

Effect of Brood Stock Size Combination on Reproductive Performance of Nile Tilapia (*Oreochromis niloticus*)

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Abstract: Two hundred and four *Oreochromis niloticus* brood stock were used in the experiment. Males and females were classified into three groups according to their body weights as follows; large (400 – 650 g), medium (160 – 300 g) and small (80-120 g) which coded A, B and C; respectively. All possible nine combinations between the three weight categories were conducted. Testes and ovary weights were significantly affected ($P < 0.001$ & $P < 0.01$ respectively) with different broodstock size groups. The highest testes and ovary weights were recorded at large-sized group, while it decreased steadily from medium to small-sized groups; respectively. Absolute fecundity, relative fecundity, and egg weight were all significantly affected ($P < 0.01$) with female size. The highest absolute fecundity and egg weight were obtained in large-sized females while the highest relative fecundity was obtained in the small-sized females. Gonado-somatic index didn't affect significantly with the various sizes of the female. Total collected fry number and growth performance represented as fry weight, length, and condition factor at the age of 6 weeks were all significantly ($P < 0.001$) affected with broodstock size combinations. The highest fry numbers were obtained from the AB, AC, and BC combinations respectively, whilst the lowest were recorded by BA and CB combinations. The highest fry weight and length at the age of 6 weeks after hatching were obtained from combinations AA, BB, and CA respectively. Nonetheless, the AC combination showed the lowest body weight and length. The highest condition factor was recorded by AB combination while the lowest was for the AC's.

Keywords: Nile tilapia, Broodstock, Size combination, Reproduction, Fecundity, Egg weight, Fry performance.

INTRODUCTION

Tilapia has been developed globally to be the second most important cultured freshwater fish. Tilapia Currently, is farmed commercially in almost 100 countries worldwide (FAO, 2011). There is a growing consensus that tilapias can become the world's most important warm water cultured fish in tropical and subtropical countries (Fitzsimmons, 2010). Global tilapia production is expected to almost double from 4.3 million tons per year in 2010 to 7.3 million tons a year in 2030. With these estimates, tilapia will likely be one of the main contributors to the fastest growth in global aquaculture aside from carp and catfish (FAO, 2014).

The main advantages of tilapia are its general hardiness, ease of breeding, rapid growth rate, low cost of production, easily spawning in captivity, tolerable to poor water quality, ability to efficiently convert organic and domestic wastes into high quality protein, flesh quality, and good taste (El-Saidy and Gaber, 2005 and Borgeson *et al.*, 2006).

The reproductive performance of Nile tilapia includes many aspects like absolute fecundity, relative fecundity, gonads weight, egg weight, gonado-somatic index (GSI), and fry performance (Bhujel, 2000; Gómez-Márquez *et al.*, 2003; Mohammed *et al.*, 2014). As there are wide variations in the reproductive performance among species and individuals within the species (Kirpichnikov, 1981; Macaranas *et al.*, 1997), many studies had been conducted to investigate different factors involved in tilapia reproductive performance. Some studies reported different relationships between female weight and absolute fecundity (Shalloof and Salama, 2008), relative fecundity (Frag, 2003), ovary weight and Gonado somatic index (Mohammed *et al.*, 2014) and egg weight (Fath El-Bab *et al.*, 2011).

Furthermore, other studies reported a mentioned some effects of female size on fry production performance of Tilapia (Bhujel, 2000; El-Saidy and Gaber, 2005; Mohammed *et al.*, 2014).

The study aimed to investigate the effects of the size combination of the breeding stock on different reproductive performance aspects of Nile tilapia in order to determine the optimum breeding stock management methods that may improve hatcheries production on the commercial scale.

MATERIALS and METHODS

Fish management and experimental design:

The experiment was conducted from 1st of May until the 20th of July 2015 at a commercial tilapia hatchery that located at San El-Hagar about 60 km northeast of Ismailia, Egypt. Two hundred and four (138 females and 66 males) *O. niloticus* broodstock were used in the experiment. The brood fish were maintained, where each sex was separated for a 25-day period in 12 m² hapas installed in a 0.25 ha pond with a stocking density 4 fishes/m². After such preparing period males and females were classified into three groups according to their body weights to large (400 – 650 g), medium (160 – 300 g), small (80-120 g) and named A, B, and C; respectively (Table 1). All possible mating combinations between the three weights categories were conducted through nine mating groups, each consisting of 2 males and 6 females, all with two replicates. Mating was carried out in 2 m² hapas installed in a 0.2 ha pond. After 14 days, swim-fry were collected separately from each hapa and then all females were checked and any eggs or fry still incubated inside the mother's mouth were also collected and counted. Fry from each mating type were stocked in two different 2 m² hapas at a stocking density 200

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fry/m² for further 6 weeks rearing period. Brood fish were fed 30% crude protein at the rate of 3% of their total body weight twice a day while Fry were fed *ad libitum* four times a day with a commercial diet containing 40% crude protein. All farm ponds filled with filtered canal water from Al-Salam canal. Water temperature ranged between 24 and 28°C, pH value ranged between 7.2-7.4 representing the alkaline medium needed for tilapia fish, and water salinity ranged between 2520 - 2700 mg/L.

Table (1): Brood-stock mating combinations in the order (♂ X ♀).

Code Weight	A (♂) (400-650 g.)	B (♂) (160-300 g.)	C (♂) (80-120 g.)
A (♀)	AA	AB	AC
B (♀)	BA	BB	BC
C (♀)	CA	CB	CC

Studied traits

A total of 60 brood-stock were used to investigate total body weight (g), total length (cm), ovary weight (g), testes weight (g), absolute fecundity, relative fecundity, egg weight (mg), and Gonado-somatic index. Absolute fecundity (AF) was determined according to Hunter *et al.* (1992) as follow: $F = (W / w) * X$; where F is the absolute fecundity, W: the weight of gonad, w: the mean weight of sub-samples and X: the counted number of mature eggs in the sub-sample. Relative fecundity (RF) was determined as the total number of eggs per unit weight of fish (Hunter *et al.*, 1992), while gonado-somatic index was calculated according to the formula: $GSI = [\text{weight of ovary (g)} / \text{female body weight (g)}] * 100$ (de Vlaming *et al.*, 1982).

In order to compare the fry production performance between all possible combinations, the

total collected fry numbers from each hapa was recorded. At 6 weeks old after hatching, thirty fry from each hapa were individually weighed (g) and their total length were measured (cm) and used to calculate the condition factor. Furthermore, survival rate was calculated for each hapa. Survival rate (%) was calculated as = Number of fish at the end of the experiment/number of fish at the start of the experiment*100, while condition factor was calculated according to the formula:

$$K = (\text{Weight (g)} / \text{Length (cm)}^3) * 100 \text{ (Le Cren, 1951).}$$

Statistical analysis

The data of brood-stock parameters were statistically analyzed using SPSS (2013) according to the following model: $Y_{ijk} = \mu + S_i + e_{ij}$, where, μ is the overall mean, S_i is the fixed effect of female or male size ($i = 1 \dots 3$), and e_{ij} is random error. The data of fry performance were statistically analyzed using SPSS (2013) program according to the following model: $Y_{ij} = \mu + C_i + h(c)_{ij} + e_{ijk}$, where, μ is the overall mean, C_i is the fixed effect of i^{th} the size brood-stock combination ($i = 1 \dots 9$), $h(c)_{ij}$ is the random experimental error of the j^{th} hapa within the i^{th} combination ($j=1,2$) and e_{ijk} is the random sampling error. Means were tested for significant differences using Bonferoni test (Bonferroni, 1936).

RESULTS AND DISCUSSION

Brood stock characterization

The results showed that males body weight, length, and girth was significantly ($P < 0.001$) affected with the different size groups. The highest male body weight, length, and girth were recorded in large-sized group while decreased for medium and small-sized respectively. Furthermore, the testes weight was also significantly ($P < 0.001$) affected with the different size groups. The large-sized group has superior testes weight while it was decreased for medium and small-sized groups respectively (Table 2).

Table (2): Least squares means for some characteristics of *O. niloticus* males classified into three different weights.

	Body weight (g)	Body length (cm)	Body girth (cm)	Testes weight (g)
Large (A)	404 ^a ±11.9	28.01 ^a ±0.5	20.8 ^a ±0.9	2.89 ^a ±0.29
Medium (B)	195 ^b ±11.9	22.6 ^b ±0.5	16.1 ^b ±0.9	1.64 ^b ±0.29
Small (C)	95 ^c ±11.9	18.1 ^c ±0.5	14.2 ^b ±0.9	1.01 ^b ±0.29
Significance	***	***	***	***

Means followed by different letter on the same column are different. *** Significantly different at ($P < 0.001$).

The results showed that females body weight, length, and girths were all significantly ($P < 0.001$) affected with size groups. The females body weight, length, and girths were highest at the large-sized group while decreased for medium- and small-sized respectively. The ovary and egg weights were significantly ($P < 0.01$) affected with female weight. The highest ovary and egg weights were recorded at large-sized females while it was decreased for medium and small-sized respectively (Table 3). The results showed that absolute and relative fecundity were

significantly ($P < 0.01$) affected with female size. The highest of absolute fecundity was obtained at large-sized group while, there values were lowered for medium and small-sized respectively. On the other hand, the highest of relative fecundity was obtained in smallest females size while, there values were lowered for medium and large-sized group respectively. The results showed that the gonado-somatic index didn't affect significantly according to different female weights.

Table (3): Least squares means for some characteristics of *O. niloticus* females classified into three different sizes.

	Body weight (g)	Body length (cm)	Body girth (cm)	ovary weight (g)	Egg weight (mg)	Absolute fecundity Egg	Relative fecundity Egg / g	GSI
Large (A)	528.3 ^a ±18.2	30.6 ^a ±0.44	22.6 ^a ±0.61	16.7 ^a ±1.6	7.17. ^a ±0.72	2058.8 ^a ±263.9	3.91 ^b ±1.02	3.17 ±0.6
Medium (B)	206 ^b ±18.2	23.26 ^b ±0.44	16.2 ^b ±0.61	7.7 ^b ±1.6	5.17 ^{ab} ±0.72	1059.3 ^b ±263.9	5.28 ^{ab} ±1.02	3.57 ±0.6
Small (c)	97 ^c ±18.2	18.3 ^c ±0.44	12.4 ^c ±0.61	3.05 ^b ±1.6	3.76 ^b ±0.72	711.3 ^b ±263.9	7.29 ^a ±1.02	3.08 ±0.6
Significance	***	***	***	**	**	**	**	ns

Means followed by different letters on the same column are significantly different. ** Significant differences at $P < 0.01$. *** Significant differences at $P < 0.001$. ns no significant differences.

Regarding to ovary and testes weights which were significantly affected with brood-stock body weight the results of the present study was in agreement with (Oso *et al.*, 2013) who mentioned a positive and significant relationship between fish weight and standard length, fish weight and weight of gonad in *Tilapia zilli* ($P < 0.05$). Furthermore, similar result in Nile tilapia was obtained with (Mohamed *et al.*, 2014) who reported that ovary weight was significantly ($P < 0.001$) affected with the female size. As for egg weight, the obtained significant effect of female size on egg weight was previously reported on some fishes as larger females often produce larger eggs (Martensdotir and begg, 2002; Rideout *et al.*, 2005). This phenomenon also reported in tilapia by Peters and Pauly (1983) who concluded that the weight of single eggs in tilapia varies widely and egg weight increases with body weight. As for fecundity, the present results revealed that total fecundity increased with increasing body size of females; which was in agreement with Hashem and El Agamy (1977) who stated that fecundity is a function related to length, weight and age of different fish species and increased with the increase in these parameters. Other studies concluded the same relationship between female size and absolute fecundity in tilapia (Watanabe and Kuo, 1985; Bhujel, 2007; Bombata and Megbowon, 2012; Mohamed *et al.*, 2014). As for relative fecundity the results showed that the highest relative fecundity was recorded in the smallest females and was lower in medium and largest ones. The present results were in agreement with Farag, (2003) who found that the highest relative fecundity increased with the decrease in female body weight. On the other hand other studies revealed that, relative fecundity was significantly ($P < 0.001$) affected with female size but increased at large-sized while decreased for medium and small-sized respectively (Mohamed *et al.*, 2013; Mohamed *et al.*, 2014).

Fry production performance

The results showed that the total collected fry number from each hapa was significantly $P < 0.001$

affected with the size brood-stock combinations. Furthermore, fry weight, length, and condition factor at 6 weeks old were all significantly $P < 0.001$ affected with the size broodstock combinations. On the other hand, survival rate wasn't significantly affected by broodstock size combinations (Table 4).

The data (Table 4) showed that AB, AC, and BC broodstock combinations were the highest concerning fry number collected from each hapa as 2960, 2430, and 2385 fry/hapa respectively while CB and BA combination showed the lowest fry number as 585 and 605 fry/ hapa. In tilapia agonistic interactions follows a hierarchical structure that forms a linear relationship based on size (Oliveira and Almada, 1996). Alpha males are the largest in the group and showed the greatest display of aggression. In addition, before reproduction in tilapia there are primary courtship displays and preparation such as digging the nest and defense displays (Oliveira and Almada, 1998). The present results suggest that using males which are larger than females in mating groups perhaps improve the courtship between males and females. The lowest fry number which obtained from combination BA and CB with a lower female size than male may also confirm the same explanation. Concerning fry body weight and length the results showed that the highest fry weight and length obtained from combinations AA, BB, and CA (1.71 g & 4.45 cm); (1.49 g & 4.32 cm); and (1.35 g & 4.20cm) respectively while BC and AC combination showed the lowest fry weight (1.27 g, 4.08 cm) and (1.2 g & 4.04 cm) respectively. Rana (1986) reported that Nile tilapia females of larger size were found to produce more and bigger eggs which were in agreement with the present results. Furthermore, El-Sayed and Gaber (2005) found that large eggs contained more yolk and led to larger fry with better growth. The previous results gave an explanation to the present results which indicate a superior weight and length of fry which resulting from combinations AA, BB, and CA and lowest weight and length of those which resulting from BC and AC combinations.

Table (4): Fry production performance of *O. niloticus* as affected with different the size brood-stock combinations in the order of (♂ X ♀).

	Fry no.	Fry weight (g)	Fry length (cm)	Fry C.F.	Survival rate %
AA	1810 ^c ±73.9	1.71 ^a ±0.05	4.45 ^a ±0.055	1.80 ^c ±0.042	93.4
BA	605 ^c ±73.9	1.25 ^d ±0.05	4.07 ^d ±0.055	1.83 ^c ±0.042	91.5
CA	1560 ^d ±73.9	1.35 ^c ±0.05	4.20 ^c ±0.055	1.82 ^c ±0.042	91.6
AB	2960 ^a ±73.9	1.32 ^d ±0.05	4.05 ^d ±0.055	1.99 ^a ±0.042	90.0
BB	1755 ^c ±73.9	1.49 ^b ±0.05	4.32 ^b ±0.055	1.88 ^b ±0.042	92.0
CB	585 ^c ±73.9	1.31 ^d ±0.05	4.14 ^d ±0.055	1.83 ^c ±0.042	93.1
AC	2430 ^b ±73.9	1.20 ^e ±0.05	4.04 ^d ±0.055	1.72 ^d ±0.042	93.5
BC	2385 ^b ±73.9	1.27 ^d ±0.05	4.08 ^d ±0.055	1.82 ^c ±0.042	93.2
CC	1430 ^d ±73.9	1.29 ^d ±0.05	4.04 ^d ±0.055	1.84 ^c ±0.042	95.1
Significance	***	***	***	***	ns

Means followed by different letters on the same column are significantly different. *** Significant differences at P < 0.001. ns no significant differences.

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"تأثير توافيق أحجام قطيع التفريخ على الأداء التناسلي لأسماك البلطي النيلي"

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استخدم في الدراسة ٢٠٤ سمكة من قطيع امهات أسماك البلطي النيلي حيث تم تقسيم الذكور والإناث إلى ثلاثة مجموعات حسب الوزن كالتالي: احجام كبيرة ٤٠٠ : ٦٥٠ جرام و احجام متوسطة ١٦٠ : ٣٠٠ جرام و احجام صغيرة ٨٠ : ١٢٠ جرام ، وقد أشير إليهم بـ "أ" ، ب ، ج" على الترتيب. تم عمل كافة توافيق التزاوجات الممكنة بين الثلاث مجموعات الوزنية بإجمالي تسعة تزاوجات ممكنة. وقد أظهرت النتائج أن وزن المناسل في الذكور والإناث تأثرت معنويًا ($P < 0.001$) و ($P < 0.01$) على الترتيب" بتقسيم المجموعات الوزنية حيث أن أعلى أوزان للمناسل سواء الخصيتين أو المبيض كانت في أحجام الذكور والإناث الكبيرة بينما انخفضت للأوزان المتوسطة والصغيرة على التوالي. أيضاً أشارت الدراسة أن قيم الخصوبة الكلية والخصوبة النسبية و وزن البيضة قد تأثروا جميعاً معنويًا ($P < 0.01$) بوزن الإناث وقد سُجلت أعلى قيم للخصوبة الكلية و وزن البيضة في أحجام الإناث الأكبر في حين كانت أعلى قيمة للخصوبة النسبية في أحجام الإناث الأصغر ثم المتوسطة والكبيرة على التوالي. أيضاً أوضحت نتائج مؤشر النضج الجنسي أنها غير متأثرة معنويًا بالأحجام المختلفة للإناث. كما أظهرت النتائج أن عدد الزريعة المنتجة وأداء النمو المتمثل في "وزن وطول الزريعة ومُعامل الحالة" على عمر ٦ أسابيع قد تأثروا جميعاً معنويًا ($P < 0.001$) باختلاف توافيق الأحجام بين قطيع التفريخ. وسُجلت أعلى قيم لعدد الزريعة في التزاوجات "أب" و "أج" و "بج" على التوالي، بينما كانت أقل قيم في التزاوجات "ب أ" و "ج أ" و "ج ب". وكانت أعلى قيم لوزن وطول الزريعة على عمر ٦ أسابيع بعد الفقس في التزاوجات "أ أ" و "ب ب" و "ج ج" على التوالي، وسُجل التزاوج "أج" أقل قيم لوزن وطول الزريعة. وقد سُجلت أعلى قيمة لمُعامل الحالة في التزاوج "أب" وأقل قيمة في "أج".