Journal of Coastal Life Medicine

journal homepage: www.jclmm.com

Original Research Article doi: 10.12980/JCLM.3.2015J5-20

Gonadal maturation and histological observations of the grey triggerfish *Balistes capriscus* Gmelin, 1789 (Teleostei: Balistidae) in the Gulf of Gabès, Tunisia

Hichem Kacem*, Lobna boudaya, Lassad Neifar

Laboratory of Biodiversity and Aquatic Ecosystems, Department of Life Sciences, Faculty of Sciences of Sfax, University of Sfax, PC 1171, 3000 Sfax, Tunisia

ARTICLE INFO

Article history:
Received 2 Mar 2015
Received in revised form 16 Mar, 2nd revised form 17 Mar, 3rd revised form 30 Mar 2015
Accepted 9 Apr 2015
Available online 14 Apr 2015

Keywords:
Gonad histology
Gonadosomatic index
Sexual maturation
Balistidae
Spawning seasonality

ABSTRACT

Objective: To determine the spawning activity using gonadosomatic index (GSI) and gonad histology the *Balistes capriscus* (Teleostei: Balistidae) of the Gulf of Gabès (Southern Tunisia, Central Mediterranean Sea).
Methods: The reproductive biology of the species, based on 756 (480 females and 276 males), collected from commercial catches at several fishing ports including Chebba, Kerkennah and Zarzis at respective GPS coordinates (34°14' N, 11°06' E), (34°45' N, 11°17' E), (33°41' N, 11°48' E) was studied over 28 months (January 2008-April 2010) using GSI and gonad histology. Sizes used in this study ranged from 11.30 to 45.60 cm in fork length.
Results: Both GSI and gonad histology suggest that spawning activity occurred mainly between July and mid-September with a peak in July, coinciding with summer time. The first maturation occurred at 20.26 cm fork length for females and 21.30 cm fork length for males. The monthly values of hepatosomatic index and condition factor (K) indicated that the liver is the main organ responsible for the mobilization process of the energizing reserves during the sexual cycle.
Conclusions: It is the first inventory of gonadal maturation and histological observations of the grey triggerfish *Balistes capriscus* Gmelin, 1789 (Teleostei: Balistidae) in the Gulf of Gabès, (Southern Tunisia, Central Mediterranean Sea).

1. Introduction

The gray triggerfish *Balistes capriscus* (*B. capriscus*) (Teleostei: Balistidae) is a reef fish broadly distributed in temperate and tropical waters of the Atlantic basin[1], from Nova Scotia to Argentina and throughout the Gulf of Mexico in the Western Atlantic and, in the Eastern Atlantic, from Ireland to Southwestern Africa[2], including the Mediterranean Sea[3]. *B. capriscus* is a member of thermophilic fishes, typical from the southern sectors of the Mediterranean and their emergence is consistent with what we would expect from the climate warming[4]. Remarkably the recent literature is rich in records of these species moving northward with respect to their previously known distribution[5].

This fish is considered as one of the most important species that are commonly associated with coral reefs, wrecks, outcroppings, artificial structure and hard bottom areas[6].

Recently, a considerable increase in the *B. capriscus* production was depicted along the Tunisian coast and particularly in the Gulf of Gabès. Catching is about six fold increase from 34 tons in 1993 to 219 tons in 2011[7].

Despite the increase of production of the gray triggerfish in the Gulf of Gabés added to both the high commercial value of this fish belonging to its appreciated quality of flesh, its traditional therapeutic practices [8-10], no peer-reviewed published data exist concerning the annual reproductive cycle of *B. capriscus* collected from the Southeastern Mediterranean Sea.

Previous studies on the reproductive biology have been focused

*Corresponding author: Hichem Kacem, Laboratory of Biodiversity and Aquatic Ecosystems, Department of Life Sciences, Faculty of Sciences of Sfax, University of Sfax, PC 1171, 3000 Sfax, Tunisia.
Tel: 0021698483426
Fax: 0021674276400
E-mail: hichemkacem2007@yahoo.fr
Foundation Project: Supported by the Faculty of Sciences, Sfax, Tunisia.

on determining the spawning season and size at sexual maturity of *B. capriscus*. They have focused on certain distribution areas: Western Atlantic, Eastern Atlantic and Northeastern Mediterranean[11].

The aim of this study was to examine the annual reproductive cycle of *B. capriscus* using the gonadosomatic index (GSI) and gonad histology and to evaluate its spawning seasonality and sexual maturation in the Gulf of Gabès.

Obtained results could provide essential life history data for any future stock assessments and may serve as a guideline for future management decisions of the gray triggerfish population of the Gulf of Gabès.

2. Materials and methods

Biological data collected monthly from 756 specimens (480 females and 276 males) obtained commercial catches at different fishing ports at several fishing ports including Chebba, Kerkennah and Zarzis at respective GPS coordinates (34°14' N, 11°06' E), (34°45' N, 11°17' E), (33°41' N, 11°48' E) (Figure 1), was studied over 28 months (January 2008-April 2010). In the laboratory, the following data were recorded for all individuals: sex, total length, fork length and standard length to the nearest mm; total weights and eviscerated fish weights to the nearest 0.1 g and the weights of gonads and livers were recorded to the nearest 0.01 g. *B. capriscus* is a gonochoristic fish. Females and males could be distinguished by the skin colour at the upper side. The former has a white colour, while the latter shows a purplish to bluish reflection. This dichromatism is confirmed with application of the squash mount technique which is more pronounced and more accentuated during the reproduction period. Sex-ratios (number of females / number of males) were calculated according to the size. Month and season were compared by applying the χ^2 test.

The monthly mean GSI, the hepatosomatic indices (HSI) and the condition coefficient (K) were calculated for all sexually identified specimens and for both sexes as follows:

$GSI\ (\%) = (GW / EW) \times 100$; $HSI = (LW / EW) \times 100$; $K = (EW / FL^3) \times 100$

Where GW = gonads weight (g); LW = liver weight (g); EW = eviscerated weight (g) and FL = fork length (cm).

Correlation between GSI and temperature was tested using the Spearman rank correlation coefficient (r_s)[12]. This temperature is the sea water average temperature which was supplied by the National Institute of Meteorology in Tunisia.

Variations in the mean of these indexes during the reproductive period were tested using One-way analysis of variance. The *post-hoc* Turkey's test was performed to determine the pair-wise differences.

Ovaries from 280 individuals were preserved in Bouin's solution in order to allow for histological analysis and were transferred to

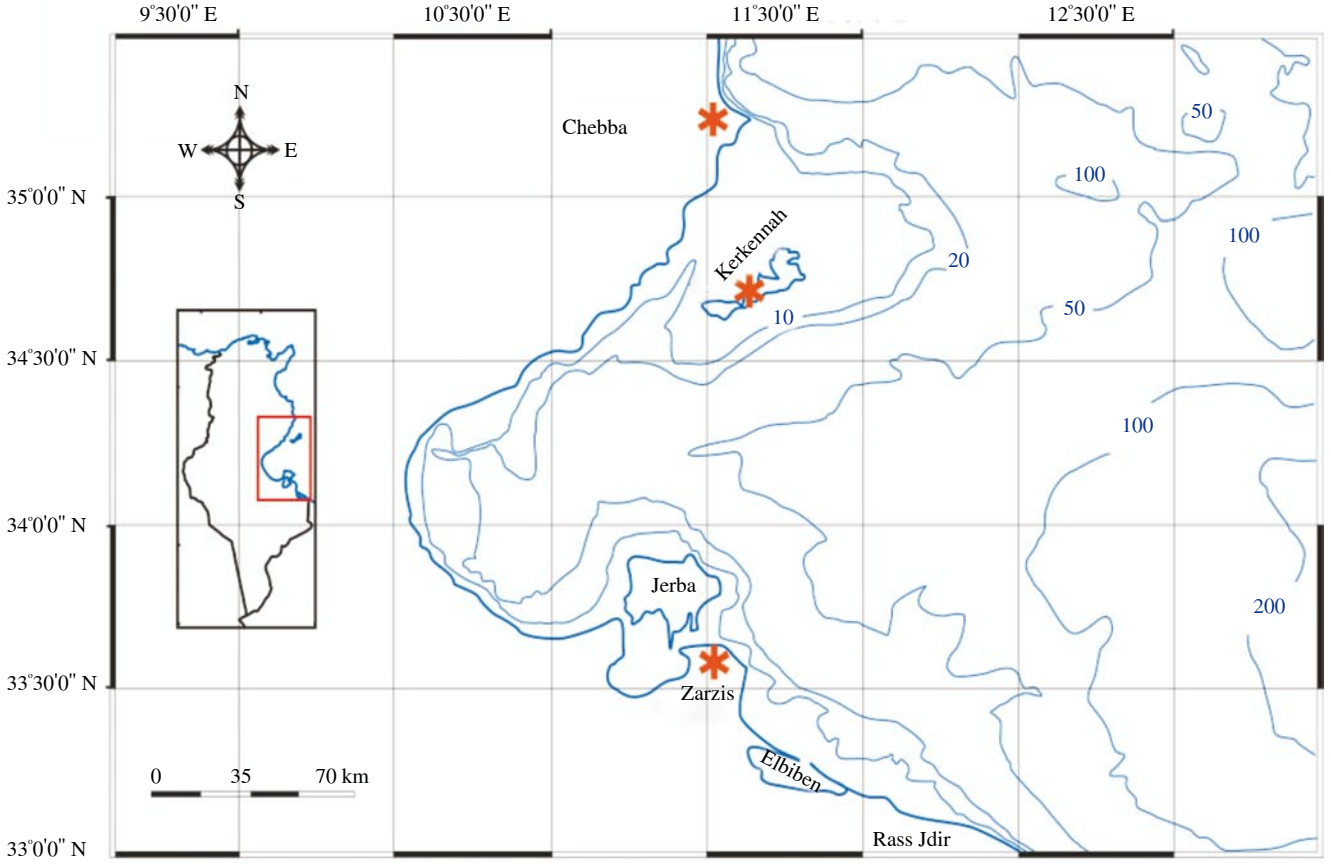

Figure 1. Map of the Gulf of Gabès describing the location of fishing port which collected *B. capriscus*.

50% ethanol after 24 h. The preserved ovaries were embedded in paraffin, sectioned into 6 μm thick slices, and stained with hematoxylin and eosin[13]. Six maturity phases were established for females as follow: I-immature, II-developing, III-spawning capable, IV-actively spawning, V-regressing, VI-recovering.

Ovarian developmental stages were assessed microscopically using a light microscope. In addition, the relative frequency of the different sizes of oocyte was estimated[14]. The diameters for each oocyte development stage were measured with an ocular micrometer. The terminology used for describing the different oocyte stages and ovarian phases is based on Brown-Peterson *et al*[15].

The mean size at first sexual maturity was estimated for both sexes by using a logistic function connecting the proportion P of mature individuals and fork length. This function was calculated as follow[16]:

$$P = \frac{1}{[1 + e^{-r(L - L_{50})}]}$$

Where P is the proportion of mature individuals; L is the fork length corresponding to the proportion (P); r is constant and L_{50} is the mean size at first sexual maturity; it was taken as the size at which 50% of individuals were mature.

3. Results

Out of 765 specimens *B. capriscus* sampled during this study, there were 480 females and 276 males. The fork length size ranges were 11.30-42.90 cm and the weight varied from 39.86 to 1 858.37 g for female whereas for male the fork length size and weight ranged between 11.90-45.60 cm and 37.97-2 318.07 g respectively.

Furthermore, sexual dimorphism was observed. The mean size of males (27.09 cm fork length) was significantly larger than females (26.74 cm fork length) based on the entire dataset ($P < 0.05$). An overall sex ratio (females / males) of (1: 1.74) which significantly deviated from the hypothetical distribution of 1:1, was significantly in favor of females (χ^2, $P < 0.05$) (Table 1).

The curves of the GSI monthly values were unimodal for both males and females (Figure 2). Thus, the *B. capriscus* is reproduced once a year.

Table 1

The fork length of *B. capriscus* in the Gulf of Gabès.

Parameter	Fork length (cm)		
	All fish	Females	Males
Min	11.30	11.30	11.90
Max	45.60	42.90	45.60
Mean ± SD	26.87 ± 8.03	26.74 ± 7.55	27.09 ± 8.91
Mode	18.00	18.00	28.50

Min: Minimum; Max: Maximum.

The GSI began to increase in May, and it reached the maximal values in July for both females (13.58) and males (0.95). The index values for July were significantly higher for females [F = 15,632; df (11,479); P, 0.001] and males [F = 2,056.36; df (11,275); $P < 0.001$]. This period constituted the phase of maturation where the gonads were well developed and occupied almost all of the abdominal cavity. This peak was followed by a fast decrease sharply until mid-September. The breeding season extended from July to mid-September. Moreover, 85% of females finished egg deposition in August and reproduction was completed in mid-September. The period between October and April constituted the phase of the sexual rest when gonads remained filamentous.

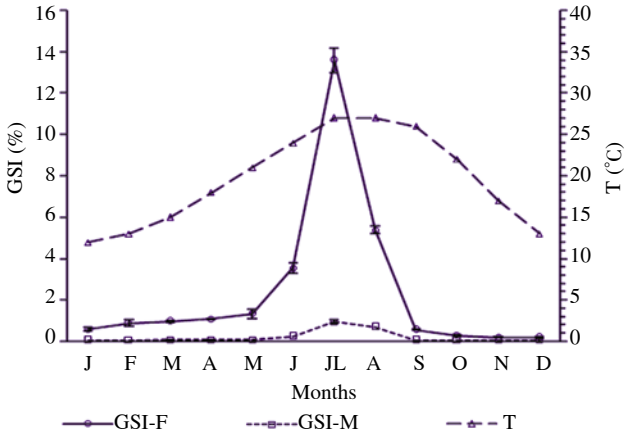

Figure 2. Monthly GSI, mean values for both sexes of grey triggerfish *B. capriscus* and of the mean temperature of sea water (T °C) in the Gulf of Gabès.

Temperature data were supplied by the National Institute of Meteorology. Values are expressed as mean ± SD. GSI-F: Females GSI; GSI-M: Males GSI; T: Mean temperature of sea water (°C).

Significant correlations were obtained between the average GSI values of both sexes (females and males) and sea surface temperature (r_s = 0.582; n = 12; $P < 0.05$ for females) (r_s = 0.628; n = 12; $P < 0.05$ for males) (Figure 2). Based on these correlations, we noted that the complete gonad maturity occurred at the highest sea surface temperature (27 °C).

The HSI of females (8.92 ± 2.01) and males (9.05 ± 1.75) were not significantly different (Mann-Whitney U-test, Z = -0.89, $P > 0.05$). HSI of both sexes showed a clear seasonal pattern, demonstrating a significant variation across months (females: Kruskal-Wallis test, H = 161.13, $P < 0.001$; males: Kruskal-Wallis test, H = 162.37, $P < 0.001$).

The HSI cyclic evolution for both sexes was slightly synchronous with that of GSI showing a peak in June (female HSI: 11.89 ± 0.62 and male HSI: 11.65 ± 0.18), while the condition factor K showed no fluctuation along the year (Figure 3).

The ovarian lamellae inside the ovarian stroma indicated the presence of germinative cells at different oocyte development stages, according to the stage of gonadal maturity. The germinative cells or oocytes developed gradually inside the ovary. Eggs began

as oogonia and went through similar stages of oocyte development, which was commonly divided into primary growth and secondary growth beginning with the CA stage and then proceeding through vitellogenesis (Vtg), which could be broken into substages associated with the extent of yolk globules or platelets in the ooplasm [primary (Vtg1), secondary (Vtg2), and tertiary (Vtg3)]. Oocyte maturation occurred after the appropriate trigger and could include germinal vesicle migration, yolk coalescence and hydration. At ovulation, the follicle ruptures and the oocyte was released. POFs remained in the ovary, where they were resorbed.

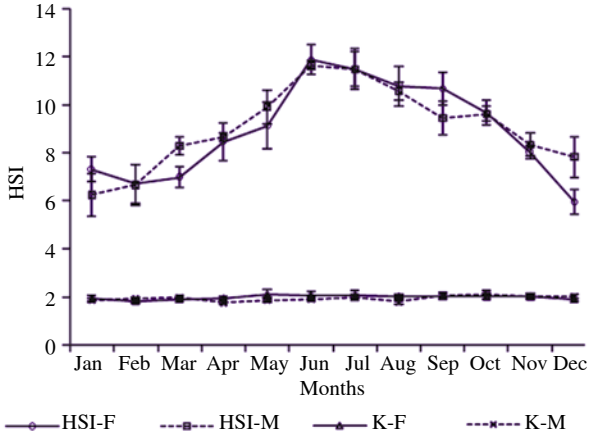

Figure 3. Monthly HSI and condition factor K for both sexes of grey triggerfish *B. capriscus* in the Gulf of Gabès.
Values are expressed as mean ± SD. HSI-F: Females HSI; HSI-M: Males HSI; K-F: Females condition factor K; K-M: Males condition factor K.

The characteristic of oocyte development stage observed on histological section of ovaries grey triggerfish *B. capriscus* Gmelin, 1789 in the Gulf of Gabès, Tunisia were described as following:

1. Chromatin nuclear: Small oocyte ranging between 53 and 75 μm, which were round and/or oval. The nucleus was large and centrally located, surrounded by a thin layer of cytoplasm and containing a large and single nucleolus (Figure 4A).

2. Perinucleolar: Round and/or oval cells measuring between 75 and 94 μm (mean = 84.5 μm) with basophilic cytoplasm, with slightly more color than during the previous stage. Nucleus increased in size and multiple nucleoli appeared at its periphery (Figure 4B).

3. Cortical alveoli formation: Cell measuring between 220 and 242 μm (mean = 231 μm), spherical vesicles started to appear at the periphery of the cytoplasm. They increased in size and number to form several peripheral rows and give rise to cortical alveoli. Oil drops began to accumulate in the cytoplasm. At this stage chorion and follicle layers seemed to be apparent (Figure 4C).

Oocyte measuring between 324 and 374 μm (mean = 350 μm) emerge of some yolk vesicles in the cytoplasm. Besides, the separation of the chorion in two different layers occurred: inner and outer zona radiate (Figure 4D).

This stage was subdivided in three different steps:

• Vtg1: Oil droplets occupied more cytoplasmic area than yolk granules (Figure 4E).

• Vtg2: Oil droplets occupied similar cytoplasmic area than yolk granules (Figure 4E).

• Vtg3: Oil droplets occupied less cytoplasmic area than yolk granules (Figure 4E).

4. Germinal vesicle migration: The nucleus started to migrate to the animal pole and the oil droplets fused to coalescence into a unique oil globule.

5. Germinal vesicle breakdown: Oocyte measuring between 370 and 489 μm (mean = 457.5 μm). The nucleus completed its migration to the animal pole and the unique oil droplet was clearly evident at central part of the oocyte (Figure 4F).

6. Hydration: Yolk granules fused in yolk plates, and eventually form a homogeneous mass. The nucleus had disintegrated and the cortical alveoli and cytoplasm were restricted to a thin peripheral layer. The oocyte significantly increased in size due to the uptake of fluids. Hydrated oocyte had a translucent appearance. At ovulation, the follicle ruptured and the oocyte was released. POF remained in the ovary, where they were resorbed.

The gonad maturation phases for females are described in Table 2. Monthly distribution of proportional female maturity phases showed that the period of spawning activity occurred mainly between July and mid-September with a peak in July (Figures 2 and 5).

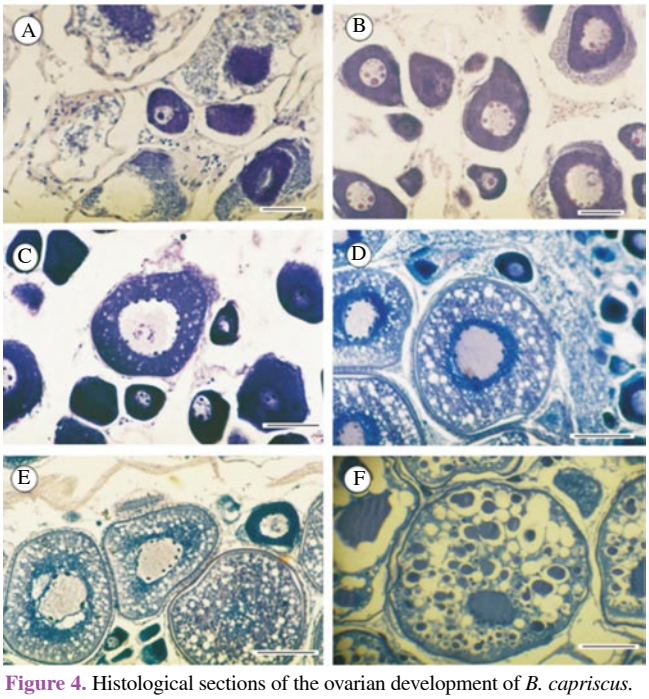

Figure 4. Histological sections of the ovarian development of *B. capriscus*.
A: Ovary in the immature stage, showing young germ cells (scale bar 40 μm); B: Oocytes in the immature stage, with multiple nucleoli within the nucleus (scale bar 40 μm); C: Cortical alveolar (scale bar 50 μm); D: germinal vesicle migration; E: Vtg1, Vtg2 and Vtg3; F: Germinal vesicle breakdown (scale bar 75 μm).

Table 2

Microscopic characterization of the maturity phases of the ovaries of grey triggerfish *B. capriscus* Gmelin, 1789 in the Gulf of Gabès, Tunisia.

Maturity phases	Microscopic characteristics
I. Immature	Well-defined cellular organization with the presence of several unyolked oocytes in perinuclear regions. Only oogonia and primary growth oocytes.
II. Developing	Oogones, CA, Vtg1 and Vtg2. No evidence of Vtg3.
III. Spawning capable	All oocyte stages present and abundance of Vtg3.
IV. Actively spawning	Hydrated oocytes (translucent) in large quantities and with few POFs.
V. Regressing	Few POFs, atresia (any stage), some residual CA and Vtg1-Vtg2 oocytes.
VI. Recovering	Reorganization of unyolked oocytes to the beginning of a new reproductive cycle, and atresia. Only oogonia and primary growth oocytes present.

Adapted from Brown-Peterson *et al.*[15], Vieira *et al*.[17] and Chen *et al*[18]. CA: Cortical alveolar; Vtg1: Primary vitellogenic oocytes; Vtg2: Secondary vitellogenic oocytes; Vtg3: Tertiary vitellogenic oocytes; POFs: Postovulatory follicles.

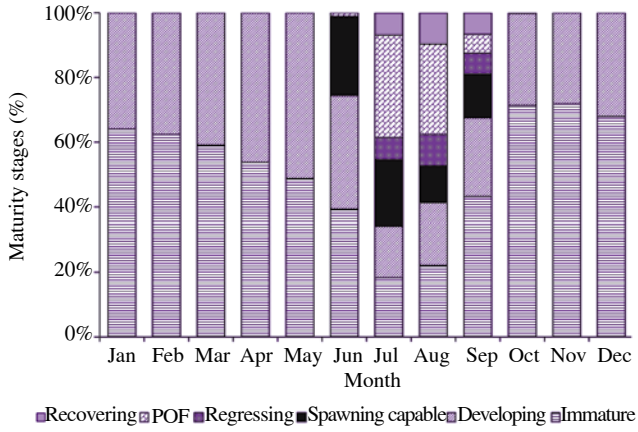

Figure 5. Monthly distribution of the maturational phases for female *B. capriscus* collected from commercial catches at several fishing ports between January 2008-April 2010 (n = 280).

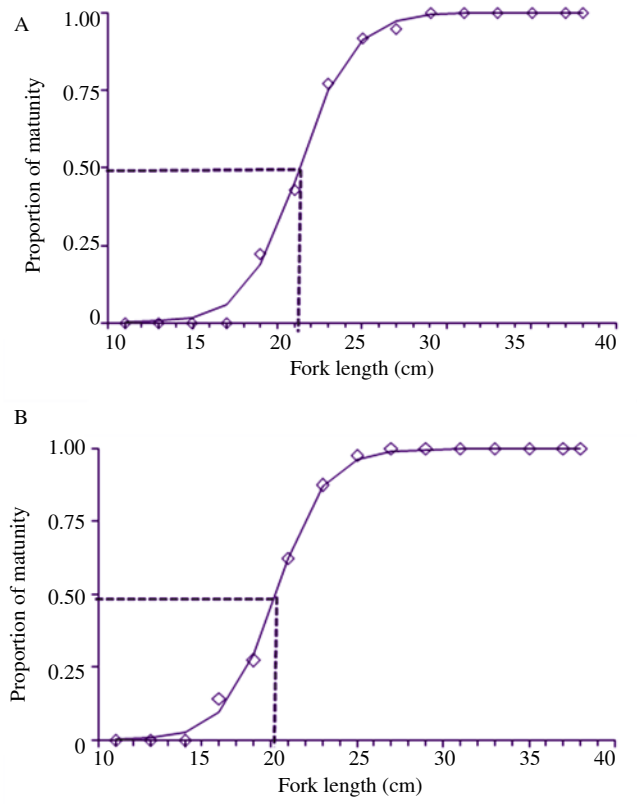

Figure 6. Size at first sexual maturity for *B. capriscus* in the Gulf of Gabès. A: Males; B: Females.

The logistic curves showed the estimated relationship between the maturity proportion in each length interval and the fork length. The results obtained by the application of the logistical function showed that the size at first sexual maturity was 20.26 cm for females and 21.30 cm for males (Figure 6). The χ^2 test did not show any significant differences between the theoretical proportions and the observed ones and this non-linear logistic regression model had an excellent fit (r^2 = 0.98).

4. Discussion

Sexual dichromatism concerning the color of the upper side was already observed in *B. capriscus*, collected from the Gulf of Gabès. This character has been described by several authors in other sites. Garnaud proved that during the period of reproduction, the male has more pronounced color[19]. Gerlotto and Stéquert reported that *B. capriscus* has a sexual dichromatism in which males and females were different in skin color at the sub-opercular region[20]. Mackichan and Szedlmayer showed that the dominant males could be identified by coloration and behavior[21].

Furthermore, sexual dimorphism in the length and weight has been also observed for this species in the Gulf of Gabès. In fact, mean lengths of males were significantly larger than that of females. Similar findings have been reported for gray triggerfish in the Southeastern US, Atlantic coast[22], and Gulf of Mexico[23]. The larger size of male gray triggerfish could be advantageous, allowing them to more adequately defend the nests in order to optimize survival of the eggs.

In the Gulf of Gabès, the sex ratio of *B. capriscus* is generally favorable to females. It is also the case in the Northeastern Gulf of Mexico[24] and in South Atlantic[25]. Only the populations of Alexandretta (Southern Turkey) have a male-dominated ratio[26]. The imbalance of the sex can be attributed to longevity, a differential catchability of the sexes or a spatio-temporal segregation of the sexes[27]. In the Gulf of Gabès, female dominance is probably related to a spatio-temporal distribution of the sexes.

Dependent upon location, *B. capriscus* population exhibited different reproduction period, which occurs mainly during the warmer months (July to mid-September) in the Gulf of Gabès, during November-December on the coast of Ghana[28] and during June-July in the Gulf of Mexico[29]. The water temperature probably plays a major role in the oocytes emission.

In general, this period coincides with favorable environmental

conditions for the survival and growth of larvae. In fact, low temperature may substantially decrease the chance of survival for fish larvae[30]. In the Gulf of Gabès, the reproduction period of *B. capriscus* begins in May, corresponding to the time when the water temperature increases and the zooplankton productivity is the highest in the Mediterranean[31] and particularly in the Gulf of Gabès[32]. Mediterranean subtidal plant communities (algae and seagrasses) begin to develop at this time[33]. This temporal coincidence allows the development of young triggerfish which find food resources in addition to the maintenance of rapid growth, development and protection within the subtidal seagrass and algae.

Like many fish species, the gonad index of *B. capriscus* is lower in males than females[34]. In fact, these index values for *B. capriscus* females in Tunisian coasts are higher than those found in the Gulf of Guinea[35]. These differences may be related to genetic potential variations and also to food resources availability. This suggests important fertility among individuals in the Gulf of Gabès.

Fish may store the energy required for spawning, in the liver or the muscles. However, the fluctuations of the HSI are relatively synchronous with those of the GSI for both females and males. However no relationship between K and spawning were clearly depicted. This suggests that the required energy for spawning might be derived from *B. capriscus* liver accumulating lipid reserves to be spent during laying. However, the muscles do not take part in the energy supplying for both sexes reproduction.

The obtained results (GSI) are agreed with gonad histology analysis. Effectively the period of spawning activity occurred mainly between July and mid-September.

Based on the method of the logistic curve, we estimated the size at maturity to be 20.26 cm fork length and 21.30 cm fork length for females and males, respectively. In São Paulo, Brazil, the length of the first sexual maturity is 16.9 cm fork length for females and 20.0 cm fork length for males[36]. However in South Atlantic sexual maturity is reached at 17.3 cm fork length for females and 18.1 cm fork length for males[25]. These differences are explained by the fact that the size of mature individuals depends on the biological factors and/or the ecological environment[37]. Human activities such as overfishing (increased fishing effort) can also explain the differences in size[38]. Fish subjected to these pressures may have ecophysiological adaptations related to the growth or the reproduction, the dwarfism or the early sexual maturity, in order to ensure their survival[39].

Male *B. capriscus* attained 50% sexual maturity (21.30 cm) almost 1 cm larger than that of female (20.26 cm). The maturation of female at smaller size than that of male is consistent with findings for triggerfish *Balistes vetula* in Brazil (fork length = 23.5 cm female, male 26.5 cm)[40]. This could be explained by the differential growth rates between the sexes[41].

Conflict of interest statement

We declare that we have no conflict of interest.

References

[1] Antoni L, Emerick N, Saillant E. Genetic variation of gray triggerfish in US waters of the Gulf of Mexico and Western Atlantic regions as inferred from mitochondrial DNA sequences. *N Am J Fish Manag* 2011; **31**: 714-21.

[2] Burton ML, Potts JC, Carr DR, Cooper M, Lewis J. Age, growth, and mortality of gray triggerfish (*Balistes capriscus*) from the Southeastern United States. *Fish Bull* 2015; **113**: 27-39.

[3] Aggrey-Fynn J. Distribution and growth of grey triggerfish, *Balistes capriscus* (Family: Balistidae), in Western Gulf of Guinea. *W Afr J Appl Ecol* 2009; doi: 10.4314/wajae.v15i1.49421.

[4] Azzurro E, Moschella P, Maynou F. Tracking signals of change in Mediterranean fish diversity based on local ecological knowledge. *PLoS One* 2011; doi: 10.1371/journal.pone.0024885.

[5] Dulči J, Soldo A. On the occurrence of the yelowmouth barracuda *Sphyraena viridensis* Cuvier, 1829 (Pisces: Sphyraenidae) in the Adriatic Sea. *Ann Ser Hist Nat* 2004; **14**(2): 225-30.

[6] Lingo ME, Szedlmayer ST. The influence of habitat complexity on reef fish communities in the Northeastern Gulf of Mexico. *Environ Biol Fish* 2006; **76**: 71-80.

[7] Food and Agriculture Organization of the United Nations. GFCM (Mediterranean and Black Sea) capture production. Roman: Food and Agriculture Organization of the United Nations; 2014. [Online] Available from: http://www.fao.org/fishery/statistics/gfcm-capture-production/en [Accessed on 28th March, 2015]

[8] Alves RR, Alves HN. The faunal drugstore: animal-based remedies used in traditional medicine in Latin America. *J Ethnobiol Ethnomed* 2011; doi: 10.1186/1746-4269-7-9.

[9] El-Deir AC1, Collier CA, de Almeida Neto MS, Silva KM, Policarpo Ida S, Araújo TA, et al. Ichthyofauna used in traditional medicine in Brazil. *Evid Based Complement Alternat Med* 2012; doi: 10.1155/2012/474716.

[10] Alves RRN, Oliveira TPR, Rosa IL. Wild animals used as food medicine in Brazil. *Evid Based Complement Alternat Med* 2013; doi: 10.1155/2013/670352.

[11] Kacem H, Neifar L. The reproductive biology of the grey triggerfish *Balistes capriscus* (Pisces: Balistidae) in the Gulf of Gabès (South-eastern Mediterranean Sea). *J Mar Biol Assoc U K* 2014; **94**: 1531-7.

[12] Zar JH. *Biostatistical analysis*. New Jersey: Prentice-Hall; 1999.

[13] Chellappa S, Câmara MR, Verani JR. Ovarian development in the Amazonian red discus, *Symphysodon discus* Heckel (Osteichthyes: Cichlidae). *Braz J Biol* 2005; **65**: 609-16.

[14] West G. Methods of assessing ovarian development in fishes: a review. *Aust J Mar Freshwater Res* 1990; **41**: 199-222.

[15] Brown-Peterson NJ, Wyanski DM, Saborido-Rey F, Macewicz BJ, Lowerre-Barbieri SK, Brown-Peterson NJ. A standardized terminology for describing reproductive development in fishes. *Mar Coast Fish* 2011; **3**: 52-70.

[16] Pauly D. On the interrelationships between natural mortality, growth parameters, and mean environmental temperature in 175 fish stocks. *J Cons Int Exp Mer* 1980; **39**: 175-92.

[17] Vieira KR, Oliveira JEL, Barbalho MC, Garcia J Jr. Reproductive characteristics of blackfin tuna *Thunnus atlanticus* (Lesson, 1831), in Northeast Brazil. *Collect Vol Sci Pap ICCAT* 2005; **58**: 1629-34.

[18] Chen KS, Crone PR, Hsu CC. Reproductive biology of albacore *Thunnus alalunga*. *J Fish Biol* 2010; **77**: 119-36.

[19] Garnaud J. [Bridge, hatch, the larva of triggerfish Grey triggerfish Linné 1758]. *Bulletin de l'Institut Océanographique de Monaco* 1960; **1169**: 1-6. Figs. 1-2. French.

[20] Gerlotto F, Stéquert B. [First results for comments on the biology of grey triggerfish *Balistes capriscus* (Gmel.) in Guinea and Senegal]. [Online] Available from: http://horizon.documentation.ird.fr/exl-doc/pleins_textes/divers11-01/02568.pdf [Accessed on 28th March, 2015] French.

[21] MacKichan CA, Szedlmayer ST, editors. Reproductive behavior of gray triggerfish, *Balistes capriscus*, in the Northeastern Gulf of Mexico. Belize City: Proceedings of the 59th Gulf and Caribbean Fisheries Institute; 2007, p. 231-6. [Online] Available from: http://aquaticcommons.org/13182/1/gcfi_59-30.pdf [Accessed on 28th March, 2015]

[22] Kelly MA. Age, growth, and reproduction of gray triggerfish *Balistes capriscus* off the Southeastern U.S. Atlantic Coast [dissertation]. Charleston: College of Charleston; 2014. [Online] Available from: http://sedarweb.org/docs/wsupp/S41_RD36_Kelly2014_GTF_AgeGrowthRepro.pdf [Accessed on 28th March, 2015]

[23] Ingram GW Jr. Stock structure of gray triggerfish, *Balistes capriscus*, on multiple spatial scales in the Gulf of Mexico [dissertation]. Alabama: University of South Alabama; 2001, p. 73.

[24] Simmons CM, Szedlmayer ST. Territoriality, reproductive behavior, and parental care in gray triggerfish, *Balistes capriscus*, from the Northern Gulf of Mexico. *Bull Mar Sci* 2012; **88**: 197-209.

[25] Kolmos KJ, Smart T, Wyanski D, Kelley A, Reichert M. Marine resources monitoring, assessment and prediction program: report on South Atlantic gray triggerfish, *Balistes capriscus*, for the SEDAR 32 data workshop. SEDAR32-DW05. SEDAR, North Charleston, SC. 2013; p. 50. [Online] Available from: http://www.sefsc.noaa.gov/sedar/download/SEDAR32_DW05_Kolmos_etal._3.4.2013_FINAL.pdf?id=DOCUMENT [Accessed on 28th March, 2015]

[26] Ismen A, Türkoglu M, Yigin CÇ. The age, growth, and reproductive of gray triggerfish (*Balistes capriscus*, Gmelin, 1789) in Skenderun Bay. *Pak J Biol Sci* 2004; **7**(12): 2135-8.

[27] Morato T, Afonso P, Lourindo P, Nash RDM, Santos RS. Reproductive biology and recruitment of sea bream in the Azores. *J Fish Biol* 2003; **63**: 59-72.

[28] Ofori-Danson PK. Reproductive ecology of the triggerfish, *Balistes capriscus*, from the Ghanian coastal waters. *Trop Ecol* 1990; **31**: 1-11.

[29] Wilson CA, Nieland DL, Stanley AL. Age, growth, and reproductive biology of gray triggerfish, *Balistes capriscus*, from the Northern Gulf of Mexico commercial harvest. MARFIN Final Report. Baton Rouge: Louisiana State University; 1995.

[30] Green BS, Fisher R. Temperature influences swimming speed, growth and larval duration in coral reef fish larvae. *J Exp Mar Biol Ecol* 2004; **299**(1): 115-32.

[31] Macpherson E, Raventos N. Relationship between pelagic larval duration and geographic distribution of Mediterranean littoral fishes. *Mar Ecol Prog Ser* 2006; **327**: 257-65.

[32] Drira Z. Contribution to the understanding of how the Gulf of Gabes: study of dynamic and structural characteristics phyto-zooplankton communities in relation to the variability environmental and hydrographic characteristics of areas coastal and ocean [dissertation]. Sfax: University of Sfax; 2009. [Online] Available from: http://indexation.univ-fcomte.fr/nuxeo/site/esupversions/86664012-6524-421e-a05b-fa1cdf1c1313 [Accessed on 28th March, 2015] French.

[33] Alcoverro T, Romero J, Duarte CM, López NI. Spatial and temporal variations in nutrient limitation of seagrass *Posidonia oceanica* growth in the NW Mediterranean. *Mar Ecol Prog Ser* 1997; **120**: 155-61.

[34] Baylis JR. The evolution of parental care in fishes, with reference to Darwin's rule of male sexual selection. *Environ Biol Fish* 1981; **6**: 223-51.

[35] Bernard S. [First acoustic evaluation of demersal resources in the continental shelf of Guinea-Bissau]. Rome: Food and Agriculture Organization; 1979, p. 91-9. [Online] Available from: http://www.documentation.ird.fr/hor/fdi:38813 [Accessed on 28th March, 2015] French.

[36] Bernardes RA. Age, growth and longevity of the gray triggerfish, *Balistes capriscus* (Tetraodontiformes: Balistidae), from the Southeastern Brazilian Coast. *Sci Mar* 2002; **66**: 167-73.

[37] Udupa KS. Statistical method of estimating the size at first maturity in fishes. Mangalore: University of Agricultural Sciences, College of Fisheries. [Online] Available from: http://www.worldfishcenter.org/Naga/na_2900.pdf [Accessed on 28th March, 2015]

[38] Jennings S, Kaiser MJ, Reynolds JD. *Marine fisheries ecology.* Oxford: Blackwell Science Ltd.; 2001.

[39] Olsen EM, Heino M, Lilly GR, Morgan MJ, Brattey J, Ernande B, et al. Maturation trends indicative of rapid evolution preceded the collapse of northern cod. *Nature* 2004; **428**: 932-35.

[40] Aiken KA. The biology, ecology, and bionomics of the triggerfishes, Balistidae. In: Munro JL, editor. *Caribbean coral reef fishery resources.* Penang: International Center for Living Aquatic Resources Management; 1983; p. 191-205.

[41] Kacem H, Boudaya L, Neifar L. Age, growth and longevity of the grey triggerfish, *Balistes capriscus* Gmelin, 1789 (Teleostei, Balistidae) in the Gulf of Gabès, Southern Tunisia, Mediterranean Sea. *J Mar Biol Assoc U K* 2015; doi: 10.1017/S0025315414002148.