REPRODUCTIVE BIOLOGY OF *TILAPIA ZILLII* (GERV, 1848) FROM ABU QIR BAY, EGYPT.

EL-SAYED H. KH. AKEL. AND MOHARRAM S. G.

National Institute of Oceanagr & Fish., Alex., Egypt.

Keywords: Reproductive aspects, condition factor, hepatosomatic index, Tilapia zillii, Abu Qir Bay.

ABSTRACT

Reproductive aspects in addition to condition factor and hepatosomatic index were analyzed for Cichlid: *Tilapia zillii* collected from the beach-seiners operating in Abu Qir Bay during the period from June 2000 to February 2001. The overall sex ratio (M/F) was 1: 1.05. Length at first sexual maturity was 8.7 cm for females and 9.7 cm for males. Gonadosomatic index revealed that spawning season lasted from June to September with peak in July. Macroscopic examination for the gonads of *T. zillii* indicated that adult females had 4 stages of gonadal development (matureII; vitellogenic stage III; ripe stage IV and spent V). The maturity stages of males were three (matureII, mature III, and ripe stage). The microscopic features of gonadal sections of both females and males showed that *T. zillii* gonads developed like other tilapian fishes inspite of their hyper saline habitat. Ripe females released in the average about 2138 eggs per cm of length and 2464 eggs per gm of gutted body weight. Condition factor showed similar trend between both sexes of increase and decrease. Hepatosomatic index in females attained higher values over males.

1. INTRODUCTION

Tilapia is the common name applied to three genera of family Cichlidae: (Sarotherodon; Oreochromis and Tilapia) including about 70 species (Meyer, 2002). Reproduction in Tilapia zillii was studied by many authors: (Maclaren, 1981; Latif et al., 1986; Khallaf et al., 1986; El-Haweet, 1991; El-Shazly, 1993; Phillips, 1994; Coward and Bromage, 1999; Negassa and Getahun, 2003 and El-Sawy, 2006). T zillii is highly euryhaline i.e. can tolerate a wide range of salinity (Bayoumi, 1969; El-Zarka et al., 1970; Fryer and Iles, 1972; Chervenski and Horing, 1973 and Meyer, 2002). El-Sayed (2006), mentioned that Tilapia zillii; Oreochromis mosambicus and O.aureus are the most salinity-tolerant tilapia species, while other tilapias are generally less euryhaline, and can grow, survive, and reproduce at 10 - 30 %, depending on the species, size and sex. Optimum temperature tolerance of *T. zillii* was found to be between 28.8 and 31.4 °C (Phillip and Ruwet, 1982). Samaan and Mikhail, (1990) pointed out that Abu Qir Bay was highly fertile. *Tilapia zillii* is a new invader to Abu Qir Bay from Lake Edku and well established in the marine habitat contributing about half of the economic catch by weight of small beachseine (Faltas and Akel, 2003). This species became a distinct population having different biometric characters from that of Lake Edku (Akel, 2005b).

The present study aimed to assess the impact of the marine habitat in Abu Qir Bay on the reproductive biology of *T. zillii*. This study included the reproductive aspects (sex ratio; length at first sexual maturity; gonadosomatic index; gonad maturation; histological examination of the gonads and fecundity) in addition to condition factor and hepatosomatic index.

2. MATERIAL AND METHODS

Abu Qir Bay lies between 30° 5' to 30° 22' E and 31° 16' to 31° 21' N in the Mediterranean Sea. It is situated between Rosetta mouth on the River Nile and Abu Qir town. It is a shallow circular basin with an average depth of 12 m and salinity ranging from 34.8 and 38.7‰ (Zakaria, 2007 – in press- p.c.) (Fig. 1).

A total of 348 specimens of *T. zillii* ranging from 4 - 16 cm total length, were collected monthly from beach – seiners operating in Abu Qir Bay, during the period from June 2000 to February 2001. In the laboratory, total length and total weight were recorded to the nearest centimeter and gram respectively. Condition factor was calculated according to the formula:

$K = W X 100 / L^{3}$

Where W = gutted body weight in g, L = total length in cm. Fishes were dissected, sex maturity stages were determined and macroscopically. Gonads and liver were removed weighed at accuracy of 0.01 gram. Gonadosomatic and hepatosomatic indices were estimated as the percentage of ovaries and liver weights respectively to the gutted body weight. For fecundity estimates the ovaries of 44 full mature and ripe ovaries preserved in 10% formalin. were Determination of maturity stages followed Latif and Saady (1973). Absolute fecundity as the number of mature ova that was likely to be spawned using ripe ovaries of higher gonadosomatic index by the method of Batts (1972). Relative fecundity as the number of eggs per length unit (cm) or gutted body weight (g) was calculated. For histological examination small pieces of gonad were fixed in Bouin's fluid, followed by dehydration in gradual series of alcohol, clearing, embedding in paraffin wax and transverse sections from 3-7 µm were obtained. Histological examination of the mature and ripe gonads was carried out on sections stained by the hematoxylin and eosin method. Oocyte size measurements were made under binocular microscope by eye piece micrometer ($\pm 1 \mu m$). Statistical analysis: t-test and Chi-square analysis were made according to Snedecor and Cochran (1967).

3. RESULTS

3.1. Sex ratio

From Table (1) it appears that the over all sex- ratio (M / F) was 1: 1.05. Occurrence of males was relatively higher than females in autumn (52.7%) and winter (56.7%), while for females, sex ratio was found to occur in higher percentage in summer (54.5%). Chi-square test (X²) showed insignificant seasonal differences between both sexes (P< 0.05).

3.2. Length at first sexual maturity

According to Pitt (1970), the length at which 50% of a fish population reaches sexual maturity (L_{50}) is considered to be the length of the onset of sexual maturity. From Fig. (2), it is also clear that L_{50} for females and males of *T. zillii* was attained at total length of 8.7 cm and 9.7 cm respectively belonging to age group II (Akel , 2005a). Both sexes were all sexually mature above 15 cm total length.

3.3. Gonadosomatic index (GSI)

Monthly variations in GSI revealed that both sexes followed nearly the same pattern. GSI showed higher values during the period from June to September with a peak in July, while the lower ones occurred during the period from October to February (Fig. 3). So, the period from June – September may be representing the spawning season of *T. zillii*. From Fig. (3), it is clear that females acquired higher GSI over males.

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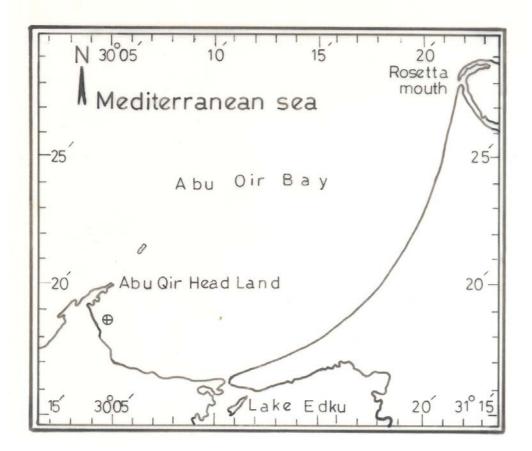


Fig. (1): Study area showing the position of sampling.

 Table (1): Seasonal variation in sex ratio for males and females of T .zillii

 from Abu Qir Bay.

Season	ð		Ŷ		Chi-square	Sex-ratio
	No.	%	No.	%		
					X ²	M / F
Summer	61	45.5	73	54.5	1.075*	1: 0.83
Autumn	58	52.7	52	47.3	0.327*	1:1.12
Winter	59	56.7	45	43.3	1.885*	1:1.31
Total	178	51.15	170	48.85	0.184*	1: 1.05

* $X^2 = 0.184$ (P < 0.05)

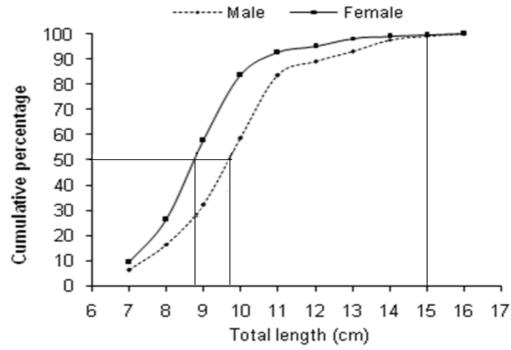


Fig. (2): Length at first sexual maturity of *T. zilii* from Abu Qir Bay.

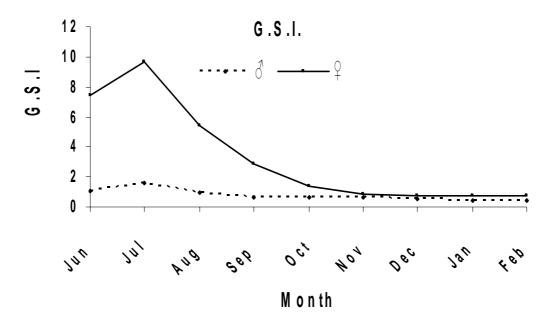


Fig. (3): Monthly variations in gonadosomatic index (GSI) for males and females of *Tilapia* zillii from Abu Qir Bay.

3.4. Gonad maturation

Macroscopic examination of the gonad of *T.zillii*, according to Latif and Saady (1973), showed that small immature males had translucent, thread – like testes while adult males out of spawning season had thread – like testes which were milky white in appearance with evidence of vascularization. The testes of mature males appeared swollen milky white and well vascularized and readily released milt under gentle pressure of the abdomen. Spent males had flaccid empty testes with evidence of hemorrhaging (bleeding).

Immature females had thread–like translucent ovaries. In maturing females (stage II) the ovaries were large occupying one to two third of the body cavity and were well vascularized with blood vessels. Ripe females (stage IV) had swollen green ovaries that occupied 2/3 of the body cavity. The spent ovaries appeared flaccid.

Percentage of gonadal maturity stages were as follows:

In females 32% of the total fish were in mature stage (II); 37% were in mature stage (III) and 31% were in ripe stage (IV). Therefore, 68% of total fish were in the reproductive process.

In males 35% of the total fish were in mature stage (II); 41% were in mature stage (III) and 24% were in ripe stage (IV). So, 65% of the total fish were in reproductive process.

Histological examinations were made for testicular & ovarian tissues of mature gonads:

3.4.1. Females

3.4.1.1. Mature stage (II)

The ovary in this stage contains oocytes in perinucleotic (PG) and previtellogenic (CA) stage. Perinucleotic oocytes were small in size measuring 15-120 μ m and the nucleus was about 60% of the oocyte diameter with one or two nucleoli and intense basophilic cytoplasm. In previtellogenic oocytes (CA) the nucleus occupies about 2/3 of the cell and contains numerous nucleoli arranged under the nuclear membrane (Fig. 4).

3.4.1.2 Vitellogenic stage (III)

Oocytes at this stage measured 450 - 750 µm and contains yolk granules (protein yolk) and large vacuoles (fatty yolk). The nucleus was about 10–15% of the total oocyte diameter (Fig.5). The Zona Radiata (ZR) was visible just beneath the follicle having a diameter of 4 µm. The follicle consisted of two layers: an outer theca layer and inner glandular granulosa layer with 11 µm in thickness (Fig. 6).

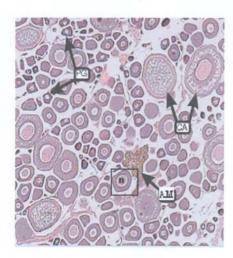
3.4.1.3. Ripe stage (IV)

At this stage (I V) the ovary was densely packed with vitellogenic oocytes and ova with hydrated yolk and could be seen through a thinly stretched ovarian tunica. The oocytes were rounded or ovoid and varied in diameter between $850 - 1550 \mu m$. The nucleus disappeared due to heavy yolk accumulation (Fig. 7).

3.4.1.4. Spent stage (V)

In this stage a few small oocytes were present and was characteized by the presence of atresia of the remaining vitellogenic ova after spawning. Atretic oocytes characterized by breaking down of Z.R and proliferation of granulosa layer which invade the dead ova.and vacant spaces in the ovary (Fig. 8).

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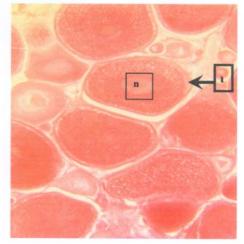


Fig.(4)- T.S. of the ovary of *Tilapia*.zillii stage (II) . X 250 Fig.(4)- 1.5. of the overy of *Inapla.* 2010 stage (II). X 250 PG = primary growth oocyte. CA = corticalalveoli oocyte n =nucleolus.AM = Atretic macrophage.

Fig.(5)- T.S. of the ovary of *Tilapia*.zillii showing the vitollogenic stage (III). X 250. a - vacolised oocyte. n - nucleolus.

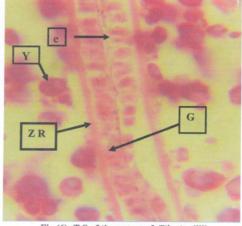


Fig.(6)- T.S.of the ovary of *Tilapia.*zillii showing the epithelial layer, granulosa and Zona-Radiata. X400. e - epithelial follicle. G – granulose. Y – yolk globule. Z R– zona radiata.

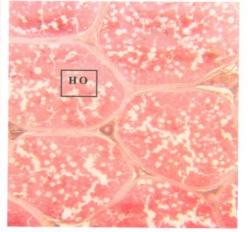


Fig.(7)- T.S.of the ripe ovary of *Tilapia.*zillii showing the ripe oocytes distended with yolk Globules HO- Hydrated oocyte

3.4.2. Males

Histological examination revealed that testis was divided into seminiferous lobules (SL) separated by interstitial tissue which contain interstitial (levding) cells. SL of mature testes contain cysts of spermatogonia and developing germ cells (spermatocytes and spermatids). In ripe testis the lumen of the SL was full of spermtozoa and open into the central sperm duct which was also full of spermatozoa. Sections of the testes of mature males had spermatogonia and spermatocytes (Fig. 9). In stage III the testes of T. zillii showed the presence of both spermatocytes and spermatozoa (Fig. 10). In a section taken from ripe males, the testes had lobules distended with spermatozoa (Fig. 11).

3.5. Fecundity

Absolute and relative fecundity were calculated in relation to fish length and gutted weight.

3.5.1. Fecundity – total length relationship

Table (2) shows the relationship between fecundity (absolute and relative) and total length for *T. zillii*. The relation was found to be curvilinear and represented by the following formulae:

$$Log Fa = 1.8618 + 1.3561 log L. (r = 0)$$

$$.9775$$
) (Fa = absolute fecundity).

Log Fr = $1.8602 + 0.3579 \log L$. (r = 0 .9715) (Fr = relative fecundity).

It was evident from table (2) that the calculated absolute fecundity for *T.zillii* ranged from 1220 and 3124 eggs with a mean 2139 ± 626 for fish of size range 8 to 16 cm total length .Also, it could be detected that the relative fecundity had the same trend of increase from 153 to 196 eggs per length unit with mean 176 ± 10.96 .

3.5.2. Fecundity – gutted weight relationship

The corresponding results concerning the relationship of absolute and relative fecundity to gutted weight of *T. zillii* were given in Table (3).The computed equations expressing these relations were as follows:

$$Log Fa = 2.46035 + 0.5981 log W. (r = 0.0510)$$

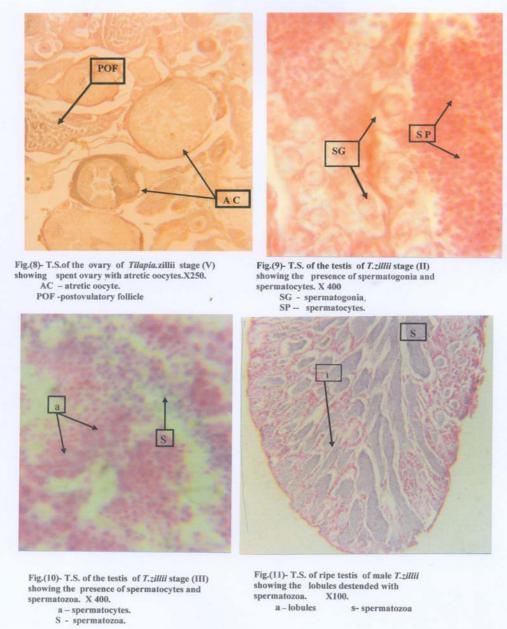
$$Log Fr = 2.1744 + 0.2250 log W. (r = 0.9603)$$

The calculated absolute fecundity ranged between 1144 and 366 eggs with mean 2464 \pm 714.15 for fish with gutted weight ranging from 10 to 70 gram; while relative fecundity showed decreasing values from 89 to 58 eggs with mean 68 \pm 8.21 as the fish increased in weight. Therefore, absolute fecundity increased as the fish increased in weight, while relative fecundity was higher in younger females than older ones.

3.6. Condition factor

Monthly variations in condition factor for males and females of T. zillii were shown in Fig. (12). It is obvious that for males higher values were detected in August (2.11) and November (1.99), meanwhile lower ones were found in September (1.61) and January (1.37). On the other hand, for females, higher values were found in August (2.01); and from October (1.81) to December (1.87) while the lowest value was noticed in September (1.48) (Fig. 12). From the figure, it was evident the conformity and nearly similarity between condition factor of both sexes i.e. they had more or less similar trend of increase and decrease. Statistical analysis (t- test), showed insignificant seasonal differences ($X^2 < 0.01$) between condition factor for males and females.

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Total length (cm.)	<u></u>	Absolute fecundity		Relative fecundity	
	No.	Obs.	Calc.	Obs.	Calc.
8	2	1350	1220	169	153
9	3	1463	1432	163	159
10	5	1680	1652	168	165
11	6	1850	1880	168	171
12	9	1954	2115	163	176
13	8	2340	2357	180	182
14	5	2725	2607	195	186
15	4	2932	2862	196	191
16	2	3448	3124	216	196

Table (2): Relationship between fecundity (absolute & relative) and total length of *T. zillii* from Abu Qir Bay.

 Table (3): Relationship between fecundity (absolute & relative) and gutted weight of T.

 zillii from Abu Qir Bay.

Gutted weight group (g)	No.	Absolute fecundity		Relative	efecundity
		Obs.	Calc.	Obs.	Calc
10	3	1450	1144	145	89
20	9	1680	1732	84	76
30	10	1985	2207	66	70
40	8	2440	2622	60	65
50	5	3225	2996	65	62
60	5	3590	3341	60	60
70	4	4120	3664	59	58

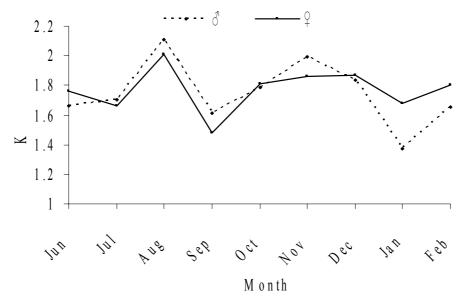


Fig. (12): Monthly variation in condition factor (K) for males and females of *Tilapia zillii* from Abu Qir Bay.

3.7. Hepatosomatic index (H S I)

Hepatosomatic index HSI of *T. zillii* is shown in Fig. (13). Generally, females had higher values from those of males particularly in December. However, indices of both sexes showed similar trend of increase in HSI after breeding season with marked increase in December for females. Lower values of HSI were recorded during spawning season for both sexes.

4. DISCUSSION

The present study in Abu Qir Bay, demonstrated that both sexes of *T.zillii* can survive and reproduce in hypersaline water. This observation conformed with the earlier investigators who reported that *T.zillii* can tolerate high salinity, survive and breed as well in the sea water (El-Zarka, 1956; Fishelson, 1966; Bayoumi, 1969 and El-Sayed, 2006).

It is worthy to mention that this species was clearly missing fom the beach seine catch during spring time and this finding may be related to two factors: A- A phenomenon took place in accordance with blooming in huge amount of the green algae (Caulerpa) and gastropods (Nudibranchia and jellyfish medusae). This flourishing and blooming in this season change the water quality and clogg the fishing nets. Moller, (1983) mentioned comments from many scientists indicating that jellyfish , under certain conditions of blooming and swarming, may interfere significantly with the development of young fish populations. As a result T. zillii was missing from the coastal fishing ground where the beach-seiners used to operate. B-This species in summer time approaches to the shoreside (as substratum spawner) to dig nests in the substratum to spawn and guard their thereafter (Loweprogeny McConnel,1982). Futhermore, the next spring sampling during the present study, confirmed the same finding, where Tilapia zillii was markedly decreased from 50 % of the total catch in winter to only three specimens in spring time. So, the present study was confined to the period from June 2000 to February 2001.

The present study showed that the sex ratio (M / F) of T. zillii in Abu Qir Bay 1 : 1.05 was more or less in agreement with other localities: in Lake Mariut 1: 0.91 (El-Shazly,1993) ; in Lake Edku 1 : 0.91 (Phillips, 1994) and 1: 0.97 (El-Sawy, 2006). Analysis of length at first sexual maturity for T.zillii from different localities was presented in Table (4). From the table, it is noticed that females reach their first sexual maturity before males : 8.7 cm (females) and 9.7 cm (males) in Abu Qir Bay (present study) and this is in agreement with Lake Manzalah 8.6 cm (females) and 10.1 cm (males) (El-Shalloof, 1991). Both sexes reached first sexual maturity at the same length 7 cm in Lake Borollus (El-Haweet, 1991). In contrast to these findings, males reached first sexual maturity at smaller lengths than females: at 9 cm TL for females and 8 cm TL for males in lake Edku (Phillips, 1994). These differences in length at first sexual maturity may be attributed to differences in genetical and environmental conditions such as food supply, population denisty and changes in temperature and salinity (Bardackci and Tanyoloc, 1990; Unlo and Balci, 1993). The present study revealed that, in Abu Qir Bay, T. zillii breeds with maximum activity during June and July. According to Maclaren (1981), cichlids exhibited prolonged spawning season in Lake Manzalah extended from April to September. Negassa and Getahun,(2003) declared that T. zillii in lake Zwai (in Ethiopia) breeds all the year round with peak activites between April and September. This observation was more or less similar to those in other localities: in Lake Borollus (El-Haweet, 1991); Lake Mariut (El-Shazly, 1993) and Lake Edku (Phillips, 1994). Gonadosomatic index of females was found remarkably higher in values over males.

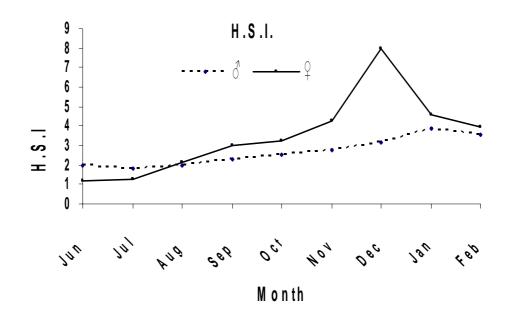


Fig. (13): Monthly variation in hepatosomatic index (H.S.I) for males and females of *Tilapia zillii* from Abu Qir Bay.

Table (4): Comparative results of length at first sexual maturity of <i>T. zillii</i> in	different
localities.	

Author	or Locality		Ŷ
El-Haweet, (1991)	Lake Borollus	7 cm	7 cm
El-Shalloof, (1991)	Lake Manzalah	10.1 cm	8.6 cm
Phillips, (1994)	Lake Edku	8 cm	9cm
Present Study	Abu Qir Bay	9.7 cm	8.7 cm

Normal oogenesis and spermatogenesis in the gonads of *T. zillii* during reproductive cycle were indicated by histological examination and were in accordance with Msiska and Costa-Pierce (1999) for the same species. Results of the present study demonstrated that the wall of vitellogenic ova consisted of three layers (zona radiata , granulosa and theca layer) as indicated by Coward and Bromage (1999)and Assem and El-Zaeem (2005) for *T. zillii*.

Cross sections of testes of *T.zillii* showed all stages of spermatogenesis . Ripe testes were composed mainly of lobules destended with spermatozoa. These findings were in agreement with those of Assem and El-Zaeem (2005).

Observations of fecundity give an indication of the state of the habitat and of the stock (Horwood, 1990). Lowe – McConnel (1982) mentioned that *Tilapia zillii* released six batches of adhesive eggs during its spawning period (6000–40000 egg per year).

The values of the absolute fecundity of *T.zillii* of the length ranging from (8-16 cm) from different lecalities were presented in Table (5) while the equations representing the relationship between absolute fecundity and total length and gutted weight were found to be as follows as given by various authors :

Lake	Log Fa = $1.881+1.388$ Log L.
Borollus	(r = 0.963)
El-Haweet,	Log Fa = $2.512+0.549$ Log W.
1991	(r = 0.977)
Lake Edku Phillips , 1994	Log Fa = $1.5744 + 1.660$ Log L. (r = 0.98561) Log Fa = $2.5931 + 0.5325$ Log W. (r = 0.975)
Abu Qir	Log Fa = $1.8618 + 1.3561 \log L$.
Bay	(r = 0.9775)
Present	Log Fa = $2.46035 + 0.5981 \log W$.
Study	(r = 0.9519)

The absolute fecundity of *T.zillii*, according to the present study, attained lower values in Abu Qir Bay 2139 ± 652 , than in other areas studied. The highest values were found in fresh water- Bhar Shbeen Canal

 (4514 ± 2270) (Latif *et al.*, 1986) followed by: Lake Borollus (2451±782) (ElHaweet, 1991); and Lake Edku (2367 \pm 903) (Phillips, 1994). Applying t-test analysis, between Abu Oir Bay and Lake Edku. the difference was insignificant at (P < 0.05) while it was significant between Abu Qir Bay and Lake Borollus. So , it can be stated that though T.zillii in Abu Qir Bay became well established in the marine environment but still resemble the same population in brackish water (Lake Edku). Rogers (1989), and Wootton (1990), stated that the variation of fecundity within wide limits of fishes of the same length may be due to ecological conditions, genetic factors or total amount of energy given to the ovary during gonad maturation.

Table (6) shows that T.zillii in Abu Qir Bay (Present study) attained values of condition factor (1.74 ± 0.1826) for males and (1.81 ± 0.1086) for females. In Lake Borollus, values of condition factor for this species were (1.6 ± 0.1296) for males and (1.63 ± 0.1311) for females (El-Haweet,1991); in Lake Edku males acquired (1.74 ± 0.027) and females (1.7 ± 0.048) (Phillips, 1994). Using the t-test analysis, the difference between Abu Qir Bay and Lake Borollus was significant at (P>0.05) while the difference between Abu Qir Bay and Lake Edku was insignificant at (P<0.05) and this confirmed that though *T.zillii* population established itself in the marine habitat of Abu Qir Bay but still looks like the same species in the adjacent Lake Edku as mentioned before.

Hepatosomatic index for females of *T. zillii* was generally higher than that for males. Brown (1957), mentioned that the accumulation and storage of fat and protein take place in the liver prior to the spawning in many fishes. Roberts (1978) showed that fermales HSI increase may be due to the increase in the hormones of the sexual activities. He also mentioned that the feeding activity increases after spawning to increase lipids, proteins and water contents of the liver to meet the requirements of yolk deposition in the developing oocytes for the next season. Ellis *et al.* (1978) declared that the

enlargement of the liver resulted from the physiological changes occurred during the prespawning period.

TL (cm)	Bahr Shbeen Canal Latif <i>et al</i> , (1986)	L. Borollus El - Haweet , (1991)	L.Edku Phillips, (1994)	Abu Qir Bay Present Study
8	1638	1332	1048	1220
9	2155	1572	1441	1432
10	2754	1824	1716	1652
11	3439	2086	2010	1880
12	4212	2858	2323	2115
13	5076	2639	2653	2357
14	6032	2929	3000	2607
15	7084	3228	3364	2862
16	8233	3595	3745	3124
Mean	4514± 2270	2451±782	2367±903	2139± 626

Table (5): Absolute fecundity of *T. zillii* in different localities.

Table (6): Comparative results of condition fa	actor K for <i>T. zillii</i> in different localities.
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Author	Locality	Mean ± S.D.		
		6	Ŷ	
El-Haweet, (1991)	Lake Borollus	1.6 ± 0.1296	1.63 ± 0.1311	
Phillips, (1994)	Lake Edku	1.74 ± 0.027	1.7 ± 0.048	
Present Study	Abu Qir Bay	1.74 ± 0.1806	1.81 ± 0.1086	

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