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REPRODUCTIVE BIOLOGY OF THE SPOTTED FLOUNDER, *CITHARUS LINGUATULA* (ACTINOPTERYGII: PLEURONECTIFORMES: CITHARIDAE), FROM SAROS BAY (NORTHERN AEGEAN SEA, TURKEY)

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Background. Understanding the reproduction of the spotted flounder, *Citharus linguatula* (Linnaeus, 1758), has been considered a major step toward understanding its population dynamics, especially, in the northern Aegean Sea. So, the presently reported study aimed to study the reproductive biology of *C. linguatula* and provided the first data of its fecundity.

Materials and methods. This study was carried out between September 2006 and September 2008 in Saros Bay (Northern Aegean Sea, Turkey), which had been closed to trawl fishing since 2000. The gonadosomatic index, length at maturity (L_{50}), and absolute fecundity were analysed.

Results. The elevated GSI values observed, suggested the spawning period was from September to November. The length at maturity (L_{50}) was at 15.0 cm TL for females and 14.0 TL cm for males**.** The relations between the absolute fecundity and total length, total weight, and age of the spotted flounder females were estimated as $F_a = 0.1878\text{TL}^{3.29}$, $F_a = 58.515\text{TW} + 36.453$, and $F_a = 887.59\text{A} - 389.43$, respectively.

Conclusion. The identification of reproductive strategy is necessary to obtain a better understanding of its biology.

Keywords: spotted flounder, reproduction, Saros Bay, *Citharus linguatula*, fecundity, length at maturity

INTRODUCTION

The spawning period has long been a principal issue in fisheries biology, ecology, and management because of its importance for the recruitment (Beaugrand et al. 2003), survival (Garvey et al. 2002), and stock biomass, and thus the fishery yield (Kjesbu and Witthames 2007). Therefore the knowledge of fecundity is useful in investigating the population dynamics of a fish species and for fish culture purposes (Dulčić et al. 1998). Consequently, the reproductive studies may be used for quantification of the reproductive capacity of fish (Murua et al. 2003).

The spotted flounder, *Citharus linguatula* (Linnaeus, 1758), is a flatfish that is distributed in the Mediterranean Sea and eastern Atlantic (Nielsen 1986). It generally occurs at depths from 10 to 100 m (Sartor et al. 2002). This species is widely distributed in the Mediterranean, including the Aegean and Marmara seas and the territorial waters of Turkey (Bilecenoglu et al. 2002). It is often discarded from commercial trawl fisheries in Turkish waters (Bayhan et al. 2009).

A number of studies on *Citharus linguatula* is available as a reference. The age, growth, and reproduction of this fish from Greek waters were studied by Vassilopoulou and Papaconstantinou (1994), while the same aspects were investigated by García-Rodríguez and Esteban (2000) and Teixeira et al. (2010) from the Iberian Mediterranean and the Portuguese coast, respectively. The diets of spotted flounder from the eastern coast of Spain and the central Tyrrhenian Sea were studied by Redon et al. (1994) and Carpentieri et al. (2010), respectively. Sartor et al. 2002 provided data on the abundance and the distribution *C. linguatula* in the Mediterranean Sea.

As for Turkish waters, the information on age, growth, and reproduction of the species come from Edremit Bay (Türker Çakır et al. 2005) and Izmir Bay (Bayhan et al. 2009, Kınacıgil et al. unpublished**, Ulutürk unpublished***).

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** Kınacıgil H.T., İlkyaz A.T., Metin G., Ulaş A., Soykan O., Akyol O., Gurbet R. 2008. Balıkçılık Yönetimi Açısından Ege Denizi Demersal Balık Stoklarının İlk Ürüme Boyları, Yaşları ve Büyüme Parametrelerinin Tespiti. [Determination of the first maturity length/age and growth parameters of demersal fish stock in the Aegean Sea from the fishery management point of view.] The Scientific and Technological Research Council of Turkey (TUBITAK), Project number: 103Y132. Ege University Faculty of Fisheries, Bornova, Izmir. [In Turkish.]

*** Ulutürk E. 2007. İzmir Körfezi'nde dağılım gösteren kancaağız pisi balığı (*Citharus linguatula* Linnaeus, 1758)'nın bazı biyolojik özellikleri. [Some biological features of spotted flounder *Citharus linguatula* (L., 1758) from Izmir Bay.] MSc Thesis, Ege University Faculty of Fisheries, Izmir. [In Turkish.]

Gürkan and Bayhan (2009) reported some morphometric features of the spotted flounder in Izmir Bay. While Cengiz et al. (2011) documented its presence in Saros Bay, Cengiz et al. (2012a) examined the relation between total length and otolith size of *C. linguatula* in same region.

No information concerning the reproduction of *C. linguatula* was so far available in from the Northern Aegean Sea. The objectives of this study were to estimate the gonadosomatic index (GSI), the length at maturity (L_{50}), and the relations between the absolute fecundity and total length, absolute fecundity and total weight, as well as absolute fecundity and age of *C. linguatula* from Saros Bay and to compare these results with those of the previous studies in other areas.

MATERIALS AND METHODS

The samples of the spotted flounder, *Citharus linguatula*, were collected monthly, between September 2006 and September 2008 using a commercial bottom trawl net of 44 mm codend stretched mesh size at depths ranging from 0 m to 500 m in Saros Bay, which had been closed to trawl fishing since 2000 (Fig. 1).

The specimens were measured to the nearest 1 mm (total length, TL), weighed to the nearest 0.01 g (total weight). The chi-square (χ^2) test was used to detect the differences in the sex ratio. The Student's *t*-test was used to analyze the differences in the mean length and weight of the sexes.

In flatfishes, the otoliths from blind side were used for age estimation, as the nucleus is more central (Fig. 2) and the zones were easier to be interpreted compared to otoliths from the ocular side (Cengiz et al. 2012a, 2012b).

Following removal, the blind side otoliths were first soaked in 5% HCL and 3% NaOH solutions, respectively, and washed in distilled water and subsequently dried. The otoliths, placed in watch glass filled with the water, were read using a stereoscopic zoom microscope under reflected light against a black background. Opaque and transparent zones were counted; one opaque zone plus one transparent zone was assumed to be one year (Cengiz et al. 2013).

Sex was determined by examining the gonads macroscopically. The maturity was assessed according to Gunderson's (1993) scale: stage I (immature), stage II (resting), stage III (developing), stage IV (ripe), and stage V (spent). The spawning period was estimated by monthly analyses, the changes of the gonadosomatic index (GSI) using the equation:

$$\mathrm{GSI} = [W_{\mathrm{g}} \times (W - W_{\mathrm{g}})^{-1}] \times 100$$

where W_{g} is the gonad weight [g] and W is the total weight [g] of fish (Ricker 1975).

Length at maturity (L_{50}), the size at which 50% of the individuals were mature, was estimated by means of

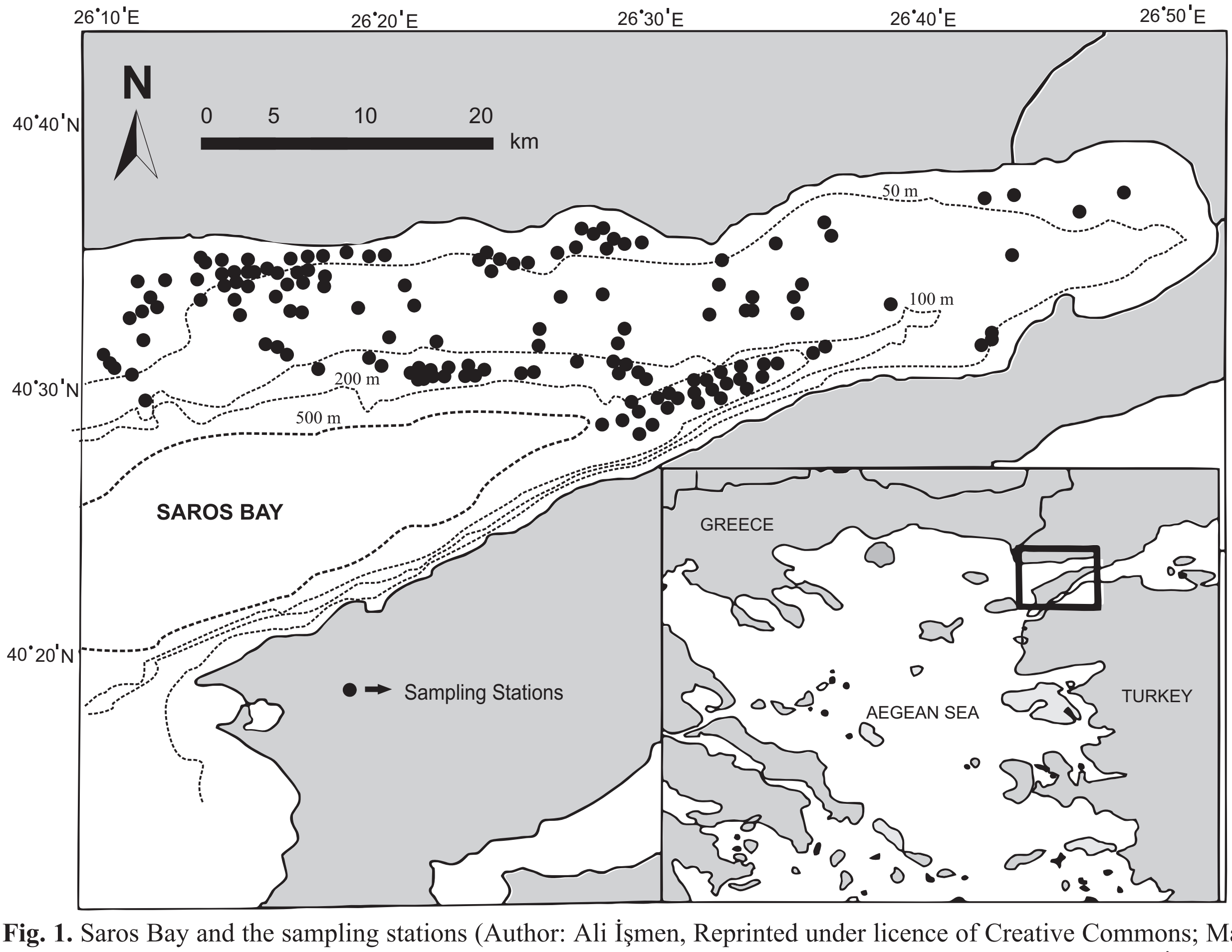

Fig. 1. Saros Bay and the sampling stations (Author: Ali İşmen, Reprinted under licence of Creative Commons; Map previously published in the following papers Cengiz et al. 2011, 2012a, 2012b, 2013, Ismen et al. 2013, İşmen et al. 2013, Yıgın and Ismen 2010, Yigin and Ismen 2012, 2013)

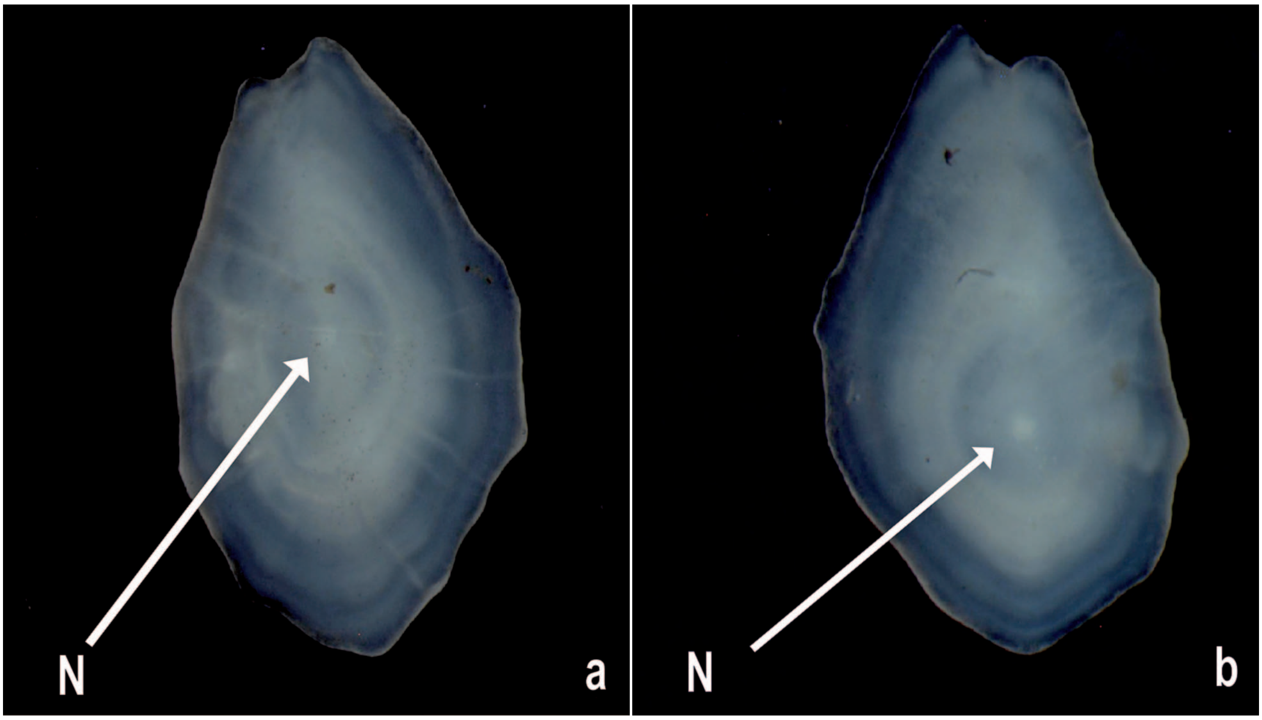

Fig. 2. Otolith of spotted flounder, *Citharus linguatula*, from Saros Bay, with N indicating the nucleus; otolith from blind side (**a**), otolith from ocular side of the fish (**b**)

a logistic function that was fitted to the proportion of sexually mature individuals by size class using a nonlinear regression (King 1995).

Gravimetric method was used for fecundity estimates (Bagenal and Braum 1978). In order to calculate fecundity, the ovaries of mature females (prior to the reproductive period) were weighed the nearest 0.0001 g (total weight), three sub-samples were taken from the front, mid- and rear sections of each ovary, weighed and then immersed separately in Gilson's fluid. These ovaries were frequently shaken to ensure the separation of oocytes from ovarian tissues. All oocytes were counted directly under stereoscopic zoom microscope. The total number of eggs in each ovary sub-sample was then estimated by using the equation provided by Yeldan and Avşar (2000):

$$F_1 = (W_g \times N) \times W_s^{-1}$$

where: F_1 is the total number of eggs in ovary sub-sample 1, W_g is the gonad weight, N is the number of eggs in the sub-sample, and W_s is sub-sample weight. Later, by taking the mean number of three sub-sample fecundities (F_1, F_2, and F_3), the absolute fecundity (F_a) for each female fish was estimated as:

$$F_a = (F_1 + F_2 + F_3)^{-3}$$

Hereby, the relations between absolute fecundity (F_a) and total length (TL), absolute fecundity (F_a) and total weight (TW), as well as absolute fecundity (F_a) and age (A) were estimated as (respectively):

$$F_a = a\mathrm{TL}^b$$

$$F_a = a + b\mathrm{TW}$$

$$F_a = a + bA$$

where a and b are the parameters of the equation (Avşar 2005).

RESULTS

Of 659 examined specimens of *Citharus linguatula*, 464 (70.4%) were females, and 195 (29.6%) males. The sex ratio (F : M) was 1 : 0.42, which is significantly different from equal representation of sexes (χ^2 test: $P < 0.05$). The mean ± standard error (and range) of total length were: 18.2 ± 0.10 (9.9–23.5) cm TL for females, 16.6 ± 0.15 (9.4–22.2) cm TL for males (Fig. 3). The respective values for total weight were: 47.59 ± 0.80 (7.81–102.42) g for females and 35.64 ± 1.03 (5.08–91.92) g for males. The Student's t-test showed significant differences between the mean lengths and between weights of the both sexes (all $P < 0.05$).

The variations in GSI values throughout the study period did not show strong changes for males, but presented a pronounced peak for females in September. All year

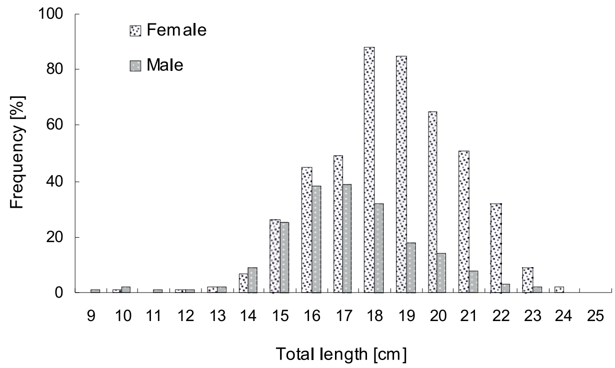

Fig. 3. The length-frequency distribution for females and males of spotted flounder, *Citharus linguatula*, from Saros Bay

around the GSI values for females were higher than the male values. GSI reached the highest values with 3.63 and 0.52 for females and males, respectively, in September 2007. The elevated GSI values suggested the spawning period was from September till the end of October. After spawning, the gonadosomatic index remained almost constant for most of the year (Fig. 4).

In presently reported study, the smallest mature female and male were found to be 9.9 and 9.4 cm TL, respectively. The length at maturity (L_{50}) was at 15.0 cm TL for females and 14.0 cm TL for males (Fig. 5).

The absolute fecundity (F_a) was estimated for 81 ripe females (age 2 to 6) caught in June–September 2008. The maximum value of 6262 eggs was recorded in a 6 year-old fish weighing 95.80 g (23.5 cm TL) while the minimum value of 1071 eggs for a 2 year-old fish weighing 19.59 g (15.2 cm TL). The mean value ± standard error of absolute fecundity was 3205 ± 129. The relations between the absolute fecundity and total length, total weight and age of the spotted flounder females were estimated as $F_a = 0.1878TL^{3.29}$, $F_a = 58.515TW + 36.453$, and $F_a = 887.59A - 389.43$, respectively (Fig. 6).

DISCUSSION

The reproductive biology of *Citharus linguatula* has been investigated for the first time in the Saros Bay (northern Aegean Sea, Turkey). The reproductive cycles of female and male individuals are synchronized. In this study, the spawning period of the spotted flounder started in September and continued until end of October. Sabatés (1988) and Ulutürk (unpublished*) reported that the reproductive period was between August and November for the Western Mediterranean and Izmir Bay. Another study conducted in Izmir Bay (Aegean Sea) by Kınacıgil et al. (unpublished*) indicated that the reproductive period was from October to December (Table 1). The spawning period has a close relation to the ecological characteristics of the water system in which the species live (İlkyaz et al. 2010) and are attributed to the factors such as stagnant or running water, temperature, and quality of food (Nikolsky 1963). According to Wootton (1990), temperature appears to be the most important factor among those that may influence the reproduction of fishes.

Our results showed that the length at maturity (L_{50}) was at 15.0 cm TL for females and 14.0 cm TL for males. Kınacıgil et al. (unpublished*) determined the length at

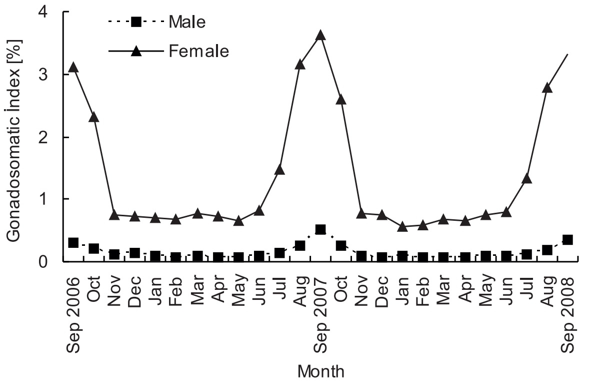

Fig. 4. The mean gonadosomatic index (%) values for females and males of the spotted flounder, *Citharus linguatula*, from Saros Bay

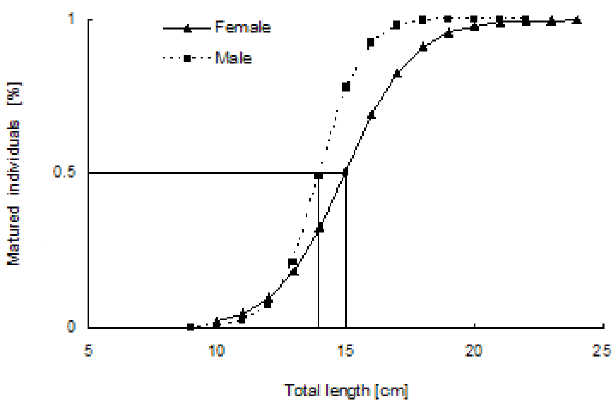

Fig. 5. Length at maturity (L_{50}) for females and males of spotted flounder, *Citharus linguatula*, from Saros Bay

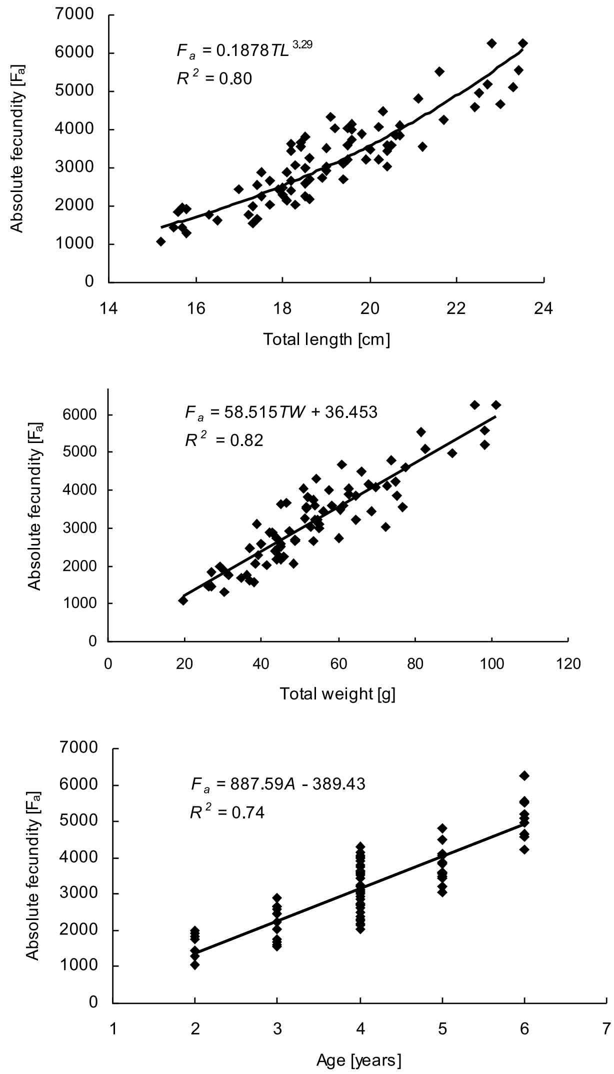

Fig. 6. The relations between the absolute fecundity and total length, total weight and age of females of spotted flounder, *Citharus linguatula*, from Saros Bay

* See footnote on page 123.

maturity (L_{50}) as 12.0 cm for females and 12.9 cm for males, whereas, Vassilopoulou and Papaconstantinou (1994) estimated the length at maturity (L_{50}) to be 15.1 cm for females and 12.4 cm for males (Table 2).

The L_{50} for both female and male appears to increase with the latitude. This regularity is related to the pattern of overall body size in flatfish species being larger at higher latitudes (Pauly 1994a). Rijnsdorp (1989) and Sampson and Al-Jufaily (1999) stated that L_{50} increased with the latitude, however, Walsh (1994) interpreted this relation as L_{50} declining with the temperature increase.

For all flatfishes studied to date, females grow larger and faster than the males (Pauly 1994a, Vassilopoulou and Ondrias 1999). This phenomenon is a common feature in many pleuronectiforms (Lozán 1992, Landa and Pinerio 2000, Cengiz et al. 2013). This could be attributed to differences in metabolism between females and males, such as differences in oxygen consumption (Pauly 1994a, 1994b), differences in the level of surplus energy between reproduction and somatic growth (Rijnsdorp and Ibelings1989), and differential food ingestion (Lozán 1992). Vassilopoulou and Papaconstantinou (1994) and Ulutürk (unpublished*) also found out that the natural mortality was higher for males than for females for *C. linguatula.*

In addition, the difference in growth rates between males and females seems to occur at the onset of maturity (Vassilopoulou and Papaconstantinou 1994). Males spawn for the first time earlier than females, as was also reported by Vassilopoulou and Papaconstantinou (1994) and by the presently reported study. Similar findings on males have been made many other flatfishes such as lemon sole, *Microstomus kitt* (Walbaum, 1792) (see Rae 1965); American plaice, *Hippoglossoides platessoides* (Fabricius, 1780) (see Roff 1983); turbot, *Psetta maxima* (Linnaeus, 1758); common sole, *Solea vulgaris* (Linnaeus, 1758); common dab *Limanda limanda* (Linnaeus, 1758); Mediterranean scaldfish, *Arnoglossus laterna* (Walbaum, 1792); and Thor's scaldfish, *Arnoglossus thori* Kyle, 1913 (see Deniel 1990). Roff (1982) insisted that male flatfish having reached the length at which they could successfully reproduce, decreased their growth, in part, as a response to the divergence of energy into reproduction and as a response to decreased foraging activity which reduced the risks of being preyed upon. Also Roff (1983) suggested that there was a clear association between the relative size of male fish and their reproductive behaviour—males that did not hold territories and did not care for their progeny were consistently smaller than females.

The fecundity results, provided by us, represent the first such data on *C. linguatula*. The marked differences in fecundity among species often reflect different reproductive strategies (Murua and Saborido-Rey 2003). Within a given species, fecundity may vary as a result of different adaptations to environmental habitats (Witthames et al. 1995). Even within a stock, fecundity is known to vary annually, undergo long-term changes (Rijndorp 1991) and has been shown to be proportional to fish size and condition. For a given size, females in better condition exhibit higher fecundity (Kjesbu et al. 1991). Fish size and condition are, thus, key parameters to properly assess fecundity at the population level. However, the fecundity–size relations has been used principally as a rapid means of predicting the fecundity of fish (Dulčić et al. 1998).

As a consequence, the data of this study will increase the life history data available for *C. linguatula* by providing the scientific support required so as to identify the cur-

Table 1

Spawning period of spotted flounder, *Citharus linguatula*, from different areas

Area	Spawning period	Reference
Western Mediterranean	August–November	Sabatés 1988
Izmir Bay, Turkey	August–November	Ulutürk unpublished*
Izmir Bay, Turkey	October–December	Kınacıgil et al. unpublished*
Portuguese coast	Autumn	Teixeira et al. 2010
Saros Bay, Turkey	September–November	This study

Table 2

Length at maturity (L_{50}) of spotted flounder, *Citharus linguatula*, from different areas

Area	Sex	Length at maturity	Reference
Northern Aegean Sea, Greece	♀	15.1 cm	Vassilopoulou and Papaconstantinou 1994
	♂	12.4 cm	
Izmir Bay, Turkey	♀	12.0 cm	Kınacıgil et al. unpublished*
	♂	12.9 cm	
Saros Bay, Turkey	♀	15.0 cm	This study
	♂	14.0 cm	

* See footnote on page 123.

rent stock state and will help conservation and management of the species.

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