Roch Diagnostics, GmbH, Monnheim, Germany kits according to (Reitman and Frankel, 1957). Serum total protein (g/dl) and albumin (g/dl) were determined calorimetrically using kits supplied by Elnasr Pharmaceutical Chemicals Co. (Egypt). Serum globulin (g/dl) levels were obtained by differences between total protein (g/dl) and albumin (g/dl). Serum triglycerides (mg/dl) were determined calorimetrically using commercial kits of Biodiagnostic Co. (Egypt). Urea was determined by urea Assay kit. Creatinine was determined according to (Pincus, 1996).

Statistical analysis:

The data obtained in this study were analyzed by one-way ANOVA procedure of Statistical Analysis System (SAS, 2000). Means were compared by Duncan's new multiple ranges test Duncan (1955).

Where:

$$Y_{ij} = \mu + D_i + e_{ij}$$

Y_{ij} = the observation of the j^{th} individual from D^{th} Diet.

μ = the overall mean. D_i = The Fixed effect of the D^{th} Diet.

e_{ij} = The random error associated with the individual j .

Economical Evaluation:

The cost of feed to raise unit biomass of fish was estimated by a simple economic analysis. The estimation was based on local retail sale market price of all the dietary ingredients at the time of the study.

Cost /kg diet (LE) = Cost per Kg diet L.E.

Consumed feed to produce 1kg fish (kg) = Feed intake per fish per period/ final weight per fish Kg/Kg

Feed cost per kg fresh fish (LE) = Step 1X step 2

Relative % of feed cost/ kg fish = Respective figures for step 3/ highest figure in this step

Feed cost /1Kg gain (LE) = Feed intake per Kg gain X step 1

Relative % of feed cost of Kg gain = Respective figures for step 5/ highest figure in this step

*Cost of 1 kg ingredients used were 3.50 L.E, Egypt Feed Ingredients Price at start of 2014.

RESULTS AND DISCUSSION

Growth Performance:

The effects of Spirulina supplementation on growth performance and feed utilization of Nile Tilapia (*Oreochromis niloticus*) fingerlings fed tested diets is presented in Table (4). No significant differences ($P < 0.05$) in initial body weight were found among the different experimental treatments, indicating the accuracy of randomization process between the experimental treatments. It was observed that groups of fish fed the diet containing Spirulina

showed higher values of body weight, weight gain, relative growth rate (RGR.), specific growth (SGR.), compared to the group of fish fed with the diet without Spirulina (control group).

Significant difference ($P < 0.05$) in BWG levels between the dietary groups compare to the control which all treatments had higher BWG compare to the control treatment during all experimental period ($P < 0.05$). Specific growth rate (SGR) is the measuring tools reflecting the fish health status under natural and experimental conditions. In the current study dried *S. platensis* found to be of potential effects on growth at an optimum replacement of 20%. Its worth mentioned that the mean value level in the group received 20% was higher in all the tested parameters with significant difference ($P < 0.05$) (Table, 4) to enhance growth performance. (Duncan and Klesius, 1996) reported that Spirulina alga was a good source of protein for animal feed, being containing high amounts of vitamins and minerals, in addition, (Nakono et al., 2003). Olvera-Novoa et al., (1998) reported that Spirulina can replace up to 20% of FM protein in diets for *O. mossambicus*, but reduced growth and feed utilization were observed at higher replacement levels to 100 and 60% by Spirulina in diets for *C. carpio* and Siberian sturgeon (*Acipenserbaeri*), respectively, without any adverse effects (Nandeesh et al., 1998 and Palmegiano et al., 2005). Other studies suggested that dietary FM can be replaced up to 100 and 60% by Spirulina in diets for *C. Carpio*; Siberian sturgeon (*Acipenserbaeri*) and Nile tilapia, respectively, without any adverse effects (Nandeesh et al., 1998; Palmegiano et al., 2005 and El-Sheekh et al., 2014).

Table 4. Growth performance and feed utilization (of Nile tilapia (*Oreochromis niloticus*) fingerlings fed diets supplemented with different levels of Spirulina

Items	Spirulina levels (%)				
	Control* T1 (0%)	T2* (5%)	T3* (10%)	T4* (15%)	T5* (20%)
Initial weight (g)	9.91±0.11	9.89±0.10	9.90±0.11	9.92±0.12	9.91±0.14
Final weight (g)	17.20 ^c ±0.12	18.90 ^d ±0.10	21.60 ^c ±0.13	26.50 ^b ±0.10	28.20 ^a ±0.11
Weight gain (g)	7.29 ^e ±0.10	9.01 ^d ±0.10	11.70 ^c ±0.10	16.58 ^b ±0.10	18.29 ^a ±0.10
Weight gain (%)	73.56 ^e ±0.10	91.10 ^d ±0.10	118.30 ^c ±0.10	167.71 ^b ±0.10	184.56 ^a ±0.10
Specific growth rate (%/day)	1.24 ^d ±0.11	1.15 ^e ±0.12	1.40 ^c ±0.15	1.76 ^b ±0.14	1.87 ^a ±0.11
Feed intake (g feed/fish)	18.75 ^e ±0.10	21.53 ^d ±0.12	25.62 ^c ±0.14	35.83 ^a ±0.11	33.43 ^b ±0.12
Food conversion ratio	2.57 ^e ±0.10	2.39 ^d ±0.11	2.19 ^c ±0.13	2.16 ^b ±0.10	1.83 ^a ±0.14
Protein efficiency ratio	1.27 ^d ±0.10	1.36 ^e ±0.11	1.49 ^c ±0.13	1.50 ^b ±0.14	1.78 ^a ±0.11
Feed efficiency	0.38 ^d ±0.10	0.42 ^c ±0.13	0.45 ^b ±0.11	0.46 ^b ±0.11	0.55 ^c ±0.13
Survival rate (%)	98.00 ^a ±0.80	98.00 ^a ±0.80	98.00 ^a ±0.70	98.00 ^a ±0.60	98.00 ^a ±0.10

Means±SD in the same letters in the same row is not significantly different at $P < 0.05$.

*Note: Control diet was without *Spirulina platensis* (0%) and T1, T2, T3 and T4 were replaced by 5%, 10%, 15% and 20% *Spirulina platensis* from diet protein, respectively.

Growth enhancement effect of Spirulina is attributed to its role in nutrient digestibility and its high contents of several nutrients, such as vitamins and minerals (Abdel-Tawwab and Ahmad, 2009 and Velasquez and Ragaza, 2015). Negative effects of high dietary inclusion levels of Spirulina on fish

growth can be resulted from reduced phosphorous availability and decreased feed palatability (Olvera-Novoa et al., 1998). The variations in Spirulina effects on fish growth performance are ascribed to different nutrient content of Spirulina species used in the studies (Nandeesh et al., 1998). Nandeesh et

al. (2001) found that FM can be totally replaced with Spirulina in diets for rohu carp (*Labeo-rohita*) and even significant higher growth can be obtained compared to the use of FM as the sole protein source, whereas nonsignificant effect was observed on growth performance of catla (*Catlacatla*) by the same Spirulina supplemented diets. Such differences in growth response of *L. rohita* and *C. catlato* dietary Spirulina were clearly showed that the growth response of fishes to Spirulina is likely to be species-specific. The other significant factor that affects the results of Spirulina administration is the composition of experimental diets in which Spirulina is incorporated (Takeuchi *et al.*, 2002). Another study was conducted in which 10-15% Spirulina alga replaced crushed fish in the feed given to Red Tilapia fish in order to bring the protein level up to 30% and this yielded the highest average daily growth rate and specific growth rate, in the Tilapia (Promya *et al.*, 2003). Similar research was done by Promya and Hongwittayakorn (2003) in which Spirulina and *Cladophora* algae were fed to fancy Carp fingerlings. The study reported that, the fish received the Spirulina powder demonstrated a higher average daily growth rate, weight gain rate, survival rate, than the fancy Carp that were fed with standard feed and feed mixed with *Cladophora*.

Research conducted by Duncan and Klesius (1996) found that *Spirulina* alga was a good source of protein for animal feed, as well as containing high amounts of vitamins and minerals. The cellular structure of Spirulina algae is easily digestible and contains a little bit cellulose.

The feed conversion rate, PER, FR and biomass gain of Nile tilapia fingerlings are reported in (Table 4). The growth performance showed that Spirulina promotes growth better than the control diet, and in particular 20% inclusion seems to be the best performance: a high biomass increase and growth rate, the best feed conversion rate and a high PER. The results are in agreement with El-Sheekh *et al.* (2014) and, Zeinab *et al.* (2015).

The average values of FCR show a significant ($P<0.05$) different between all treatments. The group of fish on T4 had significant ($P<0.05$) lower FCR (1.83), while the group of fish on control had the highest FCR (2.40). The results in the current study are in accordance with Watanabe *et al.* (1990) and Takeuchi, *et al.* (2002) who found that feed supplemented with *S. platensis* powder improved the feed conversion ratio and growth rates in striped jack, *Pseudocaranx dentex*. Lu *et al.* (2002) demonstrated that raw *S. platensis* can be an effective -feed for larval tilapia at a feeding rate of 30% (on a dry basis) of body weight. Abdel-Tawwab and Ahmed (2009) recorded that the growth and feed utilization of *O. niloticus* were obtained at 5.0 g fresh culture of *S. platensis* /kg diet. Our results show that, treatments with Spirulina platensis had feed conversion ratios (FCR) significantly ($P<0.05$) lower than those for the

control diets. The best conversion ratio (1.83) was recorded for the 20% diet of Spirulina platensis treatments. It is proved that lower the feed conversion ratio higher is the quality of the feed as observed in many fresh water fishes where the feed conversion ratio is known to decrease with increasing dietary protein content (Jha *et al.*, 2009 and Zeinab *et al.*, 2015). Similar results have been reported for feed additives used in diets for piglets (Gil, 1998). Moreover, the group of fish on T4 had significantly ($P<0.05$) higher PER, FI and FE while the group of fish on control had the lowest PER, FI and FE, this agree with the finding of Badwy *et al.* (2008) the incorporation 50% algae replacement resulted in the significant greater value of FCR (2.03 ± 0.08 and 1.76 ± 0.05), respectively. These results are agreed with those obtained by Dawah *et al.* (2002) and Jha *et al.* (2009). Who found that food conversion ratio and PER were better when the fish were maintained on artificial diets with 10% and 20% dried algae. In addition, Zeinhom (2004) found that, Inclusion of algae in fish diets insignificantly ($P<0.05$) improved the FCR (1.83), PER and (1.76), whereas feed intake was significantly ($P<0.05$) increased (33.47g) Table (4). However, these results are good agreement with those obtained by Hayashi *et al.* (1998) and Abu-Zead (2001) who found that the protein efficiency ratio ranged from 1.1 to 1.7 for Nile tilapia and common carp fed on diets containing aquatic plant and algae, while Ibrahim *et al.* (2007) reported that, feed conversion ratio gradually increased with increasing Azolla meal percentage in the diets without significant differences until 31.8% inclusion level after that, significantly ($P<0.05$) decreased, they added that, economical feed efficiency improved as the level of the dietary Azolla meal increased from 10.6 to 31.8% of the diet. Duncan and Klesius (1996) reported that Spirulina alga was a good source of protein for animal feed, being containing high amounts of vitamins and minerals, in addition, Nakono *et al.* (2003) recorded that the lack of cellulose from the cellular structure of Spirulina render it easily digestible, thus, increase fish appetite, improve feed intake and nutrient digestibility and in turn enhance the health of fish, increasing the ability to fight off infections through the reduction of stress levels. The results in the current study are in accordance with Watanabe *et al.* (1990) and Takeuchi *et al.* (2002) who found that feed supplemented with *S. platensis* powder improved the feed conversion ratio and growth rates in striped jack, *Pseudocaranx dentex*. Lu *et al.* (2002) demonstrated that raw *S. platensis* can be an effective -feed for larval tilapia at a feeding rate of 30% (on a dry basis) of body weight. Abdel-Tawwab and Ahmed (2009) recorded that the growth and feed utilization of *O. niloticus* were obtained at 5.0 g fresh culture of *S. platensis* /kg diet. Treatments with *Spirulina platensis* had significant ($P<0.05$) feed conversion ratios (FCR) lower than those for the control diets.

The best conversion ratio (1.83) was recorded for the 20% diet of *Spirulina platensis* treatments. The lower feed conversion ratio the higher is the quality of the feed as observed in many fresh water fishes where the feed conversion ratio is known to decrease with increasing dietary protein content (Jha et al., 2009 and Zeinab et al., 2015). Similar results have been reported for feed additives used in diets for piglets (Gil, 1998). Moreover, the group of fish on T5 had significant ($P < 0.05$) higher PER, FI and FE, while the group of fish on the T1 had the least PER, FI and FE, this agreed with the finding of Badwy et al. (2008) who reported that, the incorporation 50% algae replacement resulted in the significant greater value of FCR (2.03 ± 0.08 and 1.76 ± 0.05) respectively. These results are agreed with those obtained by Dawah et al. (2002) and Jha et al. (2009) who found that food conversion ratio and PER were better when the fish were maintained on artificial diets with 10% and 20% dried algae. In addition, Zeinhom (2004) found that, inclusion of algae in fish diets insignificantly ($P < 0.05$) improved the FCR (1.83), PER and (1.76), whereas feed intake was significantly ($P < 0.05$) increased (33.47g) (Table, 4). However, these results are in good agreement with those obtained by Hayashi et al. (1998) and Abu-Zead (2001) who found that the protein efficiency ratio ranged from 1.1 to 1.7 for Nile tilapia and common carp fed on diets containing aquatic plant and algae. Ibrahim et al. (2007) reported that, feed conversion ratio gradually increased with increasing Azolla meal percentage in the diets without significant differences until 31.8% inclusion. They added that economical feed efficiency improved as the level of the dietary Azolla meal increased from 10.6 to 31.8% of the diet. Duncan and Klesius (1996) reported that *Spirulina* alga was a good source of protein for animal feed, and contained high amounts of vitamins and minerals. Nakono et al. (2003) recorded that the small amount of cellulose from the cellular structure of *Spirulina* render it easily digestible, and increase fish appetite, improve feed intake and nutrient digestibility and in turn enhance the health of fish, increasing the ability to fight off infections through the reduction of stress levels. The results in the current study are in accordance with Watanabe et al. (1990) and Takeuchi et al. (2002)

who found that feed supplemented with *S. platensis* powder improved the feed conversion ratio and growth rates in striped jack, *Pseudocaranx dentex*. Lu et al. (2002) demonstrated that raw *S. platensis* can be an effective feed for larval tilapia at a feeding rate of 30% (on a dry basis) of body weight. This contributes to optimizing protein use for growth, a significant quality given that protein is the most expensive feed nutrient. The improvement in the biological value of the supplemented diets in these treatments with high population and low dietary protein demonstrated that *Spirulina* supplements performed more efficiently in stress situations (Ringo and Gatesoupe, 1998). No significant differences in fish survival among the different treatments ($P < 0.05$), and its 98% (Table 4). Some studies have shown that feeding *Spirulina* to fish could improve their survival rate and growth rate (Belay et al., 1996; Hayashi et al., 1998 and Tongsiri et al., 2010).

Fish body composition:

Spirulina significantly ($P < 0.05$) was affected whole-fish body composition (Table 5). Fish fed the control diet had the lowest protein content; however, all feed replacement with *Spirulina* appeared to improve their protein body content. Carcass lipid content was also affected by dietary treatments, being the highest in the group T4, which were statistically ($P < 0.05$) the highest than the rest of dietary replacement treatments. The supplementation of live *Spirulina* significantly ($P < 0.05$) affected the protein and lipid contents in the whole-fish body. The better feed intake of *Spirulina* diets may be due to the increased fish appetite, resulting in a higher feed intake and improved growth. On the other hand, changes in the protein and lipid contents in the fish body could be linked to changes in their synthesis, deposition rate in muscle and/or different growth rates, high feed intake and nutrient utilization, (Abdel-Tawwab and Ahmad, 2009). These results agreed with Soivio et al. (1989) who concluded that the differences in protein content of fish body composition may be related to changes in their synthesis, deposition rate in muscle and/or different growth rate.

Table 5. Body chemical composition of Nile tilapia (*Oreochromis niloticus*) fingerlings fed diet supplemented with different levels of *Spirulina* on dry matter basis

Chemical Composition	Body chemical composition (%)			
	Moisture (%)	Crude protein (%)	Ether extract (%)	Ash (%)
Initial	73.4 \pm 0.27	61.2 \pm 0.27	22.8 \pm 0.27	16.6 \pm 0.27
Control*	72.5 ^a \pm 0.27	62.2 c \pm 0.32	23.1 c \pm 0.18	14.7 a \pm 0.13
T1*	72.5 ^a \pm 0.22	62.6 c \pm 0.40	23.7 c \pm 0.12	13.7b \pm 0.23
T2*	72.5 ^a \pm 0.25	63.3 b \pm 0.21	23.8 c \pm 0.14	12.9c \pm 0.22
T3*	72.5 ^a \pm 0.22	63.7 b \pm 0.41	24.1 b \pm 0.16	12.2c \pm 0.20
T4*	72.5 ^a \pm 0.24	64.5a \pm 0.51	25.5 a \pm 0.18	11.0e \pm 0.22

* Control diet was without *Spirulina platensis* (0%) and T1, T2, T3 and T4 were replaced by 5%, 10%, 15% and 20% *Spirulina platensis* from diet protein, respectively.

Note: Means in the same row having different superscripts are significantly different ($P < 0.05$).

Table 6. Effect of Spirulina levels on blood serum components of Nile Tilapia (*Oreochromis niloticus*) fingerlings at the end of experiment

Item	Treatments				
	Control*	T (1)*	T (2)*	T (3)*	T (4)*
Total Protein (g/dl)	2.28 ^b ±0.12	2.36 ^a ±0.11	2.40 ^a ±0.10	2.41 ^a ±0.12	2.44 ^a ±0.12
Albumin (g/dl)	0.85 ^d ±0.12	0.90 ^c ±0.11	0.95 ^b ±0.12	0.98 ^b ±0.12	1.00 ^a ±0.11
Globulin (g/dl)	1.43 ^e ±0.12	1.46 ^d ±0.15	1.45 ^c ±0.10	1.43 ^b ±0.12	1.44 ^a ±0.13
Albumin/Globulin Ratio	0.59 ^a ±0.12	0.62 ^b ±0.12	0.66 ^b ±0.12	0.68 ^c ±0.12	0.69 ^d ±0.12
Serum Creatinine(Mg/dl)	0.31 ^c ±0.12	0.35 ^b ±0.10	0.38 ^b ±0.11	0.40 ^a ±0.12	0.44 ^a ±0.12
Urea (Mg/dl)	2.2 ^c ±0.11	2.50 ^d ±0.12	3.00 ^c ±0.14	3.40 ^b ±0.11	3.60 ^a ±0.12
Triglycerides (mg/dl)	88.00 ^c ±0.10	90.00 ^d ±0.12	93.00 ^c ±0.12	96.00 ^b ±0.11	97.00 ^a ±0.11
HDL-C (mg/dl) 1	31.00 ^c ±0.11	33.00 ^d ±0.10	39.00 ^c ±0.10	42.00 ^b ±0.10	46.00 ^a ±0.10
LDL-C (mg/dl) 2	81.00 ^a ±0.12	79.00 ^b ±0.12	77.00 ^c ±0.12	59.00 ^d ±0.12	56.00 ^c ±0.12
ALT3 (U/dl)	46.00 ^a ±0.12	44.00 ^b ±0.12	42.00 ^c ±0.12	41.00 ^d ±0.12	39.00 ^c ±0.12
AST4(U/dl)	125.00 ^a ±0.11	124.00 ^b ±0.12	122.00 ^c ±0.12	119.00 ^d ±0.11	117.00 ^c ±0.10

*Note: Control diet was without *Spirulina platensis* (0%) and T1,T2,T3 and T4 were replaced by 5%,10%,15% and 20% *Spirulina platensis* from diet protein, respectively

Note: Values are means ± SD of three replications. Means in the same row having different superscripts are significantly different (P <0.05).

1. (HDL-C) High – density lipoprotein cholesterol

2. (LDL-C) Low – density lipoprotein cholesterol

3. ALT (SGPT) Alanine aminotransferase

4. (AST) aspartate aminotransferase

Blood biochemical components:

Fish fed diets containing different spirulina levels exhibited higher protein, serum creatinine, urea, albumin, globulin, triglycerides HDL-C values (Table 6).

Diets containing different Spirulina levels were significantly (P<0.05) decreased albumin /globulin ratio LDL-CAST, ALT and AST values compared with the control. Serum protein (g/dl), creatinine (mg/dl), urea (mg/dl) albumin (g/dl), globulin (g/dl) and triglycerides (mg/dl) values increased significantly (P<0.05) with the increase of Spirulina levels and the highest values were obtained at T4 (20% Spirulina/kg) diet.

These results proved the improvement of fish health when fed Spirulina-supplemented diets. Moreover, the measurement of albumin, globulin, and total protein in serum or plasma is of considerable diagnostic value in fish, as it affects the general nutritional status as well as the integrity of the vascular system and liver function (Schaperclaus *et al.*, 1992).

These results suggested an improvement of fish health when fed with feed additives supplement. The measurement of AST and ALT in plasma is of considerable diagnostic value in fish, as it relates to general nutritional status as well as the integrity of the vascular system and liver function. This result agrees with Abdel-Tawwab *et al.* (2008) who investigated the use of commercial baker yeast and Spirulina as a growth and immunity promoter for Nile Tilapia; they found that biochemical parameters were improved in fish fed yeast and Spirulina. These results are in agreement with the previous study in tilapia recorded by Metwalli (2013) and other species such as sturgeon (Yue and Zhou, 2008) and sunshine bass (Zhu *et al.*, 2011).

Economic Evaluation:

Calculations of economic efficiency of the tested diets were based on the cost of feed. The costs of one Kg gain in weight and its ratio with the control group are recorded in Table (7).

Table 7. Economic analysis of Nile tilapia *O. Niloticus* fingerlings supplemented with different levels of Spirulina for 56 days

Item	Treatments				
	Control*	T1*	T2*	T3*	T4*
Cost /kg diet (LE) ¹	3.65	3.56	3.50	3.41	3.29
Consumed feed to produce 1kg fish (kg) ²	1.23	1.14	1.19	1.35	1.19
Feed cost per kg fresh fish (LE) ³	4.49	4.06	4.16	4.60	3.92
Relative % of feed cost/ kg fish ⁴	97	88	90	100	85
Feed cost /1Kg gain(LE) ⁵	10.59	8.59	7.66	7.37	6.02
Relative % of feed cost of Kg gain ⁶	100	81	72	70	57

*Note: Control diet was without *Spirulina platensis* (0%) and T1,T2,T3 and T4 were replaced by 5%,10%,15% and 20% *Spirulina platensis* from diet protein, respectively

1. Cost /kg diet (LE) = Cost per Kg diet L.E. 2. Consumed feed to produce 1kg fish (kg) = Feed intake per fish per period/ final weight per fish Kg/Kg.

3. Feed cost per kg fresh fish (LE) = Step 1X step 2.

4. Relative % of feed cost/ kg fish = Respective figures for step 3/ highest figure in this step.

5. Feed cost /1Kg gain (LE) = Feed intake per Kg gain X step 1.

6. Relative % of feed cost of Kg gain = Respective figures for step 5/ highest figure in this step.

7. Component cost/kg: commercial fish meal (4.51/k g); commercial Spirulina(1051/kg); corn meal (1.81/kg); Rice bran (1.51/kg); sunflower oil (8.00/kg); mineral mix (1.51/kg) and vit. Mix. (3.51/kg).

The lowest feed cost per kg fresh fish (3.92 LE), Relative % of feed cost / kg fish (85%), Feed cost /1Kg gain(6.02 LE) and Relative % of feed cost of Kg gain (57%) in group of fish fed diet 4 (20% Spirulina). This indicates that treatment 4 which supplemented with 20% Spirulina is most economic than the rest of dietary treatments.

In conclusion, this study demonstrates that up to 20% of Spirulina were most effective in stimulating fish growth and more economic for Nile tilapia (*Oreochromis niloticus*) fingerlings under the experimental condition.

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مقاييس نمو اصبعيات سمك البلطي النيلي المغذاة على علائق مدعمة بمستويات مختلفة من طحلب السبيرولينا

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أجريت هذه الدراسة بمركز بحوث الأسماك- جامعة قناة السويس- بهدف دراسة تأثير استخدام بروتين طحلب السبيرولينا بنسب مختلفة من بروتين العليقة لإصبعيات سمك البلطي. حيث تم اضافة السبيرولينا عند خمس مستويات من بروتين العليقة (٠، ٥، ١٠، ١٥ و ٢٠%) متوازنة في النيتروجين والسعرات الحرارية. تمت التغذية حتى الشبع مرتين يوميا لمدة ٥٦ يوم. وتم دراسة تأثيرها على أداء النمو، الاستفادة الغذائية ومكونات الجسم والتحليل الكيميائي للدم والتقييم الاقتصادي. وسجلت المعاملة الخامسة (٢٠% سبيرولينا) أفضل زيادة معنوية في الوزن النهائي، معدل النمو النوعي، الكفاءة التحويلية للغذاء، الكفاءة الغذائية، معدل استهلاك الغذاء، معامل الاستفادة من البروتين، وأعلى معدل حيوية عند مستوى معنوية ($p < 0.05$) يليها المعاملات ١، ٢، ٣، ٤ على التوالي. ووجد ان العلائق التجريبية المستخدمة في التجربة أثرت معنويا ($p < 0.05$) على التحليل الكيماوي لمكونات الجسم (البروتين والدهن) للأسماك المغذاة بنسبة ٢٠% سبيرولينا مقارنة بالمعاملات الأخرى. كما اظهرت النتائج تحسن في صحة الأسماك نتيجة عملية الاحلال الغذائي، حيث ارتفعت قيم كل من البروتين الكلي للسيرم والكرياتين واليوربا والألبومين والجلوبيولين والجليسريدات الثلاثية والكلسترول المرتفع الكثافة بينما انخفضت قيم الألبومين /الجلوبيولين والكلسترول منخفض الكثافة وانزيمات الكبد (ALT, AST) مقارنة بالعينة المقارنة عند زيادة نسبة السبيرولينا وأعلى القيم كانت بالمعاملة الخامسة. ويستنتج من هذه الدراسة التدعيم ببروتين طحلب السبيرولينا بنسبة ٢٠% من بروتين الصويا كان الافضل تأثيرا على نمو اصبعيات سمك البلطي النيلي وكذا الناحية الاقتصادية تحت ظروف هذه التجربة.