

## Reproductive biology of the common cuttlefish (*Sepia officinalis* Linnaeus, 1758) in Iskenderun Bay (Northeastern Mediterranean Sea)

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Morphometric characters and reproductive biology were studied in *S. officinalis* collected at different seasons from the Iskenderun Bay, Turkey. A total of 2006 specimens (992♀, 1014♂) were sampled from September 2005 to August using a Mediterranean type deep trawl (22 mm mesh size). Mean mantle lengths for female and male were 9.83±1.66, 9.40±1.76 cm and the mean weights were 135.78±6.71g, 115.34±6.29g, respectively. Sexual maturity lengths were calculated as 12.04 for females and 10.30 cm for males. Mean diameter of eggs was measured as 5.367±9.12 mm per spawning period. Spermatophore lengths were estimated to range from 2.8±1.05 to 12.4±2.11mm. Population consisted of gonochoristic individuals. Although, sex ratio of *Sepia officinalis* were not significantly different from the expected 1:1 ratio (p>0.05), a slightly male biased sex ratio was recorded.

[**Keywords:** *Sepia officinalis*, Levantine Sea, sex ratio, sexual maturity.]

### Introduction

The common cuttlefish, *Sepia officinalis* L., 1758 is commonly found in the eastern Atlantic, from the North Sea to the Cape of Good Hope, through the English Channel and the eastern coast of Africa to Mozambique<sup>1</sup>. It is also widely distributed in the Mediterranean Sea, up to a depth of 200 m and where it has a commercial value in countries of the Mediterranean, Aegean and Marmara Sea cost line<sup>2,3</sup>. *S. officinalis* is the most widely-available cuttlefish species in Turkey. The contribution of this species to local fisheries differs in each sea from a the total of 1163,3 to 841,6 tons in the Mediterranean Sea, to 321,4 tons in the Aegean Sea, and 0,3 tons in the Marmara Sea<sup>4</sup>.

Although cuttlefish can exhibit variations in the life cycle<sup>5, 6, 7, 8</sup>, they live for approximately two years, and exhibits mass mortality of adults following a spring spawning period<sup>9</sup>. Growth, reproduction and life span of *S. officinalis* was studied by Forsythe et al<sup>10</sup>. Authors were successfully cultured *S. officinalis* under laboratory conditions. Gauvrit et al<sup>11</sup>, studied on the wild populations of *S. officinalis* and concluded that this species has a very long spawning period. A comprehensive study was performed on

fecundity of *S. officinalis* by Laptikhovsky et al<sup>12</sup>. Laptikhovsky et al<sup>12</sup>, suggested that intermittent spawning is a natural process for wild captured *S. officinalis* specimens. Fecundity estimates for cuttlefish involve many difficulties, such as determining which ovarian eggs to count, judging the appropriate times in the breeding cycle to take samples, and estimating the number of batches that spawned. Because of these difficulties, there was a few study published on the fecundity of cuttlefish<sup>13</sup>. Because of various egg sizes and stages have prolonged spawning period of cuttlefish which have advanced maturing (developing) eggs may be produced and others (less developed ones) have the time to develop. Therefore egg counts at only one time can be very misleading<sup>14</sup>.

There are four discrete populations of *S. officinalis* in Turkish coastal waters. In the morphometric analysis, only the Marmara Sea and Iskenderun Bay samples were differentiated from each other, and the Izmir and Antalya Bay samples overlapped each other<sup>15</sup>. There is a few detailed information on the reproductive biology of *S. officinalis* in the Aegean Sea<sup>16,17</sup> and also Northeastern Mediterranean Sea<sup>18</sup>. Present study consists the reproductive biology and

some morphometric aspect of *S. officinalis* in Levantine Sea (northeastern Mediterranean Sea), based on specimens from the Iskenderun Bay.

### Materials and Methods

By-catches of cuttlefish taken were obtained from a commercial fishing vessel (equipped with Mediterranean type deep trawl) operating in the Iskenderun Bay, during September 2005 and August 2006 (Figure. 1). A total of 2006 specimens (992♀, 1014♂) of *Sepia officinalis* were collected. After capture, the specimens were kept frozen until being analysed in the laboratory. Individuals were weighed and measured. Data on reproductive components were collected from cephalopods received in a suitable condition by weighing the following components (to 0.001 g): male testis and spermatophoric complex; female nidamental gland, ovary, oviducal complex (gland+oviducts) and proximal oviduct. Paired organs were weighed and recorded. These data were used to determine the maturity indices. Maturity stages of male gametes were identified according to description given by Hatanaka<sup>19</sup> and were divided into four different stages as follows: I-juvenil-immature, II-functional and maturing, III- functionally-mature, and IV-spent. Mature females were categorized into one of the four reproductive stages (I- juvenil-immature, II-preparatory, III-maturing and IV-spawning) based on Mangold-Wirz<sup>5</sup> and Bakhoyokho<sup>20</sup>. Stages of gamet development were classified based on the appearance and relative size of the reproductive organs. Data on reproductive components were collected from cephalopods received in a suitable condition by weighing the following components (to 0.001 g): male testis and spermatophoric complex; female nidamental gland, ovary, oviducal complex (gland+oviducts) and proximal oviduct. Paired organs were weighed one-piece and recorded. These data were used to determine the maturity indices.

From intact mature males (♂), spermatophoric complexes were fixed in 10% buffered formalin (excess CaCO<sub>3</sub>) for >48 h. Total numbers of spermatophores within these complexes were counted. Measurements of fixed spermatophores were taken using a binocular microscope (Olympus CH40). Ovaries and oviducal complexes from mature females (♀) were also fixed in formalin. They were then

rinsed thoroughly in tap water and broken up, using blunt seekers and a spatula, in order to remove strands of connective tissue and other nonegg material. All eggs within the distal oviducts and oviducal glands were counted before these structures were discarded. Estimates of ovarian and (proximal) oviducal fecundities (egg numbers) were obtained gravimetrically from counts of preweighed subsamples (5x -0.02 g for ovaries; 5x ~0.04 g for oviducts). Samples (N=200) of unrinsed eggs were taken from the ovary and oviduc specimens, and measured by the method described previously for the male spermatophores.

The annual sex ratio was calculated and deviations from the 1:1 ratio were assessed using a Chi-square test.

The length at which 50% of common cuttlefish were mature was determined by fitting the logistic model to the percentage mature (Stage IV) at length:  $P=1/(1+\exp[-1*(a+b*ML)])$ <sup>21</sup>.

The spawning peaks were identified using the indices of reproductive status: females GSI Gonadosomatic index:  $(OM/BM)*10022$ .

OM= Ovary mass,  
BM= Body mass.

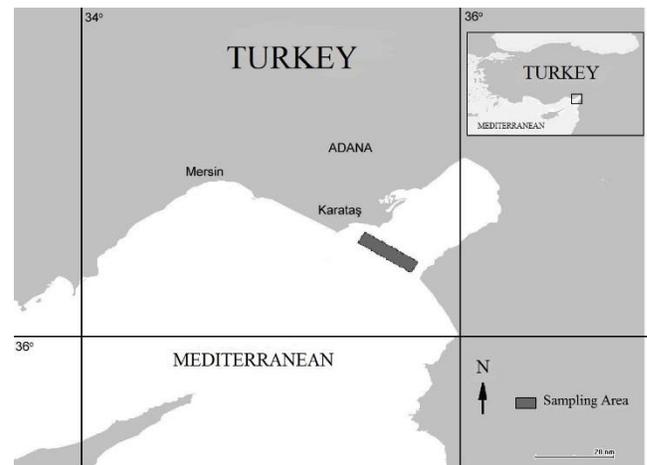


Fig. 1— Map of the Iskenderun Bay showing the study site.

### Results and Discussion

Mantle length (ML) values were ranged from 5.1 and 16.4 cm (mean  $9.83\pm 1.66$  cm) in 992 females, and from 4.8 to 19.2 cm (mean  $9.40\pm 1.76$  cm) in 1014 males. Length frequency distributions of the population are shown in Figure 2.

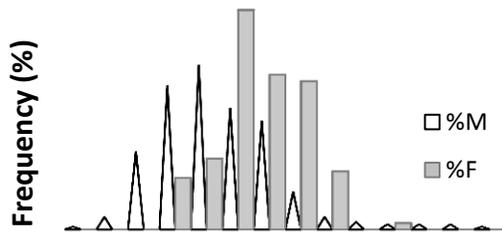
The length frequency distribution of the population showed that the 9.0-9.9 cm length classes had the highest number of specimens (Figure 2A). Females dominated at the larger size (11-14cm) whereas males

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predominated the smaller size classes (9-10cm), (Figure 2B). The mean weights for male and female were recorded as 115.34±6.29, 135.78±6.71 g, respectively.

Mantle lengths (ML) of *S. officinalis* population in Iskenderun Bay were between 4.8 and 19.2 cm in the present study. Önsoy and Salman<sup>16</sup> reported mantle lengths from 63 to 241 mm, while Türeli-Bilen<sup>18</sup> reported values between 22 and 155 mm, and Akyol<sup>17</sup> reported from 5.5 to 22.4 cm ML for this species.

A)



B)

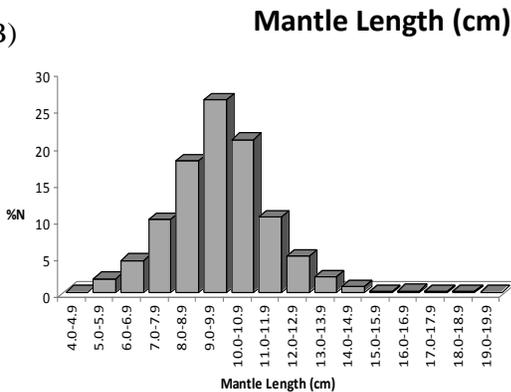


Fig-2 — A)- The length-frequency distribution of the population (n= 2006), B)- The length- frequency distribution of males (n=1014) and females (n=992).

Table 1— Ratios of absolute and relative (%) frequencies of male (□M) and female (□F) *Sepia officinalis* from Iskenderun Bay between September 2005 and August 2006 with results of the Chi-Square ( $\chi^2$ ) test for a significant difference from 1:1 in the sex ratio.

Year	Sex distribution	Sex ratio (%)	$\chi^2$
	(Female: Male)	*F: *M	
	*F: *M		

2005-2006	992:1014	49.45:50.55	241, d.f.=1,n.s.
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Our results agree with the description given for *S. officinalis* by<sup>18</sup>, while bigger ML were recorded for *S. officinalis* by<sup>16, 17</sup>. However, our findings are parallel with Karataş population<sup>18</sup>, present mantle length distribution data are differ from Aegean Sea cuttlefish populations<sup>16, 17</sup> because of environmental, nutritional and genetic diversities. Moreover, Turan and Yağlıoğlu<sup>15</sup> reported that *S. officinalis* populations are divided into four isolated populations in Turkish Seas (Aegean, Marmara and Mediterranean populations).

Of the 2006- Iskenderun Bay *S. officinalis* examined 992 (49.45%) were females, and 1014 (50.55%) were males. Annual female: male sex ratio (1F:1.02M, n= 2006) did not differ significantly ( $p > 0.05$ ) from the expected ratio of 1F: 1M (Table 1). Similar results were previously found in the English Channel, North Sea<sup>23</sup>, in the Homa lagoon (Aegean Sea)<sup>16</sup>, in the Antalya Bay (Mediterranean Sea)<sup>22</sup> and in the Sado Estuary (Atlantic Ocean, Portugal)<sup>21</sup>.

Monthly maturity stages of females and males during the sampled period are shown in Table 2. Maturing population was observed in all sampled months for females, stage IV (full matured) was observed most of the sampling period except in September, October and April (Table 2). Full matured male population was also observed in all months except in March (Table 2). Ripe and running gonads (stages III-IV for females, stages II-III for males) of both sexes were found in commercial samples from whole sample months (Table 2). In all months some maturing and mature individuals were found. These results suggest that *S. officinalis* probably spawns during the whole year.

Results of the GSI calculations for *S. officinalis* from September 2005 to August 2006, are represented in Figure 3. On the whole GSI value was higher for females than males. The monthly GSI results indicated that the reproductive season of cuttlefish ranged from February to August, with a maximal GSI of 6.91 in May for females and a maximal GSI of 1.96 in March for males (Figure 3).

In the 2005-2006 breeding season, females have two peaks in the year-round spawning period (Figure 3). First peak was observed in February, while the second one was recorded in May. Besides, the males of *S. officinalis* have homogeneous GSI values throughout the cycle.

Önsoy and Salman<sup>16</sup> reported two peaks in March and June and spawning period of cuttlefish in Homa

lagoon from March to June. Results of the present study are similar to the findings of<sup>16</sup>. Guven and Ozbaş<sup>22</sup> observed a spawning period extending throughout the year in *S. officinalis* in the Antalya Bay.

Table 2— Monthly percentage of male and female individuals maturity stages.

Months	N	Males			Females				
		I	II	III	N	I	II	III	IV
September	230	31.3	53.5	15.2	213	15.0	79.3	3.8	1.9
October	80	30.0	43.8	26.3	91	3.3	81.3	15.4	
November	44		25.0	75.0	37		13.5	29.7	56.8
December	52		44.2	55.8	49		6.1	63.3	30.6
January	69		76.8	23.2	98		13.3	80.6	6.1
February	72		58.3	41.7	80			56.3	43.8
March	25		96.0	4.0	78			98.7	1.3
April	146		82.9	17.1	55			100.0	
May	102		88.2	11.8	56			98.2	1.8
June	88	1.1	45.5	53.4	68		2.9	79.4	17.6
July	48		47.9	52.1	65		64.6	12.3	23.1
August	58		72.4	27.6	102		78.4	17.6	3.9
<b>Total</b>	<b>1014</b>				<b>992</b>				

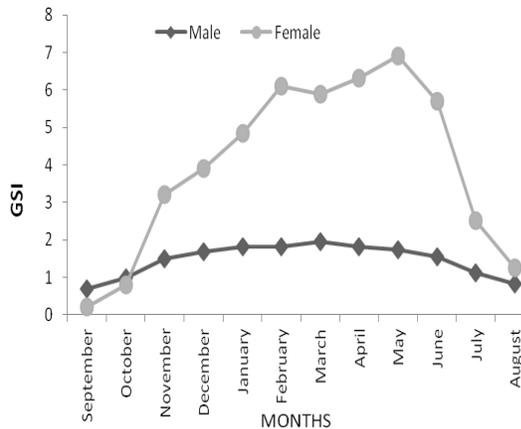


Fig-3— Gonadosomatic index (GSI) of female and male cuttlefish from the Iskenderun Bay.

At the same time, these authors noted two spawning peaks (June-July). This shows that the spawning season and the reproductive peaks for the cuttlefish are very variable and probably very closely connected to some abiotic environmental conditions. Türelı-Bilen<sup>18</sup> reported that spawning period of cuttlefish in Karataş from April to September. Furthermore, spermatophore lengths were from 5 to 14 mm in Homa lagoon<sup>16</sup> and from 2,8 to 12,4 mm in Iskenderun Bay (from the present study).

The first sexual maturity of females and males were recorded at 12.04 cm and 10.30 cm ML. First sexual maturity (50%) was smaller for males than females.

$$P=1/(1+\exp(-1*((-9.218)+(0.806)*ML))) \quad r=0.998$$

for females

$$P=1/(1+\exp(-1*((-9.435)+(0.877)*ML))) \quad r=0.981$$

for males

The length at first sexual maturity of females was higher than that of males: most of researchers have reported similar patterns<sup>16, 21, 23</sup>, but was different from the findings of<sup>17</sup>. Furthermore, Dunn<sup>23</sup> estimated size at maturity of *S. officinalis* to be 164 mm for females, 146 mm for males from English Channel. Önsoy and Salman<sup>16</sup> found that cuttlefish reached maturity 130 mm for females, 90 mm for males in Homa lagoon in İzmir. Neves<sup>21</sup> calculated length at first maturity of *S. officinalis* to be 80 mm ♀, 59 mm ♂ from Sado Estuary. Akyol<sup>17</sup> reported maturity mantle length 80 mm ♀, 110 mm ♂ in İzmir Bay. The present study sample sizes at first sexual maturity estimate of 120 mm ♀ and 103 mm ♂ for *S. officinalis* in Iskenderun Bay were lower than other studies<sup>16, 23</sup>. But, our sample sizes at maturity estimates were higher than<sup>21</sup> and also<sup>17</sup>. There was a variation detected in size at maturity for *S. officinalis*. We suggested that food availability, age, locality, and environmental conditions were playing the major role for the maturation size.

Spermatophore lengths were ranged from 2.8 to 12.4 mm. The Table 3 indicates that spermatophore lengths increase with reproduction periods of *S. officinalis*.

The mean egg lengths of female individuals were calculated as 5.367±9.12 mm. The diameter of egg lengths were ranged from 2.804 to 8.50 mm females of *S. officinalis*.

Table 2— Monthly percentage distribution of spermatophore lengths according to mantle length groups.

MONTHS	N	Spermatophore lengths (mm)										
		2.0-2.9	3.0-3.9	4.0-4.9	5.0-5.9	6.0-6.9	7.0-7.9	8.0-8.9	9.0-9.9	10.0-10.9	11.0-11.9	12.0-12.9
September	24	0	0	4.2	12.5	54.2	29.2	0	0	0	0	0
October	44	0	0	13.6	20.5	31.8	27.3	4.5	0	2.3	0	0
November	45	0	0	2.2	2.2	2.2	17.8	22.2	42.2	8.9	0	0
December	40	0	0	0	0	2.5	15.0	45.0	22.5	12.5	2.5	0
January	54	1.9	0	0	3.7	38.9	35.2	18.5	0	0	0	0
February	66	0	0	1.5	0	0	21.2	37.9	24.2	7.6	6.1	1.5
March	24	0	0	0	0	8.3	41.7	45.8	4.2	0	0	0
April	135	0	0	0	0	3	35.6	44.4	14.1	3.0	0	0
May	84	0	0	0	1.2	0	22.6	51.2	20.2	3.6	0	0
June	33	0	0	0	6.1	36.4	36.4	18.2	0	3.0	0	0
July	18	0	0	0	0	33.3	44.4	33.3	5.6	0	0	0
August	13	0	0	0	23.1	38.5	38.5	0	0	0	0	0

The present study provides detailed bio-information on mantle length at first maturity of *S. officinalis* in Iskenderun Bay, which is reported for the first time. This will assist in the fisheries management and also alternative culture pilot studies of common cuttlefish in the Levantine Sea region.

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