EFFECT OF PROTEIN LEVEL ON SPAWNING PERFORMANCE OF NILE TILAPIA *OREOCHROMIS NILOTICUS* IN EGYPT

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Abstract

The effect of protein levels (15, 25 and 35% protein) on spawning performance of Nile tilapia (*Oreochromis niloticus*) were investigated in outdoor tanks. Spawning was monitored; the data on egg characteristics (fecundity and egg size) were recorded.

The results indicated that increasing the protein level fed from 15% to 35% increased significantly the body weight and fecundity of *O. niloticus* females. On the other hand, supply of inadequate protein (15% protein level) resulted in prolonged intervals between spawning. Means of egg diameter between and within treatment were not significantly different. The present study shows the important of high-protein diet in enhancing reproductive performance of Nile tilapia. Also, these findings give fish farmers an option in the management of feeding of Nile tilapia brood stock.

Based on our results, we suggest that a minimum of 35% protein be included in the diet female Nile tilapia brood stock

Key Words: Nile tilapia, protein level, tank culture, spawning performance.

INTRODUCTION

Among all cultured tilapia species, Nile tilapia (*Oreochromis niloticus*) has emerged as the single most important species. The attributes which make Nile tilapia so suitable for fish farming are its general hardiness, ease of breeding, rapid growth rate, ability to convert efficiently organic wastes into high quality protein and good taste (Yi *et al.*, 1996) and resistance to environmental stress.

Reproducing output is maintained at the expense of somatic growth. However supply of inadequate protein for long periods results in slow ovarian recrudescence (Gunasekera and Lam, 1997), prolonged intervals between spawning (Gunasekera *et al.*, 1996) and a complete halt to reproduction. Brood fish should, therefore, be provided with optimum levels of protein in the diet from a young age up to the egg producing stage

In a study carried out by (Gunasekera *et al.*, 1995), Nile tilapia fed a low protein diet (17%) did not show oocyte maturation, and females fed 25% protein diet showed slow oocyte growth, where as females fed > 32% protein level had early oocyte

growth and maturation. The 20% dietary protein displayed lowest protein content in both ovaries and muscle of female swordtail brood stock (Alexander, 2004)

Fecundity is very flexible in tilapia fish. It is responsive to food availability and time. It is the biological item concerned with estimating and counting the more advanced groups of eggs depending on the fact that these groups are only spawned in the spawning season (Asem, 1992).

Wee and Tuan (1988) found that , the absolute and relative fecundities of *O. niloticus* were found to be significantly higher in the fish fed on medium dietary protein levels (27.6 and 35%) than those fed on the higher protein levels (42.6 and 50.1 %). Brood stock nutrition is still poorly understood due difficulties in conducting studies involving proper feeding and reproduction of brood stock. The current study was design to investigate the effects of dietary of protein level on reproductive performance of Nile tilapia.

MATERIALS AND METHODS

Diets

Three isocaloric diets (T1, T2 and T3) containing three protein levels 15, 25 and 35% were formulated utilizing fish meal and soybean meal as protein sources (Table 1). Proximate analysis of diets was conducted according to AOAC (2000) for verification of nutrient levels. The pellets were stored at – 20°C until used.

Fish

The female and male of *O. niloticus* used in this experiment were obtained from Fish Research Center, Suez Canal Univ. form the same brood stock. Fish were fed a commercial diet until they were sexually mature.

A total number of inserted 144 females of *O. niloticus* were distributed randomly into nine tanks (300 liters), each stocked with 16 individuals (each 70 gm, body weight) and fed with one of 15, 25 and 35% protein diet to each tank (3 replicate in each treatment). All females were tagged with plastic number in selected into dorsal fin. After 2 months of feeding and at least five spawning, 10 female from each dietary protein level were randomly assigned to new tank. One female and one male were kept in each tank and they were kept separated by a partition frame across each tank in order to avoid male aggression outside the mating period. It will know that *O. niloticus* females usually continue their spawning cycle even in the absence of a male. The males were introduced into the tanks to ensure success of spawning cycle (one male to 3 female).

The fish in each tank were fed twice daily at rate 2% of body weight (two times/day). The females were observed 4 times daily to check whether they had

spawned. Spawning was recorded when actual spawning was observed, or when eggs were seen in the mouth of the brooder. Eggs were removed from brooder's becalm cavity not later than 1 h after spawning and the number of eggs in each spawn determined and the brooder's body weight was recorded. This spawns were used for determination of all the parameters studied. However, those spawns resulting from the non-observation period cap (proximately between 2.00 and 06 .00 h) were used for fecundity estimation only.

The spawning intervals for each female were recorded and the 1st spawning intervals was considered as a number of days between 1st and 2nd spawning after females were transferred into individual tanks. Four spawning, hence four spawning intervals, were monitored for each treatment.

The data on egg characteristics, viz. fecundity egg size (diameters) were collected from the first 4 spawning only.

Females were transferred back to the same tank and observation data collections were repeated for another four spawning.

The experiment was lasted for 8 month with regular and careful observation (from March 2005 to November 2005).

Statistical Analysis:

All data were analyzed by analysis of variance and Duncan's multiple range tests using SAS program (1996).

RESULTS AND DISCUSSION

Mean spawning interval for four consecutive spawning in *O. niloticus* fed diets containing three protein levels are presented in Table 2. The averages of spawning interval for four consecutive spawning were 22.5; 28.4; 44.3 and 44.2 for treatment one (15% protein level) and 20.2; 20.7; 20.2 and 21.3 for treatment two (25% protein level) and 21.3; 19.6; 19.8 and 20.8 for treatment three (35% protein) respectively. These results indicate that, mean spawning interval decreased with increasing of protein level. The differences between the means were significant Table 2. These results are in accordance with those obtained by Gunaskera *et al.,* (1996) and Abee *et al.,* (1990) who found that supply of inadequate protein for long period's results in prolonged intervals between spawning.

Mean of body weight for four spawning of *O. niloticus* females fed three protein levels are presented in Table 3. The averages of body weight for four consecutive spawning were 70.3; 72.2; 70.3 and 75.2 g. for treatment one (15% protein level) and 75.4; 90.5; 100.3 and 100.2 g for treatment two (25% protein level) and 96.3; 100.2; 120.4 and 135.2 g. for treatment three (35% protein level), respectively. These

results indicate that, mean of body weight increased gradually with each increasing of protein level. The differences between the means were significant (Table 3). These results are agreement with these obtained by Abella *et al.*, 1990 , and Gunaskera *et al* ., 1995 who reported that Nile tilapia attained puberty and oocyte maturation earlier when fed higher levels of dietary protein and concluded that this was due to the effect of diet on fish growth. Moreover, the group of fish fed a low protein diet (15%) did not show good maturation, this indicates limited or insufficient protein for maintenance and oocyte development. In tilapia total weight increase with higher protein level. Larger female have also been reported to display higher success in spawning rates which is in agreement with El-Sayed *et al.* (2003). Also, Ahmed (2006) reported that female and male Nile tilapia average weight 300g each is better weight for reproductive traits

Mean of egg diameter for four spawning of *O. niloticus* females fed three protein levels are presented in Table 3. The averages of egg diameter for four consecutive spawning were 2.25 ; 2.26 ; 2.30 and 2.35 mm for treatment one (15% protein level) and 2.37 ; 2.25 ; 2.30 and 2.36 mm for treatment two (25% protein level) and 2.39 ; 2.33 ; 2.40 and 2.36 for treatment three (35% protein level), respectively .These results indicate that means of egg diameter between and within treatment were not significantly different ($P< 0.05$). In this Connection Watanab, (1985), kjorsvik *et al.* (1990) and Bhujel, (2000) reported that feed quality, protein quality, lipid composition and vitamins have an effect on egg and larval viability.

Means of fecundity at four consecutive spawning of *O. niloticus* females fed three protein levels are presented in Table 4. The averages of fecundity for four consecutive spawning were 330 ; 320 ; 320 and 316 for treatment one (15% protein level) and 390 ; 440 ; 480 and 480 for treatment two (25% protein level) and 440 ; 520 ; 520 and 630 for treatment three (35% protein level), respectively. These results indicate that, means of fecundity increased gradually with each increasing of protein level .The differences between the means were significant (Table 4).

Mean s of number of eggs per g body weight (relative fecundity) are presented in Table (4) .The number of eggs for four consecutive spawning were 4.5 ; 4.3 ; 4.4 and 3.8 for treatment one (15% protein level) and 4.7 ; 4.8 ; 4.6 and 4.3 for treatment two (25% protein level) and 4.9 ; 5.2 ; 4.2 and 4.6 for treatment three (35% protein level) , respectively . These results indicate that means of number of eggs per g. body weight were not significantly different ($p<0.05$). These results are partly agreement with those obtained by Bhujel, (2000) and Wee and Tuan (1988) who found that, the absolute and relative fecundities of *O. niloticus* were found to be significantly higher in the fish fed on medium dietary protein levels (27.6 and 35%)

than those fed on the higher protein levels (42.6 and 50.1%) also Rana(1988), reported that, absolute fecundity is related to body weight , while De Silva (1986) found that absolute fecundity is related to body length .Also Estay *et al* ., (1997) and Alexander *et al.*,(2004) reported that , the relative fecundity decreased with increasing of female body weight.

Table 1. Composition and proximate analysis of experimental diets.

Ingredients	Dietary Protein level (%)		
	15	25	35
Fish meal (7o %)	3	4	5
Soybean meal (44%)	18	43	70
Yellow corn (8%)	71	45	17
Vegetable oil	6	6	6
Vitamin mixture(1)	1	1	1
Mineral premix(2)	1	1	1
Total	100	100	100
Chemical composition % on dry matter basis			
Moisture	6.45	7.25	8.61
Protein	15.70	25.40	35.6
Ash	4.85	5.94	7.43
Ether extract	8.90	9.70	8.09
Crude fiber	2.97	4.20	5.55
NFE(3)	67.65	54.76	43.33
Gross energy kcal/g diet(4)	4.49	4.58	4.11

(1)Vitamins mixture contained (as g/ kg premix): Thiamine 2.5; Riboflavin 2.5, pyridxine 2.0 Inositol 100.0; Biotin 0.3; pantothenic acid 100.0; folic acid 0.75; Para-aminobenzioic 2.5Choline 200.0 Nicotinic acid 10. Cyanocobalmine 0.005; tocopherol acetate 20.1; Ascorbic acid 50.0; Mennadione 2.0.; Retinol palmitate 100.000IU ; Cholecalciferol 500.000 IU.

(2) Minerals premix (as g/ kg of premix) ChaHpo4.2H20 727.7775; MgSo4; H20 127.5; Kcal 50.0; Nacl 60.; FeSo4. 7H20 25.; ZnSo4. 7H20 5.5; MnSo4. 4H20 2.53; CuSo4.5H200.785; CoSo4. 7 H20 0.4775; Calo3.6H2 0.295; CrC13.6H20 0.1275

(3) NFE (Nitrogen free Extract) = 100 – (protein + lipid + fiber+ ash).

(4) Gross energy were calculated, the energy value (Kcal/ g) for protein 5.65; for lipid 9.45 and carbohydrate 4.1.

Table 2. Mean spawning interval for four consecutive spawning in *O. niloticus* fed diets containing three protein levels. (the data are based on the spawning of 10 fish for each treatment).

Protein Level	Spawning interval			
	1	2	3	4
15%	22.5a	28.4a	44.31a	44.2a
25%	20.2a	20.7b	20.2b	21.3 b
35%	21.3a	19.6b	19.8b	20.8b

* Values with the same superscript in each column are not significantly different (P <0.05)

Table 3. Mean of body weight and egg diameters four spawning of *O. niloticus* females fed three protein levels

Protein level	1 st. spawning	2 nd. spawning	3 rd. spawning	4 th. spawning
(%)	Body weight (g)			
15	70.3 c	72.2c	70.3c	75.2c
25	75.4 b	90.5 b	100.3b	100.2b
35	96.3 a	100.2a	120.4	135.2a
Egg diameter (mm)				
15	2.25	2.26	2.3	2.35
25	2.37	2.25	2.3	2.36
35	2.39	2.23	2.4	2.36

*Means in each column having the same super script letter are not significantly different (P<0.05) .

* Means of egg diameter between and within treatment were not significantly different (P<0.05)

Table 4. Means of Fecundity and relative fecundity at four consecutive spawning of *O. niloticus* females fed three protein levels (data one based on 10 spawning fish in each treatment)

Protein level (%)	1 st spawning	2 nd spawning	3 rd spawning	4 th spawning
Fecundity	330 c	320c	320 c	316c
15	390 b	440 b	480 b	480 b
25	440 a	520 a	520 a	630 a
35				
Number eggs per body weight (relative fecundity)				
15	4.5c	4.3c	4.4b	3.8c
25	4.7b	4.8b	4.6a	4.3b
35	4.9a	5.2a	4.2c	4.6a

* Means in each column having the same superscript letter were not significantly different (P< 0.05) .

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تأثير مستوى البروتين في العليقة على أداء التبويض في اسماك البلطي النيلي في مصر

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أجريت هذه التجربة بهدف دراسة اثر مستوى البروتين في العليقة (١٥ ،٢٥ ،٣٥ % بروتين خام)على أداء التبويض لأسماك البلطي النيلي المرباه في عدد ٩ تنكات حجم كل منهما ٣٠٠ لتر ، تم تسكين عدد ١٦ أم بوزن ٧٠ جرام للسمكة في كل تنك مع تكرار كل معاملة ثلاث مرات . تم تسجيل الصفات التناسلية (عدد البيض الناتج من كل أنثى ــ حجم البيض ــ عدد البيض لكل جرام من وزن السمكة ــ الفترة الزمنية البينية لوضع البيض).

وقد أوضحت النتائج الآتي:-

ــ زيادة مستوى البروتين في العليقة من ١٥ % إلى ٣٥ % أدى إلى زيادة ملحوظة في معدلات أوزان الأمهات المرباة وعدد البيض الناتج من كل أنثى ومن ناحية أخرى أدى تغذية الأسماك على أحتياجات بروتينية غير كافية (١٥ % بروتين) الى طول الفترة الزمنية بين التبويض والتبويض التالي له .

ــ عدم وجود تأثير ملحوظ لمستوى البروتين على متوسطات أحجام البيض داخل وبين المعاملات .

ــ أهمية المستوى العالي من البروتين (٣٥ %) في تنشيط الأداء التناسلي لأمهات اسماك البلطي النيلي وهذا يعطي المزارعين حرية اختيار نظم التغذية المناسبة لأمهات اسماك البلطي النيلي .