EFFECT OF PROTEIN LEVEL AND HAPA EXCHANGE TIMES ON GROWTH PERFORMANCE AND PROFITABILITY OF NILE TILAPIA (*Oreochromis niloticus*) FRY DURING SEX REVERSAL STAGE IN COMMERCIAL HATCHERY, FAYOUM GOVERNORATE, EGYPT

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SUMMARY

Two trials were conducted for three weeks (one during June and the other during July 2014) to evaluate the effect of protein level and hapa exchange times on growth performance and profitability of Nile tilapia (*Oreochromis niloticus*) fry during sex reversal stage in a commercial hatchery, Fayoum governorate, Egypt. The first one was conducted in six hapas (2 per each treatment) with size of 10.2 m³ (4 × 3 × 0.85 m). The hapas were stocked with Nile tilapia fry at a rate of 50,000 fry/hapa. Three diets varying in protein level [fish meal as animal protein source] were formed as follows: the D72 (100% fish meal (FM) = 72 % Crude Protein (CP)); the D53 (70% FM + 30% wheat flour = 53% CP) and D37 (33.33% FM + 33.33 % wheat flour + 33.33 % commercial diets = 37% CP). The diets were offered to the fry five times/day at a rate of 30% of total biomass weight/first week then decreased gradually by 10% every week. The second trial was to study the effect of hapa exchange times [zero time (0TT); one time (1TT) and two times (2TT) treatments, where, every treatment had two replicates] on the fry growth performance. The fry were fed with D37 (37% CP) which was mentioned previously at the first trial at the same feeding rate and times/day. The hapas of both first and second trials were fixed in an earthen pond (3000 m²).

The results obtained from the first trial revealed that significant differences (P<0.05) were found for all growth performance parameters. The D72 has the best values for final weight, weight gain, daily gain, specific growth rate. On the other hand, survival rate and feed conversion ratio have an opposite trend for that treatment (D72) when it was compared with the other treatments. But the profit index (PI) was significantly higher (P ≤ 0.05) for fish fed D37 followed by D53 and finally D72.

The results of the second trial demonstrated that there were significant differences (P<0.05) for all growth parameters the best values was obtained with both 1TT and 2TT when were compared with 0TT.

**Keywords:** Nile tilapia (*O. niloticus*), fish meal, protein level, hapa exchanged growth performance, profit index.

INTRODUCTION

Tilapia is the second aquaculture species globally. Egypt occupies the second ranked in the production of tilapia after China (FAO, 2014), because of it’s easy to adapt in tropical and sub-tropical areas of the world (Shelton, 2002). Tilapia have numerous advantages as an aquaculture species (Teichert-Coddington et al., 1997) but the ability to reproduce in the production setting has resulted in various techniques being developed to control unwanted reproduction.

Net enclosures or hapas are used in tilapia hatcheries (Guerrero, 1997). Hapas has many attributes that make them an excellent hatchery system for tilapia, especially in developing countries (El-Sayed, 2006). These include easy construction, easy management, easy seed harvest and low cost. Hapas can also be suspended in fertilized earthen ponds, deep water bodies and concrete tanks supplied with clear water.

As the main and most expensive component of the diet, protein draws greater attention in nutrition requirement studies (Loum et al., 2013).
Fry feed generally contains higher level of protein because protein and energy requirements are higher in the early stages of life. Protein content of fry feed for tilapia farming has not been standardized yet although some farms use feed stuffs having 40% protein. So, it is essential to recommend the appropriate protein level of fry feed for economic production of healthy fry and maximize its lifespan as well (Loum et al., 2013). So, the amount of protein in the diet should be just enough for fish growth where the excess protein in fish diets may be wasteful and cause diets to be unnecessarily expensive (Ahmad, 2000). Reducing feeding costs could be a key factor for successful development of aquaculture.

Fry growth is improved with increasing protein levels. Larvae of tilapia tend toward carnivores, yet as they mature they become more herbivorous (Suresh, 2003). Thus, dietary formulations may require modification as fish grow and ingredient usage may shift from animal-based ingredients such as fish meal to plant-based feedstuffs. Most early life-history stage diets for tilapia contain higher concentrations of fish meal than grow out diets (Gonzales et al., 2007).

Klanian and Adame (2013) reported that water was changed with new water, to dilute the high levels of ammonia and nitrites. Water exchange and flow rate may affect the growth and physiological functions of tilapia. Little information’s are available on the subject. Continuous water exchange generally sustains the good quality of culture water while low or zero water exchange may reduce the quality of water lead to reduce growth and increased mortality (El-Sayed et al. 2005).

This work aimed to study the effects of protein level and hapa exchange times on growth performance, survival rate and profitability of Nile tilapia (O. niloticus) fry during sex reversal stage in commercial hatcheries.

MATERIALS AND METHODS

Two experiments were conducted at commercial hatchery in Shakshouk village, Fayoum governorate, Egypt for three weeks during summer season 2014 (the first during June and the second during July) to evaluate the effect of protein level and hapa exchange times on growth performance and profitability of Nile tilapia (O. niloticus) fry. After hatching and before consumption of yolk sac, the fry were collected, graded, selected and weighed to be prepare for the study.

The first experiment was conducted in six hapa (two hapas per each treatment) with size of 10.2 m$^3$ (4 × 3 × 0.85 m). They were stocked with Nile tilapia fry at a rate of 50,000 fry/hapa. The fry were fed with three diets [72%, 53% and 37% crude protein (CP)] as follows: the D$_{72}$ [100% fish meal (FM) = 72% CP]; the D$_{53}$ (70% FM + 30% wheat flour = 53% CP) and the D$_{37}$ (33.33% FM + 33.33 % wheat flour + 33.33 % commercial diets =37% CP). The diets were treating with 17 α methyl testosterone hormone at a rate of 100 mg/kg diet (Al-Hakim et al., 2012). The diets were applied to the fry at a rate of 30% of total biomass, where the daily quantities were divided to five portions/day. After the first week, the quantities were decreased gradually by 10% every week (three weeks experimental period).

In the second experiment the fry were stocked inside the hapas (with 10.2 m$^3$) in an earthen pond (0.75 feddan = 3000 m$^2$) with different exchange times (two replicates per each). Firstly, zero time treatment (0TT), where, 50,000 fry were stocked at hapa (10.2 m$^3$) from the first day till 21 days without any exchange. Secondly, one time treatment (1TT), where 50,000 fry were stocked at hapa (10.2 m$^3$) from the first day until 10 days, then the fry were transferred in a new hapa (10.2 m$^3$) till 21 days. And finally, two times treatment (2TT), where 50,000 fry were stocked at hapa (10.2 m$^3$) after 7 and 15 days to the end of the trail (21 days). Fry were fed with D$_{37}$ (37% CP) plus hormone incorporated feed five times/day. Weekly samplings of both water and fry were done to test the water quality parameters and to determine the growth performance characteristics and profitability. All treatments were started during the first 48th h, this due to the hatchery production of fry was not enough to start all treatment in the same time. A plan of the second trail is shown in Table (1).

Parameters measurements:

Some water quality parameters were recorded weekly during the study period for the two trails. Water temperature, pH, dissolved oxygen (DO), ammonia nitrogen (NH$_3$-N), and salinity were measured weekly by
centigrade thermometer, Orion digital pH meter model 201, oxygen meter, Cole Parmer model 5946, HACH test kit ammonia mid-range 0-3 mg/L model NI-8, and TDS apparatus, respectively.

Table (1). A plan of the second trail.

<table>
<thead>
<tr>
<th>Item</th>
<th>Sex reversal stage in days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>0TT</td>
<td></td>
</tr>
<tr>
<td>1TT</td>
<td></td>
</tr>
<tr>
<td>2TT</td>
<td></td>
</tr>
</tbody>
</table>

Note that:
- 0TT = zero time treatment (the hapa was not exchanged);
- 1TT = one time treatment (the hapa was exchanged only one time) and
- 2TT = two time treatment (the hapa was exchanged two times).

By the end of both trails, some growth parameters and survival rate were measured
- Weight gain = Final weight - Initial weight (Effiong et al., 2009),
- Daily gain = Weight gain, g /period in days (Effiong et al., 2009),
- Specific growth rate (SGR %) = 100 (ln Final weight-ln Initial weight)/period in days, where ln is the natural log (Effiong et al., 2009),
- Feed conversion ratio (FCR) = feed offered/weight gain (Effiong et al., 2009),
- Survival rate (SR) % = Final number of fish /Initial number of fish x 100 (Charo-Karisa et al., 2006) and
- Condition factor (CF) = 100 x W/L³ where W = Weight (g), L = Total length (cm) (Ng and Wang, 2011).

Economic analysis:
A simple economic analysis was used to assess the cost effectiveness of diets used in the feed trial. The cost of feed was calculated using market prices, taking into consideration the cost of feed and the transport fare with the assumption that all other operating costs remained constant (e.g. cost of constructing hapa, cost of broodstock and labor).

Indices for economic evaluation included:
(i) Incidence cost (IC), which was calculated as:
IC = Cost of feed/ No. of fry produced
IC is actually the cost of feed to produced 1000 fry (relative cost per unit), and the lower the value, the more profitable using that particular feed (Nwanna, 2003; Abu et al., 2010).
(ii) Profit index (PI), which was calculated as:
PI = value of fish produced/ Cost of feed

Statistical Analysis:
Data were computed and statistically analyzed using a one-way analysis of variance using SPSS version 16 (2007). Mean of treatments were compared by Duncan multiple range test when the differences were significant (Duncan 1955).
RESULTS AND DISCUSSION

Water quality parameters:

Some water quality parameters were recorded during the study. The dissolved oxygen (DO) was 5.7 ± 0.14 mg/l. The pH was 8.05±0.13. The temperature was 29±1.0 °C. The NH$_3$-N concentration was 0.4±0.17 mg/l and salinity 2.8±0.22 ‰. These values were within the safe and acceptable ranges for the spawning and growth of Nile tilapia fry and agreed with that reported by many authors (Magid and Babiker, 1975; Ross, 2000; El-Sayed, 2006 and El-Sherif & El-Feky, 2008).

First experiment: Diets’ protein levels:

As shown in Table (2), the D$_{72}$ has highest significant (P≤0.05) values for all growth performance parameters except survival rate and FCR, where it has the lowest one. This may be due to firstly to: higher protein content of D$_{72}$ and fish meal diets are also assumed to contain androgenic hormones which are beneficial for fish growth, and, secondly to: the androgenic steroids which promote releasing of growth hormone from pituitary somatotrophs fish (Higgs et al., 1976).

Table (2). Effects of diets’ protein levels on some parameters of growth performance of Nile tilapia (O. niloticus) fry reared in hapa (50,000 fry/10.2 m$^3$) for 21 days.

<table>
<thead>
<tr>
<th>Item</th>
<th>Initial weight, mg</th>
<th>Final weight, mg</th>
<th>Weight gain, mg</th>
<th>Daily gain, mg</th>
<th>SGR, %</th>
<th>Survival rate, %</th>
<th>FCR</th>
</tr>
</thead>
<tbody>
<tr>
<td>D$_{72}$</td>
<td>10.99</td>
<td>149.99$^a$</td>
<td>138.00$^a$</td>
<td>6.57$^a$</td>
<td>12.41$^a$</td>
<td>75.55$^b$</td>
<td>1.38$^b$</td>
</tr>
<tr>
<td>D$_{53}$</td>
<td>10.99</td>
<td>101.47$^b$</td>
<td>90.48$^b$</td>
<td>4.31$^b$</td>
<td>10.58$^b$</td>
<td>92.85$^a$</td>
<td>1.72$^a$</td>
</tr>
<tr>
<td>D$_{37}$</td>
<td>10.99</td>
<td>99.87$^b$</td>
<td>88.88$^b$</td>
<td>4.23$^b$</td>
<td>10.51$^b$</td>
<td>94.17$^a$</td>
<td>1.73$^a$</td>
</tr>
<tr>
<td>SED</td>
<td>0.01</td>
<td>3.24</td>
<td>3.24</td>
<td>0.15</td>
<td>0.12</td>
<td>2.16</td>
<td>0.04</td>
</tr>
</tbody>
</table>

Note that:
- Values in the same column with different superscripts are significantly different (P≤0.05).
- SED, standard error of a difference between 2 means= √ (2×Error MS/r)
- D$_{72}$ [100% fish meal (FM) = 72% CP]; D$_{53}$ (70% FM + 30% wheat flour = 53% CP) and the D$_{37}$ (33.33% FM + 33.33 % wheat flour + 33.33 % commercial diets =37% CP).

D$_{37}$ and D$_{53}$ have highest significant (P≤0.05) values for both survival rate and FCR. These may be due to a higher content of carbohydrates which lightweight and caused feed mash to float on the water surface. This provide a chance for fry to consume feed, on the other hand for that of D$_{72}$ , where the feed mash were sinking quickly, and so that, all fry were not capable of consuming feed mash at the same time and lead to more mortality.

The trend of the growth of Nile tilapia fry during trial period revealed that tilapia fed with fish meal consistently increases rapidly in terms of body weight followed by those treated with fish meal with flour (70: 30) and the fish meal with flour and artificial feed (equally).

Several dietary protein requirements of several tilapia species have been estimated to range between 20-56% (El-Sayed and Teshima, 1991). De Silva and Perera (1985) reported that the optimum dietary protein level for optimum growth of Nile tilapia fry was 30% crude protein. As reported by Ahmad et al. (2004), the growth performance of Nile tilapia fry was highest at 45% protein diets. Al-Hafedh (1999) found that better growth of tilapia was obtained at high dietary protein levels (40-46%) rather than 25-35%. In the present study, fish meal diets contain the highest protein level compared to the other diets which contain 53 and 37% crude protein.

El-Sayed and Teshima (1991) reported that the dietary protein requirements of several species of tilapia have been estimated to range from 20 to 56%. Therefore, protein content of the test diet used in this study was within the suitable range for tilapia culture.
Table (3). Effects of diets’ protein levels on profitability of Nile tilapia (O. niloticus) fry reared in hapa (50,000 fry/10.2 m³) for 21 days.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Feed offered, Kg</th>
<th>Price of hormone, L.E</th>
<th>Feed cost, L.E</th>
<th>Fry No sales</th>
<th>Values of fry sales, L.E</th>
<th>Profit index</th>
<th>Incidence cost</th>
</tr>
</thead>
<tbody>
<tr>
<td>D72</td>
<td>7250</td>
<td>26.1</td>
<td>131</td>
<td>38000b</td>
<td>950b</td>
<td>6.07b</td>
<td>3.45</td>
</tr>
<tr>
<td>D33</td>
<td>7250</td>
<td>26.1</td>
<td>98</td>
<td>46650a</td>
<td>1166a</td>
<td>9.41b</td>
<td>2.10</td>
</tr>
<tr>
<td>D37</td>
<td>7250</td>
<td>26.1</td>
<td>62</td>
<td>47150a</td>
<td>1179a</td>
<td>13.44a</td>
<td>1.31</td>
</tr>
<tr>
<td>SED</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>867</td>
<td>21.68</td>
<td>0.18</td>
<td>0.13</td>
</tr>
</tbody>
</table>

Note that:
- Values in the same column with different superscripts are significantly different (P≤0.05).
- SED, standard error of a difference between 2 means = √ (2 x Error MS/r)
- D72 [100% fish meal (FM) = 72% CP]; D33 [70% FM + 30% wheat flour = 53% CP] and the D37 (33.33% FM + 33.33 % wheat flour + 33.33 % commercial diets =37% CP).

As illustrated in Table (3), the cost per kilogram of feed was highest for diet contained 100% fish meal (D72) and lowest for D37. D37 has the highest Survival rate with estimated value of 1179 L.E, but D72 has the lowest fry fed with estimated value of 950 L.E. Total harvested fry with estimated values for diets was in the following order: diet D37 > D33 > D72. Among treatments, D72 recorded the highest significant (P ≤0.05) value of cost/1000 fry (4.13 ± 0.29 L.E) followed by D33 (2.66 ± 0.35) and, finally, D37 (1.87 ± 0.36). The profit index (PI) showed that the cost analysis revealed a lowest cost and highest profit index for the D37. Low cost value means low cost to produce 1000 sex reversed fry. Meanwhile, a high profit index indicates a high amount of profit for every cost incurred in feeding. These are important factors for fish farmer minding when feed ingredients were chosen to minimize cost and maximize profit (Teves et al., 2014).

Second experiment (hapa exchange rate):

Fry hapas in commercial hatcheries are not washed or cleaned for fear of increased mortality for that exchange hapa or replaced by anther was necessary. Table (4) demonstrated that 1TT and 2TT treatments lead to improve the growth performance and survival rate and consequently increase profit index. There were significant (P≤0.05) differences for all growth performance parameters and the higher values were obtained from 1TT and 2TT treatments than 0TT treatment. These may be due to cleaning mesh nets allowed to renew and pass the water through the nets and thereby increase the dissolved oxygen and decrease the ammonia concentration, which lead to increase growth rates. Moreover, the 0TT treatments provided a chance to accumulate and fill inside mesh nets of the hapa by the feces and feed mash wastes. This prevent to renew and pass the water through the nets, so, lack of oxygen inside the hapa caused weakens growth rate and increased mortality.

Table (4). Effect of frequency hapa exchanged on growth performance and profit index of Nile tilapia fry reared in hapa (50,000 fry/10.2 m³) for 21 days.

<table>
<thead>
<tr>
<th>Item</th>
<th>Initial weight</th>
<th>Final weight</th>
<th>Weight gain</th>
<th>SGR</th>
<th>Survival rate</th>
<th>FCR</th>
<th>Profit index</th>
</tr>
</thead>
<tbody>
<tr>
<td>0TT</td>
<td>10.99</td>
<td>89.53b</td>
<td>78.55b</td>
<td>9.99b</td>
<td>71.22b</td>
<td>2.75a</td>
<td>9.65</td>
</tr>
<tr>
<td>1TT</td>
<td>10.99</td>
<td>107.62a</td>
<td>96.63a</td>
<td>10.86a</td>
<td>83.96a</td>
<td>1.83b</td>
<td>10.84</td>
</tr>
<tr>
<td>2TT</td>
<td>10.99</td>
<td>103.45a</td>
<td>92.46a</td>
<td>10.68a</td>
<td>87.00a</td>
<td>1.84b</td>
<td>10.70</td>
</tr>
<tr>
<td>SED</td>
<td>0.07</td>
<td>5.97</td>
<td>5.91</td>
<td>0.26</td>
<td>2.62</td>
<td>0.063</td>
<td>1.33</td>
</tr>
</tbody>
</table>

Note that:
- Values in the same column with different superscripts are significantly different (P≤0.05).
- SED, standard error of a difference between 2 means = √ (2 x Error MS/r)
- 0TT = zero time treatment (the hapa was not changed);
- 1TT = one time treatment (the hapa was changed only one time) and
- 2TT = two time treatment (the hapa was changed two times).
CONCLUSION

The results obtained from the first trial revealed that the D72 has highest significant (P≤0.05) values for all growth performance parameters except survival rate and FCR when compared with the other treatments. But, the profit index (PI) was significantly higher (P ≤ 0.05) for D37 followed by D53. The D37 has lowest cost and the highest profit index in. The second trail demonstrated significant (P≤0.05) differences for all growth parameters and the 1TT recorded the best values of the studied growth parameters. Depending on the above discussion, exchanging hapa with intense care, at least for one time or more, is preferable to improve environment water quality parameters this will reflex on healthy fry with high growth performance parameters.

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تأثير مستوى البروتين ومعدل تغيير الهابات على مظاهر النمو والربحية لزريعة البلطى النيلى أثناء مرحلة التحول الجنسي في المفرخات التجارية بمحافظة الفيوم

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أجريت تجربتان لمدة ثلاثة أسابيع الأولى في شهر يونيه والثانية في شهر يوليو 2014 لدراسة تأثير مستوى بروتين العليقة ومعدل تغيير الهابات على مظاهر النمو والربحية لزريعة البلطى النيلى في المفرخات التجارية بمحافظة الفيوم، مصر. أجريت الأولى في ستة هابات (هابتان/معالجة) بحجم 10.2 م³ (4x3x0.85 م) حملت زريعة البلطى بمعدل 50000/هابة. كونت ثلاثة علائق تختلف في نسبة البروتين الأولى (100% مسحوق سمك = 72% بروتين)، الثانية (70% مسحوق سمك + 30% دقيق قمح = 53% بروتين) أما الثالثة (33.3% مسحوق سمك + 33.3% دقيق قمح + 33.3% علبة تجارية = 37% بروتين). قدمت العالقات إلى الزريعة بعد المعالجة الهرمونية بمعدل 100 ملجم 17 الفا ميل تسترون/كم خمس مرات/يوم بمعدل 30% من وزن الزريعة في الأسبوع الأول ثم تناقصت تدريجيا بمعدل 10% لكل اسبوع.

التجربة الثانية درست تأثير عدد مرات تغيير الهابات (بدون تغيير، تغيير مرة، تغيير مرتين) كل مععلة بمكررين على مظاهر النمو. ثبت الهابات في حوض ترابي (3000 م³) وغذت الزريعة بال العليقة الثالثة (37% بروتين) المستخدمة في التجربة الأولى بنفس معدل التغذية وعدد المرات في اليوم.

أظهرت النتائج المحصلة عليها من التجربة الأولى وجود اختلافات معنوية بين كل معالجة مظاهر النمو وكانت العلقة الأولى في الأحسن في الوزن النهائى والزيادة في الوزن والزيادة اليومية ومعدل النمو النسبى أما معدل الأعاشة والتحويل الغذائي اختلاف معنوي بين المعالجات. ناتج الربحة كان أعلى مع النمو مع المحلاة المعنوي على العلقة الثالثة ثمها العلقة الثانية وأخيرا الأولى. تحليل التكاليف أظهر انخفاض مدى التكاليف وارتفاع دليل الربحية مع العليقة الثالثة.

نتائج التجربة الثانية أظهرت وجود اختلافات معنوية بين كل مظاهر النمو وأحسن القم التي تم الحصول عليها كانت مع الهابات التي تغيرت مرة واحدة أو مرتين بالمقارنة بالهابات التي لم تتغير.

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