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# EFFECT OF DIFFERENT STOCKING DENSITIES AND SEX RATIO ON REPRODUCTIVE PERFORMANCE OF NILE TILAPIA (*Oreochromis niloticus*) IN CONCRETE PONDS

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***Abstract***

These experiments was conducted to investigate the effects of broodstock stocking density, sex ratio and there interaction on reproductive performance of Nile tilapia (*Oreochromis niloticus*) broodstock. Three trials was design, the first, two different broodstock density (2 fish/$m^3$ and 3 fish/$m^3$) at six ratio of 1♂:2♀, the second trail three different broodstock sex ratio (1♂:1♀, 1♂:2♀ and 1♂:3♀) at 2 fish/m stocking density and the third interaction between them. The experimental trials were assigned according the one way analysis of variance design with two replicates per treatment giving total number of 22 concrete ponds and contained for two months. The results of broodstock density at 2 fish/$m^3$ had the highest fry production /female. The results obtained with sex ratio 1♂: 1♀ was significantly higher than other sex ratio/female. The results of interaction between stocking density and sex ratio cleared that significant effect obtained and the highest fry production /female obtained with density 2 fish/$m^3$ with sex ratio 1♂:1♀ and the lowest fry production obtained with density 3 fish /$m^3$ and 1♂:3♀ sex ratio. But total fry production for pond was more efficient with sex ratio 1♂:3♀ and density 3 fish/$m^3$ was suitable for commercial hatcheries.

**Key words:** Nile tilapia, broodstock, sex ratio, stocking density, reproductive **and hatcheries.**

## INTRODUCTION

Tilapia is an ideal candidate for warm-water aquaculture. They spawn easily in captivity, use a wide variety of natural foods as well as formulated feeds, tolerate poor water quality, and grow rapidly at warm temperatures. These attributes, along with relatively low input costs, have made tilapia the

most widely cultured freshwater fish in tropical and subtropical countries (Biswas*et al.,* 2005 and Fasakin*et al.,* 2005).

Not only tilapia is one of the most widely cultured fish in the world but also it is one of the most important commercial fish in Egypt, whereas Egypt production of tilapia was 867557 ton and this amount represented about 58.5 % of Egyptian fish production in 2014 (GAFRD, 2014).

Reproductive success in many fish species influenced by, the broodstock, stocking density, sex ratio, age, size, nutrition and feeding regime (Hammouda *et al*., 2008 and Ibrahim *et al.*, 2008).

Stocking density can influence seed production in tilapia under culture conditions (Obi and Shelton, 1988). Many other workers indicated that lower stocking is better than the higher stocking; the lower stocking density had the highest seed/ female. (Siddiqui*et al.*1997) found that fecundity is positively related to the size of fish, the growth of fish decreased with increasing density of fish, it appears that social interaction and social stress were responsible for inefficient food utilization, poor growth and consequently low fecundity at high stocking density. Also Allison *et al.* (1979) reported an inverse relationship between stocking density and fry produced in *O. aureus.*

The sex ratio can also be influenced by the distribution of individuals in time and space, temperature and precopulatory guarding of multiple mates (Debuse*et al.,* 1999) and sex ratio of brood fish can have both economic and technical impacts, suboptimal ratios, in which either males or females are unable to spawn clearly waste resources and increase cost (Ridha and Cruz, 1998). Many of researches were conducted to determine the optimum of tilapia broodfish sex ratio such as Muntaziana*et al*. (2011) investigated the effect of broodfish sex ratio on seed production of red tilapia (*Oreochromis sp*.) using hapa nets suspended in an earthen pond, they reported that there were no significant difference ($p>0.05$) in seed production and female broodfish performance between the two sex ratios tested which were 1:1 and 1:3 (male:

female). On the other hand, Khater (2002) reported that sex ratio of 1:3 male to female was better economically for fry production.

This study aimed to determine the optimum stocking density and sex ratio of tilapia broodstock during the reproductive season, which leads to increase of fry production with economic efficiency from the same area

## MATERIALS AND METHODS

Three experiment were conducted in a greenhouses commercial fish hatchery at Central Laboratory for Aquaculture Research, Abbasa, Egypt to investigate the effects of broodstock stocking density, sex ratio and there interaction on reproductive performance of Nile tilapia (*Oreochromis niloticus*) broodstock. Three trials was design, the first, two different broodstock density (2 fish/$m^3$ and 3 fish/$m^3$) at six ratio of 1♂:2♀, the second trail three different broodstock sex ratio (1♂:1♀, 1♂:2♀ and 1♂:3♀) at 2 fish/ $m^3$ stocking density and the third interaction between them. An average body weight of broodstock was 160±8 and 150±5 gm for female and male respectively.

The experimental trials were assigned according the one way analysis of variance design with two replicates per treatment giving total number of 22 concrete ponds and contained for two months. Water capacity of each pond was 20 $m^3$ and feeding rate was 1% of fish body weight and feeding was twice daily. This study continued for two months. Broodstock were fed on diet containing 34.8 % crude protein (Table, 1).

**Table 1.** Composition of the experimental diet for Nile tilapia (*Oreochromis niloticus*) broodstock.

| Ingredients | % |
|---|---|
| Fish meal | 30 |
| Rice bran | 21 |
| Wheat bran | 16 |
| Yellow corn | 14 |
| Molasses | 2.75 |
| Dicalcuim phosphate | 1 |
| Vit. & Min. premix | 0.25 |
| Total | 100 |
| Proximate analysis | |
| Protein | 34.85 |
| Lipids | 6.20 |
| Ash | 7.7 |
| Fiber | 6.13 |
| NFE | 45.12 |

Chemical analysis was determined according to (AOAC, 2000) and NFE was calculated by difference.

**Water quality:**

Water temperature, pH, dissolved oxygen (DO), salinity and ammonia throughout experimental periods were measured periodically in the morning and at noon by centigrade thermometer, Orion digital pH meter model 201, oxygen meter (Cole Parmer model 5946) and Hanna instruments test kits (HI4829) for ammonia respectively.

**Statistical analysis:**

The analysis of variance and LSD of Duncan Waller were used to compare treatment means. Data were analyzed using stat graphic package software **(SPSS, 2007)** SPSS Inc. Released 2007. SPSS for Windows, Version 16.0. Level of significant was 0.05.

## RESULTS AND DISSCUSION

### Water quality:

Water quality parameters were showed in table (2) The averages of water temperature, water salinity, dissolved oxygen, water pH, and ammonia-N concentration, were 29.20°C, 0.58‰, 6.45, 8.01 and 0.40 respectively. These values were within the acceptable limits for tilapia Brood stock as reported by Popma and Lovshin (1996) and Popma and Masser (1999).

**Table 2.** Average water quality parameters during the experimental period.

| *Water quality parameters* | |
|---|---|
| **Temperature** | 29.20°C |
| **pH** | 8.01 |
| **Dissolved oxygen, mg/l** | 6.45 |
| **Salinity, ppt** | 0.58 |
| **Total ammonia, mg/l** | 0.40 |

### First trial: Effect of stocking density on Nile tilapia reproductive performance:

As shown in Table (3) fry production were significant differences between the treatments, whereas the lower density (2 broodfish /m³ ) had the highest of fry production per female in average (477 fry).

**Table 3**. Effect of stocking density on Nile tilapia fry production (± SE)

| **Item** | **Stocking density/m$^3$** | |
|---|---|---|
| | **2** | **3** |
| **Fry production/ female** | | |
| **Harvest 1** | 501.00± 17.08 | 465.83±8.69 |
| **Harvest 2** | 485.50±11.51 | 457.33±9.22 |
| **Harvest 3** | 484.83$^a$±10.08 | 451.83$^b$±9.64 |
| **Harvest 4** | 472.17$^a$±8.41 | 436.67$^b$±10.84 |
| **Harvest 5** | 467.17$^a$±9.07 | 428.67$^b$±11.34 |
| **Harvest 6** | 448.83±7.10 | 421.67±18.22 |
| **Average** | 477$^a$±9.54 | 444$^b$±9.21 |

In the present study the fry production was the highest at the lower density may be return to increase of broodfish density lead to competition for space and feed, this causes social stress and affecting reproductive efficiency. Also an increasing of broodfish density due to increase competition between territorial male or fighting between males leading to reduction in courtship, egg fertilization and incubation .Moreover, Guerrero (1982) reported that, tilapia mucus contains a substance that may cause auto allergic responses and inhibit reproduction at high broodstock densities. This result was in agreement with these of Tahoun*et al.* (2008) who reported that, increasing broodstock density significantly ($P \leq 0.05$) reduced broodstock fecundity. Similar observations for Nile tilapia reported by Little (1989) the reduced fry production at a higher stocking density indicates a lower fertilization success of eggs and those females were less successful in incubating the fry and this was attributed to the increased interference during spawning and the stress during incubation.

In the same trend, Allison *et al.* (1979) reported an inverse relationship between stocking density and fry produced in *O. aureus*. Also, high broodfish density often reduces seed production (Bevis, 1994).

Bhujel (2000) reported that, an inverse relationship between stocking density and the percentage of spawning females has been found in production hybrids of tilapia, *O. niloticus* and *O. hornorum*, probably due to some chemical or behavioral factors. High stocking density inhibits reproduction possibly due to the presence of a substance in tilapia mucus, which might cause an auto allergic response.

On the other hand, many of researchers had suggests in this trend such as Tsadik and Bar (2007) suggested lower stocking density of 3 broodfish/ $m^3$ to improve seed production of hapa -in- pond tilapia hatcheries. Behrends and Smitherman (1983) suggested that 5 fish/$m^2$ be adopted for optimum seed production of Nile tilapia in suspended net enclosures. Little (1992) found that 6 fish/m2 was optimum for Nile tilapia reared in hapas suspended in fertilized

ponds, and Ridha and Cruz (1999) found that 4 fish/$m^2$ had better seed production and spawning synchrony than 8 and 12 fish/$m^{2.}$

**Second trial: Effect of sex ratio on Nile tilapia reproductive performance.**

As shown in Table (4) the sex ratio significantly affected on fry production per female. Whereas there were significant differences between the treatments. The sex ratio (1 male : 1 female) had the highest fry production per female (486 fry /female) followed by (1male : 2 female) but the sex ratio (1male : 3female) was the worst in fry production per female.

**Table 4.** Effect of sex ratio on Nile tilapia fry production (± SE).

| Item | Sex ratio (♂:♀) | | |
|---|---|---|---|
| | **1:1** | **1:2** | **1:3** |
| **Fry production** | | | |
| **Harvest 1** | $519.75^{a}$±18.46 | $477.25^{ab}$±8.99 | $453.25^{b}$±7.05 |
| **Harvest 2** | $497.5^{a}$±10.85 | $473.25^{ab}$±8.22 | $443.5^{b}$±8.34 |
| **Harvest 3** | $493.5^{a}$±10.72 | $469.75^{ab}$±9.18 | $441.75^{b=}$±10.58 |
| **Harvest 4** | $480.5^{a}$±9.35 | $454.25^{ab}$±8.81 | $428.5^{b}$±13.02 |
| **Harvest 5** | $476.5^{a}$±10.12 | $446^{ab}$±9.41 | $421.25^{b}$±14.00 |
| **Harvest 6** | 450.5±10.49 | 451±18.06 | 404.25±14.95 |
| **Average** | $486^{a}$±10.43 | $462^{b}$±12.52 | $432^{c}$± 11.96 |

This result agreed with Hango and Brummett (1998) found that, fry production was significantly higher in 1:1 male to female compared to male: 3female of *O. shiranus* this may be due to decrease of competition between the female on the male , whereas number of males equal number of the female compared with the second and third treatments. And maybe increase in fertilization of egg with increase of male number. Also, available males with the sex ratio male per 2 or 3 females are not sufficient to fertilize eggs. And this result was partial in agreement with; Salama (1996) found that, the sex ratio male to 2 females achieved the highest of Nile tilapia fry compared with male per 3, 4, and 5 females. As well as Nouret *al.* (2008) evaluated the spawning results of three broad tilapia species, (*Oreochromis niloticus, O. aureus* and *O. galilaeus*) with two sex ratios (1:2 or 1:3; male: female) on the criterias of fry

production. The results indicated that *O. niloticus* broodstock with (1♂:2♀) sex ratio produced the highest number of fry. And Hughes and Behrends (1983) found that, in Nile tilapia, a male: female ratio of 1: 2 produced more seed than a 1: 3 ratio.

But this opining differed with, higher male density led to increase aggression and male competition which could reduce the opportunity for female to spawn cited by (Muntaziana *et al*., 2011). Also the higher spawning frequency occurred when fewer males were encountered during spawning activity (Mills and Reynolds, 2003).Under the same treatment this result differed with Khalfalla *et al.* (2008)who found that, the highest fry production of blue tilapia (*Oreochromis aureus*) obtained with the sex ratio (1♂:2♀) from three different broodstock sex ratios (1:1, 1:2 and 1:3 male: female).

On the other hand, Khater (2002) did not find any significant differences with different sex ratio of male to female, male to 2females and male to 3 females in number of Nile tilapia fry per g female.

**Third trial: The effect of interaction between stocking density and sex ratio on Nile tilapia fry production.**

The effect of interaction between stocking density and sex ratio on Nile tilapia fry production per female was shown in table (5) there were significant differences between the treatments at level (0.05). The low density (2 broodfish /$m^3$) was better than (3broodfish /$m^3$) also the sex ratio one male to one female was the best in fry production than one male to two females or one male to three females.

Table (5) cleared that, the sex ratio (1:1) with the lowest density (2 broodfish/ m3) was the best in all treatments followed by (1:2) and (1:3) with the lowest density, while the worst fry production per female was achieved with the sex ratio (1:3) with 3broodfish per $m^3$. This result agreed with Wafeek and Ali (2013) reported that, the lowest broodstock density resulted in the highest egg number, fertilization rate and hatching rate per female. Moreover, Bhujel

(2000) reported that, an inverse relationship between stocking density and the percentage of spawning females has been found in production hybrids of tilapia, *O. niloticus* and *O. hornorum*, probably due to some chemical or behavioral factors. High stocking density inhibits reproduction possibly due to the presence of a substance in tilapia mucus, which might cause an auto allergic response.

**Table 5.** The effect of interaction between stocking density and sex ratio on Nile tilapia fry production (± SE).

| **Interaction between stocking density and sex ratio** | | | | | | |
|---|---|---|---|---|---|---|
| **Stocking density/$m^3$** | **2 brood/$m^3$** | | | **3 brood/$m^3$** | | |
| **Sex ratio♂:♀** | **1:1** | **1:2** | **1:3** | **1:1** | **1:2** | **1:3** |
| **Fry production/ female** | | | | | | |
| **Harvest 1** | 551±9.0 | 487±17.0 | 465±4.0 | 488.5±3.50 | 467.5±2.50 | 441.5±2.50 |
| **Harvest 2** | 515.5±7.50 | 484±13.0 | 457±7.0 | 479.5±1.50 | 462.5±2.50 | 430±2.0 |
| **Harvest 3** | 512±2.0 | 482.5±12.50 | 460±2.0 | 475±1.0 | 457±5.0 | 423.5±1.50 |
| **Harvest 4** | 496±6.0 | 469.5±0.50 | 451±1.0 | 465±3.0 | 439±1.0 | 406±2.0 |
| **Harvest 5** | 494±1.0 | 462±4.0 | 445.5±0.5 | 459±1.0 | 430±2.0 | 397±1.0 |
| **Harvest 6** | 468.5±2.50 | 448±2.0 | 430±2.0 | 432.5±2.50 | 454±44 | 378.53.5 |
| **Average** | $506^a$ | $472^b$ | $451^b$ | $467^b$ | $452^b$ | $413^c$ |

Also, Khater and Ali (2008) found that, fry production per female was negatively affected with an increase of number broodfish above 3broodfish per $m^2$. And these results partial agreed with Siddqui *et al.* (1997) stated that, hybrid tilapia brood fish should be stocked at a density of 2 fish/$m^2$ with a sex ratio of male to 2 female or 3 female to maximize seed production in concrete tanks.

In the same trend, this result also agreed with several researchers studied the effects of different stocking densities on the reproductive performance particularly, on Nile tilapia *O. niloticus* have demonstrated that increasing the level of stocking density significantly ($P \leq 0.05$) reduces spawning success and in turn mass production (Ridha and Cruz, 1999 and Bhujel, 2000). However, Obi and Shelton (1988) who found that fry production per unit of

area (m2) in tilapia, *O. hornorum* (Trewavas) tended to increase with the increase of broodstock stocking density.

In relation to effect of sex ratio it can be said that, an increase of females' number increases fry productions per pond but take place reduce of fertilization eggs. Whereas, the sex ratio (1:1) was better than (1:2) and (1:3) and this agreed with Broussard *et al*. (1983) found that increasing broodstock density, at a fixed male: female sex ratio of 1: 3, had a negative effect on fry production of Nile tilapia reared in ponds. The authors attributed that effect to increasing competition between territorial males and/or constraints imposed by feed availability. On the other hand, Muntaziana *et al.* (2011) reported that, seed production of red tilapia in hapas was not affected by the ratio of males to females.

**Fry production evaluation:**

This evaluation was conducted on basis the fry production per pond and fry production per pond were shown in Table (6).

**Table 6.** Fry production per pond under efficiency of stocking density, Sex ratio and interaction between stocking density and sex ratio.

| Treatments | Fry production/ pond |
|---|---|
| **Stocking density** | |
| **Stocking density (2broodfish/$m^2$)** | $12243^{b}$ |
| **Stocking density (3broodfish/$m^2$)** | $17020^{a}$ |
| **SED*** | 3.11 |
| **Sex ratio** | |
| **1 male : female** | $12150^{c}$ |
| **1 male : 2 female** | $15477^{b}$ |
| **1 male : 3 female** | $16200^{a}$ |
| **SED*** | 403.3 |
| **Interaction between stocking density and sex ratio** | |
| **1 male : female with (2broodfish/$m^2$)** | $10120^{f}$ |
| **1 male :2 female with (2broodfish/$m^2$)** | $12744^{e}$ |
| **1 male : 3female with (2broodfish/$m^2$)** | $13530^{d}$ |
| **1 male : female with (2broodfish/$m^2$)** | $14010^{c}$ |
| **1 male :2 female with (2broodfish/$m^2$)** | $18080^{b}$ |
| **1 male : 3female with (2broodfish/$m^2$)** | $18580^{a}$ |
| **SED*** | 9.8 |

*, Standard error of differences.

From Table (6) it can be concluded that, the high stocking density (3broodfish /$m^3$) had the highest fry production per pond (17020 fry) compared with the other density. Also an increase of female number lead to increase of fry production per pond this achieved with the sex ratio male to 3 female (16200fry). The interaction between stocking density and sex ratio cleared also that, high stocking density with the sex ratio had the highest fry production per pond. These results were in agreement with **Salama (1996)** reported that, low production of tilapia seed could be attributed to very low density of broodfish and inappropriate sex ratio. Likewise, Khater (2002) reported that, the sex ratio male: 3 female is more economical for fry production.

## CONCLUSION

The results of broodstock density, the lower density had the highest seed production/female. The results obtained with sex ratio 1♀:1♂ was significantly higher than other sex ratio/female. The results of interaction between stocking density and sex ratio cleared that significant effect obtained but the highest seed production obtained with density 2 fish/$m^3$with sex ratio 1:1 and the lowest obtained with density 3 fish /$m^3$with 3:1 sex ratio/female. But total fry production for pond was more efficient with sex ratio 1♂:3♀ and density 3 fish/$m^3$ was suitable for commercial hatcheries.

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# تأثير الكثافات والنسب الجنسية المختلفة على الكفاءة التناسلية لأمهات اسماك البلطى النيلى فى الاحواض الاسمنتية

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## الملخص العربى

أجريت هذه التجارب لدراسة تأثير الكثافات والنسب الجنسية المختلفة لأمهات أسماك البلطي النيلي والتداخل بين هذه الكثافات والنسب الجنسية على الأداء التناسلي لأمهات اسماك البلطي النيلي فى الاحواض الاسمنتية.

أجريت ثلاث تجارب فى احواض اسمنتية عددها 22 حوض لمدة شهرين :

- التجربة الأولى : استخدم فيها كثافتين مختلفتين وهى 2 سمكه / م$^3$ و 3 سمكة / م$^3$ ونسبة جنسية 1 ذكر : 2 أنثي.
- التجربة الثانية : ثلاث نسب جنسية هي 1 ذكر : 1 أنثي و 1 ذكر : 2 أنثي و 1 ذكر : 3 أنثي مع كثافة 2سمكة / م$^3$.
- التجربة الثالثة : التداخل بين الكثافات 2 سمكة / م$^3$ و 3 سمكة / م$^3$ والنسب الجنسية (1 ذكر : 1 أنثي و 1 ذكر : 2 أنثي و 1 ذكر : 3 أنثي).

وأظهرت النتائج أن كثافة 2 سمكة / م$^3$ أعطت كميه زريعة لكل سمكة أكثر من كثافة 3 سمكه / م$^3$ والنسبة الجنسية1 ذكر: 1 أنثي أعطت كميه زريعة لكل سمكة أكثر من النسب الأخرى (1 ذكر : 2 أنثي و 1 ذكر : 3 أنثي)، والتداخل بين الكثافات والنسب الجنسية فإن النسبة 1ذكر : 1 انثى مع الكثافة2 سمكة / م$^3$ اعطت كمية زريعة لكل سمكة اكثر من الكثافات والنسب الاخرى. بينما اجمالى انتاج الحوض من الزريعه أكثر كفاءة مع النسبة الجنسية 1 ذكر : 3 أنثي مع كثافة 3 سمكة / م$^3$ وهذا مناسب للمفرخات التجارية.