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Reproductive histology of the saddled seabream *Oblada melanura* in the southern Mediterranean Sea (Tunisian coast)

by

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Abstract

The saddle seabream, *Oblada melanura*, is an important species in Mediterranean coastal fisheries. The artisanal gillnet fishery on Tunisia's northern coast was sampled on a monthly basis between April 2015 and March 2016. The sex ratio varied by month throughout the year, with females dominating from November to December and equal sex ratios during the rest of the year. For both sexes, gonad maturity and gonadosomatic index (I_G) indicated that spawning occurs from May to September. In terms of energy investment, fish feed throughout the year. During the spawning period (May to September), gonadal production (I_G) appears to be supported by liver reserves (I_H). These reserves came from external sources (i.e. food) in the period before spawning. During the spawning period, it was necessary for the fish to continue feeding in order to support their body development. Estimated lengths at 50% maturity were similar for females (L_{50} = 17.14 cm) and males (L_{50} = 17.01 cm).

Key words: biology, maturity, Sparidae, Tunisia

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1. Introduction

The saddled seabream *Oblada melanura* (Linnaeus 1758) is a commercially important coastal seabream (Bauchot & Hureau 1990; Harmelin-Vivien et al. 1995). It has a wide geographical distribution in the tropical and temperate regions of the eastern Atlantic Ocean, from the Gulf of Biscay to Angola (including the Canary Islands, the Cape Verde islands and São Tomé Island), and the Mediterranean Sea and the Black Sea (Bauchot, Hureau 1986; Fredj, Maurin 1987; Froese et al. 2014). It is widely distributed along the coast of Tunisia (Bourgeois & Farina 1961; Lubet, Azzouz 1969; Azzouz 1971; 1974; Bradai 2000).

O. melanura is fished commercially in the Mediterranean Sea using gear such as gillnets (Akyol et al. 2014) and trammel nets (Ali Bacha et al. 2017). It is also caught by recreational fishermen (Russell et al. 2014), and has been considered for aquaculture (Suquetet al. 2009). In Tunisia, the species is mainly targeted by coast-based fisheries. While the species fishing in the north is specific. In addition to gillnets, fishermen use trammel nets in turbid waters after bad weather, using wheat bran as bait to catch fish of this species.

According to the International Union for Conservation of Nature (IUCN) Red List, *O. melanura* is assessed as Least Concern (LC), but the need for further monitoring and re-assessment has been indicated, given the potential for local overfishing and competition with Lessepsian migrants (Russell et al. 2014).

Various aspects of the fisheries ecology of *O. melanura* in Tunisian waters have been previously investigated, including its distribution (Bourgeois & Farina 1961; Lubet & Azouz 1969; Azouz 1971; 1974; Bradai2000), stock identification (Barhoumi et al. 2017), feeding habits (Lassidi & Chakroun-Marzouk 2015; Khaldi et al. 2016), growth (Khaldi et al. 2016) and parasitology (Ben Abdallah & Maamouri 2008). The reproductive biology of the species in the study area is less researched, although it has been described from other locations in its range, including the eastern Adriatic Sea (Pallaoroet al. 1998; Tsikliraset al. 2010; Antolović et al. 2010; 2013), north-western Greece (Nikolioudakis et al. 2006), Sicilian coastal waters (Cavallaro et al. 1985) and Egyptian Mediterranean waters (El-Maghraby et al. 1981; Zaki et al. 1995; Daban et al. 2020).

Research on reproduction is a key element of aquaculture and is essential for genetic biotechnology research (Wootton & Smith 2014). In addition, information on reproduction is crucial for studies of population dynamics and life history (Hilborn & Walters 1992; Corgos & Freire 2006; Muchlisin 2014). Length or age at maturity are among the primary reproductive variables incorporated into models (Wootton & Smith 2014) and considered important for life history, demographic analysis and spawning stock biomass (SSB) estimation (Chen & Paloheimo 1994; Vitale et al. 2006). Of key importance in estimating SSB is the relationship between stock and recruitment (Murawski et al. 2001). Consequently, variability in the impact of SSB on assessment results and the accuracy of assessment projections affect fisheries management.

In addition, length at maturity is also used as a benchmark to establish a minimum catch (Corgos & Freire 2006) to ensure that fish are not captured before they reach maturity to sustain the stock. Length at maturity is estimated by fitting a logistic curve to length data and the proportion of the population that is mature. Nevertheless, the estimation of the probability of maturity is based on observations; fish must be physiologically mature and produce viable gametes (Restrepo & Watson 1991). However, the question arises about the biological criteria for identifying mature fish. Several methods have been used to identify mature individuals, including inspection of gonads, estimation of the gonadosomatic index and histological staining of tissue (Khoufi et al. 2014; Flores et al. 2015; Jaziri et al. 2015).

Oblada melanura is mainly a gillnet-fished species in the south-central Mediterranean Sea, where biological studies of this fish are limited. This paper aims to progress the biological understanding of this species following stock identification studies (Barhoumi et al. 2017). Our objective is to assess the reproductive biology of *O. melanura* from the south-central Mediterranean Sea by describing the sex ratio, spawning season, and length at maturity in the population of this species in northern Tunisian waters. Such data are necessary for future stock assessments and reporting on fisheries management in the area.

2. Materials and methods

2.1. Data collection

Monthly samples of *O. melanura* commercially caught in Rafraf, northern Tunisia, the southern Mediterranean Sea (Fig. 1), were examined over a period of one year (April 2015 to March 2016). The samples were collected using commercial gillnets deployed in waters with a depth of 30 m. In total, 345 males and 472 females were sampled, body weight measurements were recorded, and histological examinations of the gonads were performed.

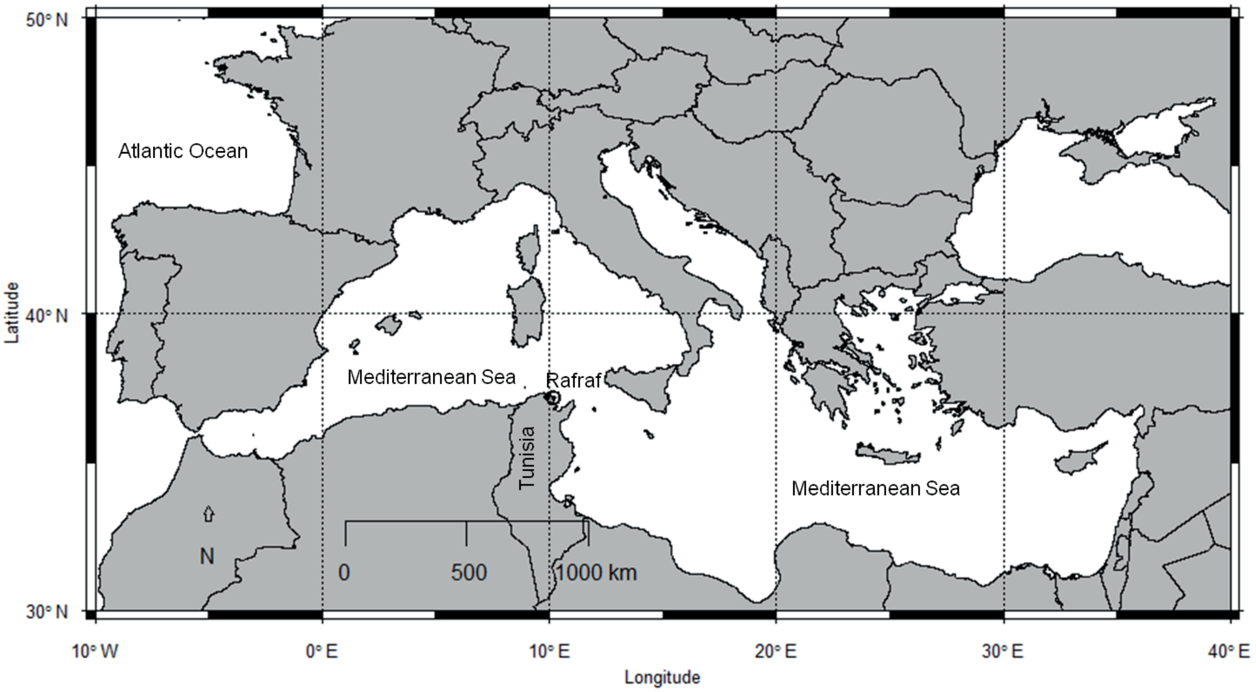


Figure 1

Map of the location where biological material of *Oblada melanura* was collected.

The weight measurements collected included total weight (W_T ± 0.1 g), eviscerated weight (W_E ± 0.1 g), as well as gonad weight (W_G ± 0.001 g) and liver weight (W_L ± 0.001 g).

The sex and maturity stages were determined for all individuals using microscopic analysis according to the standardized terminology proposed by Brown-Peterson et al. (2011) and modified by the authors of this paper (Table 1). The gonads were preserved in 10% buffered formaldehyde and then histologically processed using standard paraffin embedding and hematoxylin-eosin staining techniques.

2.2. Data analysis

The sex ratio (SR) was obtained by dividing the number of females by the total number of individuals. The behavior of males and females was analyzed by season using the χ^2 test.

The length–weight relationship (LWR), the condition factor (CF; Le Cren 1951), the gonadosomatic index (I_G) and the hepatosomatic index (I_H) were calculated using the following equations:

$$W_{TC} = aL_T{}^b$$

$$CF = \frac{W_T}{aL_T{}^b}$$

$$I_G = \frac{W_G}{W_E} \times 100$$

$$I_H = \frac{W_L}{W_E} \times 100$$

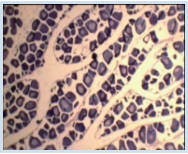
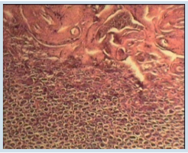
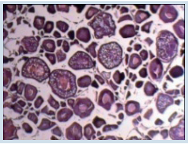
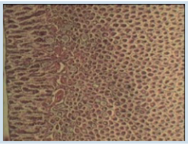
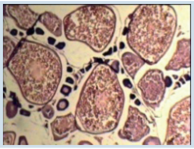
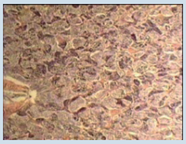
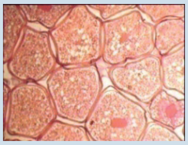
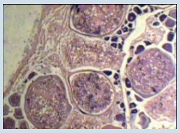
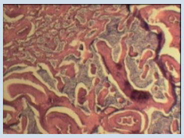
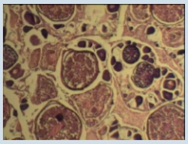
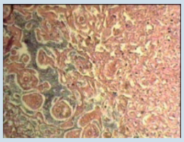
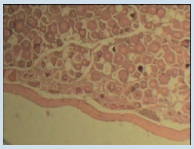
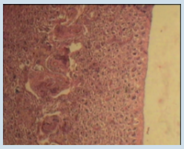
Where W_{TC} is the theoretical (calculated) total weight, L_T is the observed total length, a is the constant of LWR and b is the power function coefficient of LWR, W_T is the observed total weight, W_G is the gonad weight, W_L is the liver weight and W_E is the eviscerated weight.

For both sexes, maturity stages were described based on histological analysis. Physiologically, the spawning period was analyzed for both sexes based on different reproductive phases and reproductive indices. The reproductive modality of *O. melanura* was described, where reproductive indices were studied along with the estimation of the relationship between the condition factor and I_G and I_H using the Spearman correlation.

Physiologically, maturity ogives for both sexes were estimated during the spawning season and for

Table 1

Microscopic classification of *O. melanura* gonad maturity (Brown-Peterson et al. 2011; modified by the authors). (Sg1 = primary spermatogonia; Sg2 = secondary spermatogonia; Sc1 = primary spermatocyte; Sc2 = secondary spermatocyte; St = spermatid; Sz = spermatozoa).

Stage	Maturity classification and description	Photo (Female)	Photo (Male)
I	Juvenile (immature): Female: Only unyolked oocytes. Male: Only Sg1present; no lumen in lobules.		
II	In maturation: Female: Presence of cortical alveoli and vitellogenesis phases. Male: Spermatocystsvisible along lobules. Sg2, Sc1, Sc2, St, and Sz may be present in spermatocysts. Sz not present in lumen of lobules or in spermducts.		
	Pre-spawning: Female: Presence of all vitellogenesis phases. Nuclei migration phase, but post-ovulatory follicles are not present. Male: Only Sg1, Sg2, and Sc1.		
III	Spawning: Female: High percentage of hydrated oocytes at the beginning of the hydration process and/or the youngest post-ovulatory follicles (day 0), together with vitellogenic oocytes in different stages. Male: Sz in lumen of lobules and/or spermducts. All stages of spermatogenesis (Sg2, Sc, St, Sz) may be present. Spermatocysts throughout the testis; active spermatogenesis.		
IV	Post-spawning: Female: Older post-ovulatory follicles (day 1 and day 2) and vitellogenic oocytes in different stages for the next batch. No hydrated oocytes. Male: Sz in lumen of lobules and/or spermducts.		
V	Regression: Female: Few or absent vitellogenic oocytes and no post-ovulatory follicles younger than 72 h, with a large number of blood vessels, ovarian wall swelling, atresia, disorganization of ovary structures. Male: Residual Sz present in lumen of lobules and in spermducts. Widely scattered spermatocysts near the periphery containing Sc2, St, Sz.		
	Regeneration: Female: Only oogonia present. Muscle bundles, enlarged blood vessels, thick ovarian wall and/or gamma/delta atresia or old, degenerating post-ovulatory follicles may be present. Male: No spermatocysts. Lumen of lobule often absent. Proliferation of spermatogonia throughout testes. Small amount of residual Sz occasionally present in lumen of lobules and in spermducts.		

the entire year. The comparison between the two was carried out using generalized linear models. Physiological maturity by sex was also estimated.

3. Results

3.1. Size, sex ratio and length–weight relationship

Table 2 shows the range of length and weight of all sampled individuals of *O. melanura* of both sexes. During the study period, the sex ratio showed monthly variation, ranging from 48% to 78% for females (Table 2). The proportion of females was significantly higher between November ($\chi^2 = 3.971 > 3.841$) and December ($\chi^2 = 9.103 > 3.841$), with more equal sex ratios at other times of the year (Table 2). For both sexes, there was a strong correlation between length and weight. Table 2 shows $W_T = 0.017{*}L_T^{2.881}$ ($R^2 = 0.981$, $p < 2.2e\text{-}16$) for females and $W_T = 0.017{*}L_T^{2.874}$ ($R^2 = 0.980$, $p < 2.2e\text{-}16$) for males.

Table 2

Sex ratio, length and weight range of females (n = 472) and males (n = 345) of *Oblada melanura* from Tunisian waters. Length and weight values are mean ± SD, with the range in parentheses; SR is the sex ratio (derived as the number of females to the total number of individuals).

	Female	Male
Total length (L_T, cm)	21.22 ± 3.73 (13.20–30.20)	21.25 ± 3.80 (10.90–29.50)
Total weight (W_T, g)	123.0 ± 62.7 (27.0–324.0)	123.0 ± 59.1 (13.0–298.0)
Eviscerated weight (W_E, g)	114.2 ± 59.3 (24.0–304.0)	114.2 ± 54.7 (12.0–282.0)
Length–weight relationship	WT = 0.017 $LT^{2.881}$	WT = 0.017 $LT^{2.874}$

Month	April	August	December	February	January	July	June	March	May	November	October	September
SR	0.537	0.447	0.784	0.569	0.551	0.483	0.485	0.560	0.556	0.759	0.667	0.688

3.2. Spawning period

For both sexes, the monthly occurrence of different reproductive phases was paralleled by the presence of active spawning individuals with hydrated oocytes and/or post-ovulatory follicles (Fig. 2 A) and spermatozoa and sperm (Fig. 2 B) from May to September. For females, the highest proportion of spawners was observed in June (17%), and for males – in May (30%). The I_G increased from May (0.92 for females and 1.2 for males), reaching the maximum in June (1.08 for females and 1.30 for males), decreasing slowly from July (0.83 for females and 0.93 for males) to January (0.44 for females and 0.34 for males). Considering the monthly changes in I_G and the maturity stage for both sexes, the spawning period corresponds to the period from May to September.

For both sexes, the reproductive indices (I_H and I_G) showed lower values from September to February (Figure 3) and began to slowly increase for I_G and

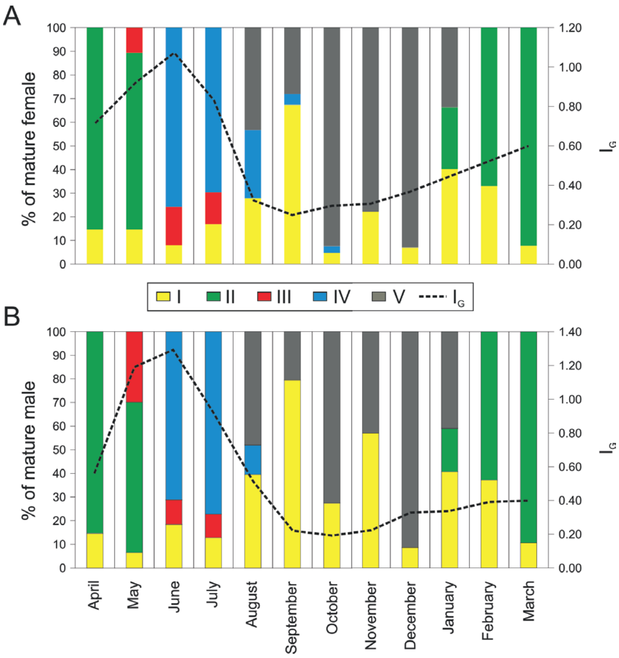


Figure 2

Monthly distribution of spawning stages with *Oblada melanura* IG evolution: (A) for females and (B) for males. IG – gonadosomatic index. For maturity stages, see Table 1.

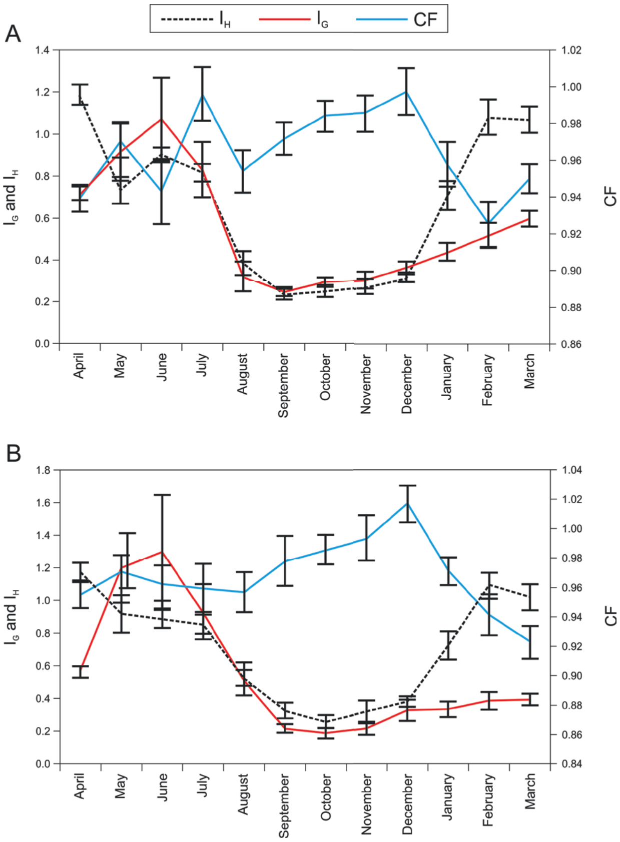


Figure 3

Evolution of somatic and reproductive indices for *Oblada melanura*, with standard deviation: (A) for females and (B) for males. CF – condition factor; IH – hepatosomatic index; IG – gonadosomatic index.

Table 3

Relationship between the condition factor and reproductive indices of *Oblada melanura* in northern Tunisia. CF – condition factor; IH – hepatosomatic index; IG – Gonadosomatic index; p* < 0.05 (significant).

	Relationship	N	R (spearman)	t(N-2)	*p*
Female	CF and I_H	12	–0.720	–3.284	0.008*
	CF and I_G	12	–0.385	–1.318	0.217
	I_H and I_G	12	0.790	4.078	0.002*
Male	CF and I_H	12	–0.825	–4.619	0.001*
	CF and I_G	12	–0.566	–2.174	0.055
	I_H and I_G	12	0.706	3.155	0.010*

abruptly for I_H at the end of the period (January and February). The indices increased to reach their maxima in April for I_H and in June for I_G. In contrast, CF showed similar trends for both sexes, peaking in July and December for females (Fig. 3 A) and in May and December for males (Fig. 3 B). For both sexes, there was a positive correlation between I_H and I_G and no significant correlation between CF and I_G, in contrast to a significant correlation between CF and I_H (Table 3).

3.3. Ogive maturity

The maturity ogives obtained for May to September (spawning period) and for the whole year (Fig. 4) were compared with the analysis of generalized linear models and showed no significant differences ($p > 0.05$) between the two datasets (Table 4; Fig. 4). The estimated L_{50} during the spawning period (16.95 cm) was very similar to that observed for the whole year (17.15 cm).

The comparison of maturity ogives for *O. melanura* by sex showed similar values for females (L_{50} = 17.14 cm) and males (L_{50} = 17.01 cm; Fig. 5). The GLM analysis showed no significant differences between the sexes ($p > 0.05$; Table 4).

Table 4

Results of generalized linear models (GLM).

	Deviance	AIC	*p*
TL	547.17	553.17	< 0.001
Period of the year	285.68	291.68	0.76
TL	223.847	229.847	< 0.001
Sex	90.581	96.581	0.58

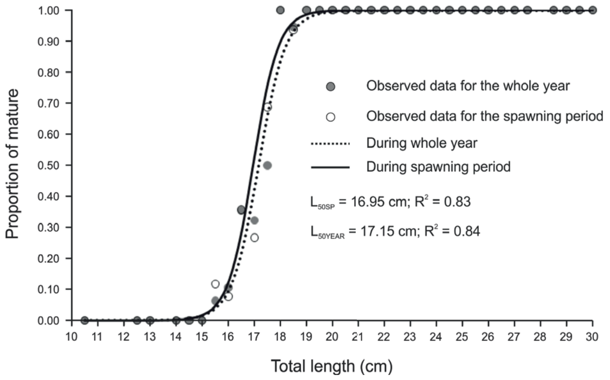


Figure 4

Estimation of maturity ogives for combined sexes of *Oblada melanura* from the Tunisian coast during the spawning period (May to September) and throughout the year. L_{50} – length at first maturity; R^2 – determination coefficient; SP – during the spawning period; YEAR: throughout the year; L_T – total length.

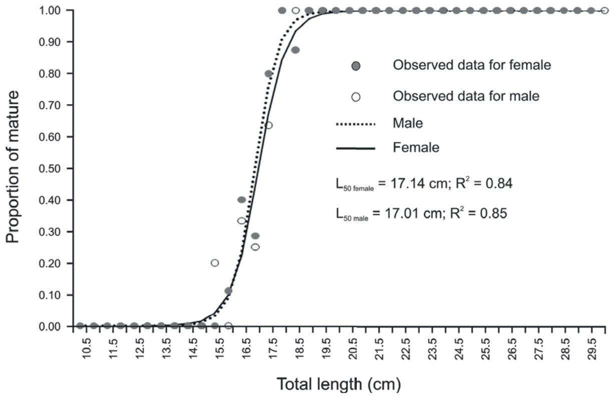


Figure 5

Estimation of maturity ogives during the spawning period (May–September) of *Oblada melanura* from the Tunisian coast for each sex. L_T – total length.

4. Discussion

According to the first analysis, the total length of *O. melanura* samples ranged from 10.9 cm to 30.2 cm. Compared to other studies, the recorded ranges were similar to those reported from the Atlantic Ocean and Mediterranean Sea, with some differences that depend on the gear used to catch fish of this species. For the smallest individuals, a beach seine (mesh size = 4 mm) was suitable to catch individuals between 2 and 6.5 cm (Pallaoroet al. 1998). For the largest individuals, it appears that *O. melanura* can reach a length of more than 30 cm in the southern Aegean Sea (Akyol et al. 2014), reaching 36.6 cm on the French Catalan coast (Crec'hriou et al. 2013), but the average length for *Oblada melanura* is 20 cm (Bauchot & Hureau 1990). Such large individuals can be explained by the presence of the species near the aquaculture zone, where the habitat is rich in nutrients (Akyol et al. 2014).

For some species, or even the same population at different times, the sex ratio may vary (Oliveira et al. 2012) due to several factors. In fact, the relationship between the sex ratio and the total length appears to vary geographically. Along the northern coast of Tunisia, both females and males reached large length classes (up to 30 cm), but in the eastern Adriatic Sea, males of *O. melanura* dominated in smaller length classes and females dominated in larger length classes (Pallaoroet al. 1998), while in the northern Adriatic Sea, females dominated in smaller length classes and males in larger length classes, although the largest individuals were females (Pallaoro 1996), similar to what was observed in coastal waters of Sicily (Cavallaro et al. 1985). In our case, this may be monomorphism, while in other mentioned regions– dimorphism. In fact, sexual size dimorphism varies among species and taxa due to differences in the relative intensity of selective forces. These forces are fecundity for females and a high degree of territoriality, sperm competition and/or paternal care for males (Horne et al. 2020). The case of the species on the Tunisian coast could be the result of a balance between these forces for both sexes.

According to the somatic condition (length, weight, and CF), *O. melanura* did not show any difference in evolution between the sexes, however, the reproductive indices such as I_G and I_H were different for males and females. Considering the factor of time, all these parameters were different during the study period.

Considering the physiological state, monthly changes in I_G indicate an active gonad from May, peaking in June and decreasing from July to September. For both sexes of *O. melanura*, the spawning period lasted from May to September. Compared to different areas in the Mediterranean Sea, this period is slightly longer, as observed in other studies (Table 9). The spawning period along the Algerian coast lasted from April to June (Breder & Rosen 1966), from March to June at Benghazi (Rafalah & El-Mor 2018), from April to June (El-Maghraby et al. 1982) and from May to July off the coast of Egypt (Zaki et al. 1995) and Syria (Ali Bacha et al. 2017), as well as the North Aegean Sea (Daban et al. 2020), and from June to July in the Adriatic Sea (Cetinić et al. 2002). These small differences may be related to several environmental factors, such as temperature and food availability.

To understand the reproductive strategy in terms of energy investment, seasonal variation in somatic and gonadal condition was studied. Monthly variation in body condition (CF) revealed an increase in body size in terms of weight and length throughout the year, with low condition from February to April compared to the period from May to January, thus confirming the foraging of this species throughout the year. While food supply depends on environmental conditions, *O. melanura* has been reported to be an opportunistic predator (Pallaoroet al. 2003). For the same periods, the gonadal production (I_G) appears not to be affected by somatic condition (CF), but is negatively correlated with liver reserves (I_H). When gonadal production increases, the liver mass decreases, indicating the use of liver reserves to support gonadal growth. The acquisition of lipid reserves comes from external sources of energy (i.e. food), in addition to muscle reserves, which explains the decrease in CF when I_H increases from January to April, i.e. the period preceding the preparation of animals for the reproductive season. From May, the CF begins to increase until December, preparing to maintain the body development. As on the Syrian coast, the CF appears not to affect the I_G and the I_H plays a major role in I_G (Ali Pacha et al. 2017). However, several environmental (such as temperature and food availability) and biological factors affect the reproductive behavior and traits of fish populations

Table 5

Summary of reproductive parameters of *Oblada melanura* from the Mediterranean Sea.

Area (country)	Spawning period	L_{50} (cm) (female–male)	Reference
Algerian coast	April–June		Breder, Rosen, 1966
Benghazi (Libya)	March–June	(18.6–17.5)	Rafalah, El-Mor, 2018
Egypt	April–June		El-Maghraby et al. 1982
Egypt	May–July		Zaki et al. 1995
Tartous (Syria)	May–July	(17.3–18.6)	Ali Bacha et al. 2017
North Aegean Sea	May–July	(18.83–18.97)	Daban et al. 2020
Adriatic Sea	June–July	(17.5–16.64)	Cetinić et al. 2002
Rafraf (Tunisia)	May–September	(17.14–17.01)	present work

(Recasenset al. 1998; Murua 2006; Domínguez-Petit & Saborido-Rey 2010).

Reproductive parameters, mainly length at first maturity (ogive), play a fundamental role in determining the reproductive potential and spawning pattern of fish species (Coleman et al. 1996; Trippel 1999), and together with growth parameters and natural mortality, determine population dynamics and are relevant to stock assessment and management. The ogive was determined using maturity data determined by microscopic observations. The first results showed no differences between the sexes, even when using the whole year or the spawning period. In our study, the length at first maturity L_{50} was 17.14 cm and 17.01 cm for females and males, respectively, and these results were similar to other Mediterranean areas (Table 5). In Benghazi, the L_{50} was 18.6 cm for females and 17.5 cm for males (Rafalah & El-Mor 2018), in Syrian waters, the L_{50} was 17.3 cm for females and 18.6 cm for males (Ali Pacha et al. 2017), and in the Adriatic Sea, the L_{50} was 17.5 cm for females and 16.64 cm for males (Cetinićet al. 2002).

Both sexes of the species inhabit the same area throughout the year. They are exposed to the same environmental conditions, especially in terms of trophic resources, which equally affect their life history traits. By comparison, in most marine species, the two sexes regroup mainly during the spawning period.

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