

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

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Abstract: Two hundred and four *Oreochromis niloticus* broodstock 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 (GSI) didn't affected significantly with the various females' size. Total collected fry number and fry 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 combination. The highest collected fry numbers were obtained from AB, AC, and BC broodstock size combinations, respectively, whilst the lowest were recorded by BA and CB combinations. The highest fry weight and length were obtained from combinations AA, BB, and CA, respectively. On the hand, AC showed the lowest body weight and length. The present results advanced the thought that forming mating groups with males which are larger than females was better in fry production activities.

Keywords: 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 (Farag, 2003); ovary weight; GSI (Mohammed *et al.*, 2014) and egg weight (Fath El-Bab *et al.*, 2011). Furthermore, other studies

reported some effects of female size on fry production performance of Tilapia (Bhujel, 2000; El-Saidy and Gaber, 2005 and 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 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-days period in 12 m² hapas installed in a 0.25 ha earthen pond with a stocking density of four fishes/ m². After such preparing period males and females were classified into three groups according to their body weight 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 weight categories were conducted through nine mating groups, each consisting of two males and six females, all with two replicates. Mating was carried out in 2 m² hapas installed in a 0.2 ha pond. After 14 days, swim-up 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 randomly selected and stocked in two different 2 m² hapas at a stocking density

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200 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 7.2-7.7 representing the alkaline medium needed for tilapia fish, and water salinity ranged between 2500 - 2700 mg/L.

Table (1): Broodstock 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 broodstock 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 (GSI). Absolute fecundity (AF) was determined according to El-Sayed, (1996) 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

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 ($P < 0.001$).

Table 3 shows that females body weight, length, and girth were all significantly ($P < 0.001$) affected with size groups. Females body weight, length, and girth 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. Absolute and relative fecundity were also significantly ($P < 0.01$) affected with female size. The highest absolute fecundity was obtained at large-sized group while, there values were lowered for medium and

recorded. At 6 weeks old after hatching, 30 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: $K = (\text{Weight (g)} / \text{Length (cm)}^3) * 100$ (Le Cren, 1951).

Statistical analysis

The data of broodstock characterization were statistically analyzed using SPSS (2013) according to the following model: $Y_{ij} = \mu + S_i + e_{ij}$, 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 production 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 broodstock size 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

Broodstock characterization

The results showed that males body weight, length, and girth were all significantly ($P < 0.001$) affected with the different size groups. The highest males 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).

On the other hand, the highest of relative fecundity was obtained in the 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 differ significantly according to different female weights.

Regarding to ovary and testes weights which were significantly affected with broodstock body weight the results of the present study were in agreement with Oso *et al.* (2013) and Mohamed *et al.*, (2014) who mentioned a positive and significant relationship between broodstock body weight and gonads weight in *Tilapia zilli* and Nile tilapia, respectively.

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.

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 (Marteinsdottir & Begg, 2002 and Rideout *et al.*, 2005). This phenomenon also reported in tilapia by Peters and Pauly (1983) who concluded that the weight of single egg in tilapia varies widely and egg weight increases with body weight increase. Concerning fecundity, the present results revealed that total fecundity increased with increasing body size of females. Hashem and El Agamy, (1977) stated that fecundity is a function related to length, weight and age of different fish species and increased with the increase in these parameters. The same relationship between female size and absolute fecundity in tilapia were reported by Bhujel, (2007) and Bombata & Megbowon, (2012). 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, another studies reveled that, relative fecundity was significantly affected with female size but increased at large size while decreased for medium and small sizes, respectively (Mohamed *et al.*, 2013; Mohamed *et al.*, 2014).

Fry production performance

The results (Table 4) showed that the total collected fry number from each hapa was significantly ($P < 0.001$) affected with the broodstock size combination. AB, AC, and BC broodstock size combinations were the highest concerning fry number collected from each hapa as 2960, 2430, and 2385 fry/hapa, respectively while CB and BA size combinations showed the lowest fry number as 585 and 605 fry/ hapa, respectively. The present results suggest that using males which are larger than females in mating groups could improve the fry production. 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. Gozlan *et al.* (2003) mentioned that females choose males for mating according to their aggressiveness. Beeching *et al.* (2004) reported that females can also choose males according to body size. In addition, before reproduction in tilapia there are primary courtship displays and preparation. Courtship behavior involves all the behavioral interaction between female and male which lead up before the fertilization. Goncalves-de-Freitas and Nishida, (1998) described courtship act in Nile tilapia as undulation and parallel movements around the female and near spawning unit bottom. Engtrom-ost and Candolin, (2007) found that females choose males according to their courtship performance. The current results led to suggest that using males larger than the females in the breeding groups was accompanied with desirable aggressive males and better courtship performance which resulted in higher fry production.

Fry weight, length, and condition factor at 6 weeks old were all significantly $P < 0.001$ affected with the broodstock size combination. On the other hand, survival rate% wasn't affected significantly by broodstock size combination. Concerning fry body weight and length the results showed that the highest fry weight and length obtained from combinations AA (1.71 g & 4.45 cm), BB (1.49 g & 4.32 cm), and CA (1.35 g & 4.20cm), respectively while BC and AC combinations showed the lowest fry weight and length (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. Furthermore, El-Saidy and Gaber (2005) found that large eggs contained more yolk and led to larger fry with better growth. Such previous results may gave an explanation to the present results which indicate a superior weight and length of fry which resulting from females size group A and B and lowest fry weight and length of those which resulting from females size group C.

Table (4): Fry production performance of *O. niloticus* as affected with different broodstock size 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 ^e ±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 ^e ±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 التجربة ٢٠٤ من قطيع تفريخ البلطي النيلي حيث تم تقسيم الذكور والإناث إلى ثلاثة مجموعات حسب الوزن كالتالي: أحجام كبيرة ٤٠٠ : ٦٥٠ جرام وأحجام متوسطة ١٦٠ : ٣٠٠ جرام وأحجام 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 تأثرا معنويا ($P < 0.001$) 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. أشارت الدراسة أن قيم الخصوبة الكلية والخصوبة النسبية و وزن البيض قد تأثروا جميعا معنويا ($P < 0.01$) بوزن الإناث وقد سُجلت أعلى قيم للخصوبة الكلية و وزن البيض في أحجام الإناث الأكبر في حين كانت أعلى قيمة للخصوبة النسبية في أحجام الإناث الأصغر ثم المتوسطة والكبيرة على التوالي. دليل وزن المناسل : وزن الجسم لم يتأثر معنويا بالأحجام المختلفة للإناث. عدد الزريعة المنتجة وأداء النمو المتمثل في وزن وطول الزريعة وعامل الحالة على عمر ٦ أسابيع تأثروا جميعا معنويا ($P < 0.001$) باختلاف توافيق الأحجام بين قطيع التفريخ. سُجلت أعلى قيم لأعداد الزريعة المنتجة في التزاوجات "أب" و "أج" و "بج" على التوالي، بينما كانت أقل قيم في التزاوجات "بأ" و "جأ" و "بب". أعلى قيم لوزن وطول الزريعة على عمر ٦ أسابيع بعد الفقس كانت في التزاوجات "أأ" و "بب" و "جأ" على التوالي، وسجل التزاوج "أج" أقل قيم لوزن وطول الزريعة. النتائج الحالية تطرح فكره ان استخدام ذكور أكبر من الإناث في مجموعات التزاوج أفضل في ممارسات انتاج الزريعة.