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Reproductive biology of *Eledone moschata* (Cephalopoda: Octopodidae) in the Gulf of Cádiz (south-western Spain, ICES Division IXa)

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Reproductive aspects of *Eledone moschata* from the Spanish waters of the Gulf of Cádiz are reported for the first time. The species is relatively abundant over the continental shelf and it is caught as by-catch by the bottom-trawl fleet. Monthly samplings throughout one annual life-cycle showed a sex ratio with a female dominance (0.45:1). A four stage maturity scale was used for both sexes. The reproductive period was determined from monthly evolution of these stages as well as that of several maturity and condition indices. The breeding season extended from October to June, with spawning peaks in October and March–May, the second one being more important. Length and weight at maturity (mantle $\text{length}_{50,}$ body weight_{50}) were estimated at 7.8 cm and 97 g in males, and at 12.2 cm and 274 g in females, respectively. Total fecundity, estimated from 52 mature females, was 443.5 ±154.4 oocytes on average. The mean size of mature oocytes and the mean length of mature spermatophore in mature females and males was 10.24 mm (±1.07) and 13.88 mm (±1.60), respectively.

INTRODUCTION

The musky octopus *Eledone moschata* (Lamarck, 1799) was formerly described as an exclusively Mediterranean species. However, this species is present in the Iberian Atlantic waters (Guerra, 1982, 1992; Mangold, 1983; Reis et al., 1984; Roper et al., 1984). In the Gulf of Cádiz, *E. moschata* is a relatively abundant species which is distributed all over the continental shelf (Figure 1). This species is caught as by-catch by the Spanish local bottom-trawl fleets, although in many cases it is discarded because of its low commercial value.

The major part of the scarce literature on the biology of *E. moshata* has been compiled by Mangold (1983), including field and laboratory studies both by this author (Mangold-Wirz, 1963) and Boletzky (1975), which were based on wild-caught and reared specimens from Mediterranean populations from the Gulf of Lion and the Catalonian Sea. Most recently, Lefkaditou et al. (1998) and Belcari et al. (2002) analysed the distribution, abundance and demographic structure of this species in different areas of the Mediterranean basin. Ezzeddine-Najai (1997) also analysed some aspects of the reproductive biology of the species from the Tunisian coasts. At present, however, the biology of *E. moschata* in Spanish Atlantic waters is unknown.

The present paper studies for the first time some aspects of the biology of *E. moschata* in the Spanish waters of the Gulf of Cádiz and attempts to contribute to the knowledge of the biology of this species in the westernmost part of its distribution range. The study presents data on bathymetric distribution, length–weight relationships, sex ratio, maturation, reproduction, condition and fecundity of the species.

MATERIALS AND METHODS

Monthly samples of *Eledone moschata* from commercial bottom-trawl catches landed in the main landing port (Sanlúcar de Barrameda) of the Gulf of Cádiz were taken from June 1998 to June 1999 (Figure 1). A total of 1306 individuals randomly selected were sampled (406 males and 900 females) and analysed in the laboratory for a detailed biological study on the species. The dorsal mantle length (ML) and total body weight (BW) were recorded in all specimens rounded downwards to the nearest cm and g, respectively, both variables being recorded in fresh specimens. For detailed observations of reproductive organs the following data were collected: ovary and testis weight (OW and TW), oviducal complex weight (OCW), oviducal gland length (OLG) and Needham's complex weight (NCW).

The bathymetric distribution by depth strata of the species was described from the results obtained in the scientific bottom-trawl surveys series carried out by the IEO (Spanish Oceanography Institute) in the Spanish waters of the Gulf of Cádiz (1993–1999) (Figure 1). The surveys were carried out on-board the RV 'Cornide de Saavedra', with a Baka 44/60 trawl gear equipped with a 20 mm mesh cod end liner. The sampling design adopted in these surveys series followed a stratified random sampling scheme, with the number of hauls per depth stratum being prorportionally allocated to the trawlable surface of each stratum. Methodologies of these surveys have been described by Jiménez et al. (1998).

Length–weight relationships (ML–BW) for the species and by sex were estimated. Equations of the form $BW = a * ML^b$, where a and b are constants of regression,

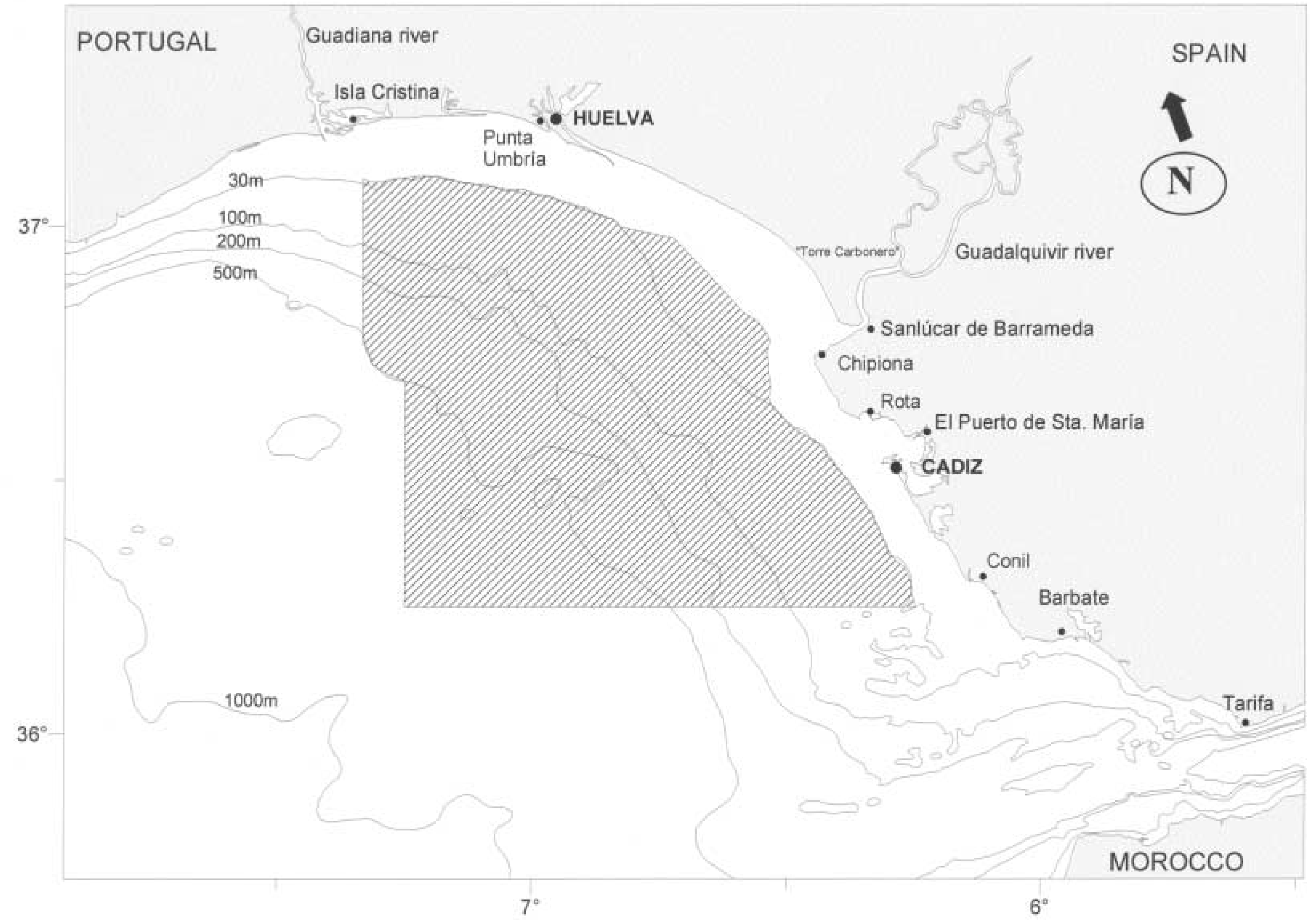

Figure 1. Spanish waters of the Gulf of Cádiz showing the main landing ports in the region and the surveyed area in the research cruises.

were fitted by transforming the data into logarithms and deriving the regression line by the least squares method. Goodness of fits were expressed by their respective coefficients of determination, r^2, and analyses of covariance (ANCOVAs) were used to test for differences between the slopes obtained for each sex.

The sex ratio was calculated annually and monthly, by bathymetric strata (<30, 31–100, 101–200, 201–500, >500 m depth) and in relation to body size-ranges (4–6, 7–9, 10–12, 13–15 cm ML). Significant deviations from the 1:1 sex ratio were tested by the χ^2-test ($P<0.05$). Maturation and reproduction were assessed by maturity scales and indices. A four-stage scale was used in both sexes (I, immature; II, maturing; III, mature, and IV, post-spawning) following that proposed by Ezzeddine-Najai (1997). The indices used were gonadosomatic index (GSI): GSI=(OW/BW−OW)*100; oviducal gland index (OGI): OGI=(DML/OGL)*100; Guerra's index (GI) in males (Guerra, 1975): GI=NCW/(NCWTW); Guerra's index (GI) in females (Guerra, 1975): GI=OCW/(OCW−OW).

The size (length) at maturity ($MLm_{50\%}$) was estimated after fitting, by the least squares method, the relative length−frequency distribution of mature individuals (Stages III and IV) to a logistic curve from the expression: $P_i = 1/1+\exp-(a+bML_i)$, where P_i represents the relative frequencies of fully mature individuals in length-class ML_i, a and b are the regression constants, and $MLm_{50\%}=-a/b$. Body weight at maturity ($BWm_{50\%}$) was also estimated following the same procedures. For these estimates, mature animals were grouped in one cm ML and 50 g BW classes, respectively.

Individual fecundity was gravimetrically estimated from the ovaries of 52 mature females, with a range of sizes and of weights of 8–15 cm ML and 120–500 g BW, respectively. During the egg counting of the whole ovary, the residual oocytes were differentiated, as described in other octopodid species (Mangold, 1983; Cortéz, 1995). Residual oocytes are opaque and much smaller than mature ones, and with a minimum probability of being laid during the spawning. The mean size of the whole

Table 1. *Parameters of the relationship between dorsal mantle length (ML) and body weight (BW) for males, females and total population of* Eledone moschata *from Spanish waters of the Gulf of Cádiz.*

Sex	a	b	*r*	N
Males (M)	0.2613	2.794	0.959	406
Females (F)	0.3573	2.660	0.960	900
M+F	0.3233	2.702	0.959	1306

N, number of individuals; *r*, coefficient of determination.

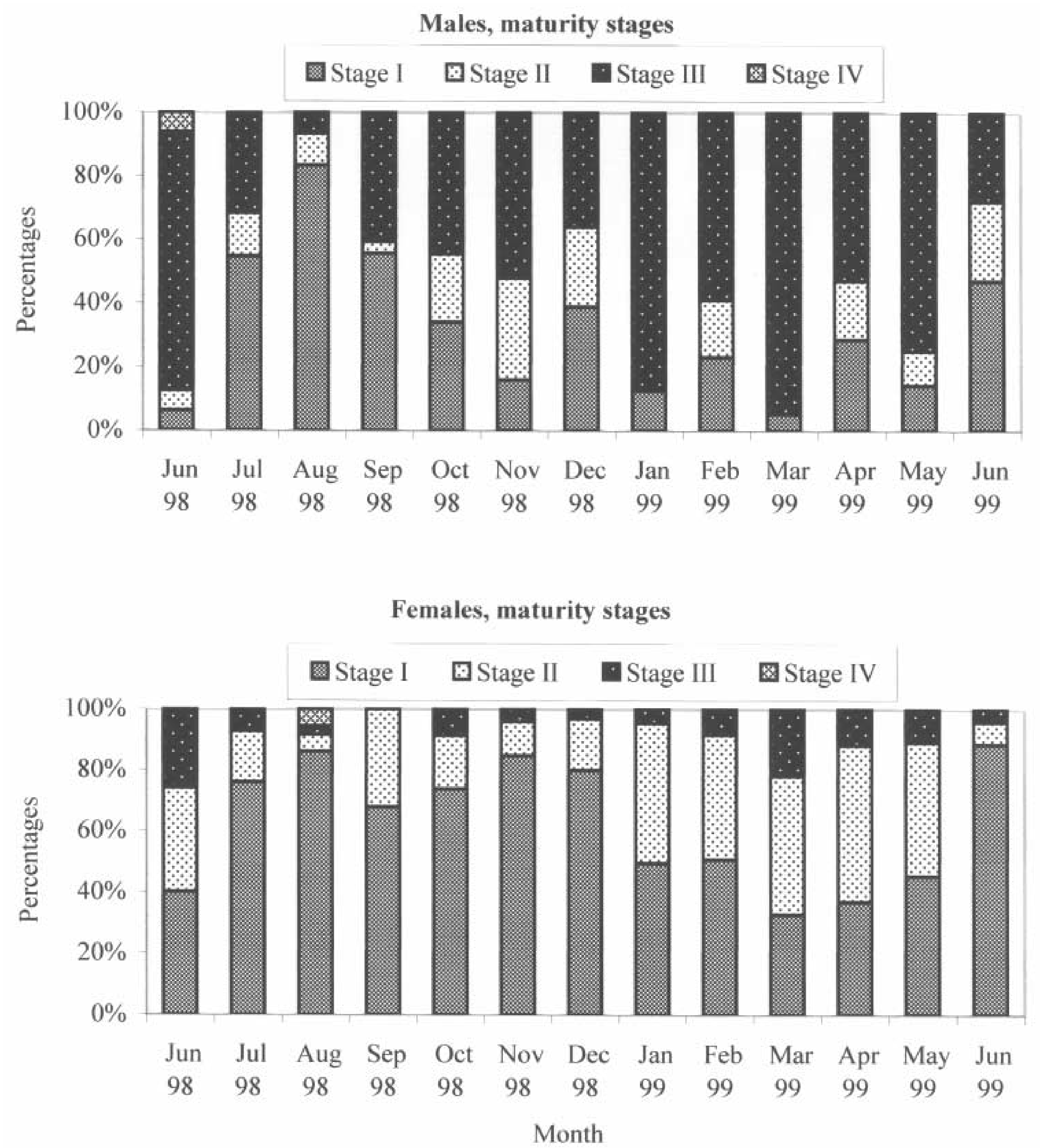

Figure 2. *Eledone moschata*. Monthly percentages of each maturity stage (I–IV) in males and females.

non-residual oocytes and only of those of largest size (i.e. oocytes >9 mm; Ezzedine-Najai, 1997) was estimated. In males, the mean length of fully-developed spermatophores (i.e. mature spermatophores) occurring in all Stage III individuals was estimated.

RESULTS

Bathymetric distribution

The species occupied a wide bathymetric range from 21 to 438 m. The most coastal specimen fished measured 7 cm ML and weighed 130 g BW. The deepest specimen caught measured 5 cm ML and weighed 45 g BW. Results from bottom-trawl surveys showed that the maximum number of specimens occurred between 50 to 75 m depth, registering 32% of the total of the sampled population. Likewise, it was observed that 85% of the caught specimens are located in shallower depths than 100 m, and only 0.2% in bottoms deeper than 200 m depth.

Length–weight relationships

The size–range was between 4–15 cm ML for males and females. The weight varied from 20–640 g BW and from 14–510 g BW for males and females, respectively. Table 1 shows the parameters of the size–weight (ML–BW) relationship by sex and for the total population for the sampled period. The results of the ANCOVA showed that the differences of the size–weight relationships of both sexes were not significant ($P>0.05$).

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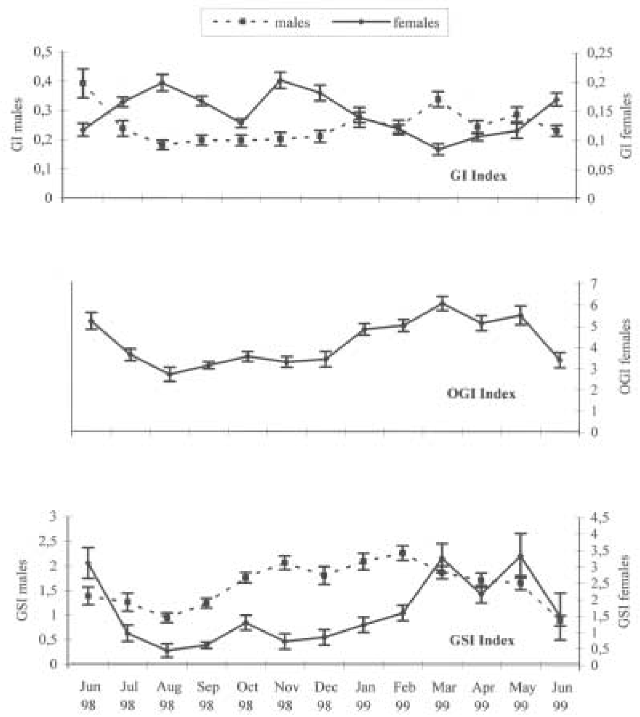

Figure 3. *Eledone moschata*. Monthly changes of the gonadosomatic index (mean GSI ±SE) in males and females, oviducal gland index (mean OGI ±SE) in females and Guerra's index (mean GI ±SE) in males and females, (SE, standard error).

Sex ratio

The annual sex ratio (males:females) of the sampled specimens was 0.45:1. The application of the χ^2-test showed that this difference was significant ($P<0.05$), even when analysing the sex ratio by month. The only non-significant difference between sexes ($P>0.05$) was found in August (0.9:1). The sex ratio by size-range and by bathymetric strata showed significant differences in favour of the females ($P<0.05$), in all cases.

Spawning season

The monthly evolution of the maturity stages of males and females during the sampled period is shown in Figure 2. Excepting September, mature females were observed in all the monthly samples, although in low proportions. The highest monthly percentage of mature females (28%) was recorded in June 1998, although this situation was not repeated in the following year. Notwithstanding the above, higher reproductive activity seems to occur since early winter, which culminates through late-winter and spring months, peaking in March 1999. A secondary autumn peak in reproductive activity may also be deduced from the percentage of mature females recorded in October 1998.

In males, an important fraction (over 30%) of mature males appears throughout the whole year, excluding August (8%). The most important proportion of mature males was found from January to May 1999. In March, almost 100% of the males were mature. As mentioned previously for the females, differences in the month of June in both years were observed.

The maturity and condition indices of males and females show a similar trend to that of the proportion of maturity stages over the year. Figure 3 plots the monthly evolution of the GSI for both sexes. Females have their minimum values in August. Since then, there is an

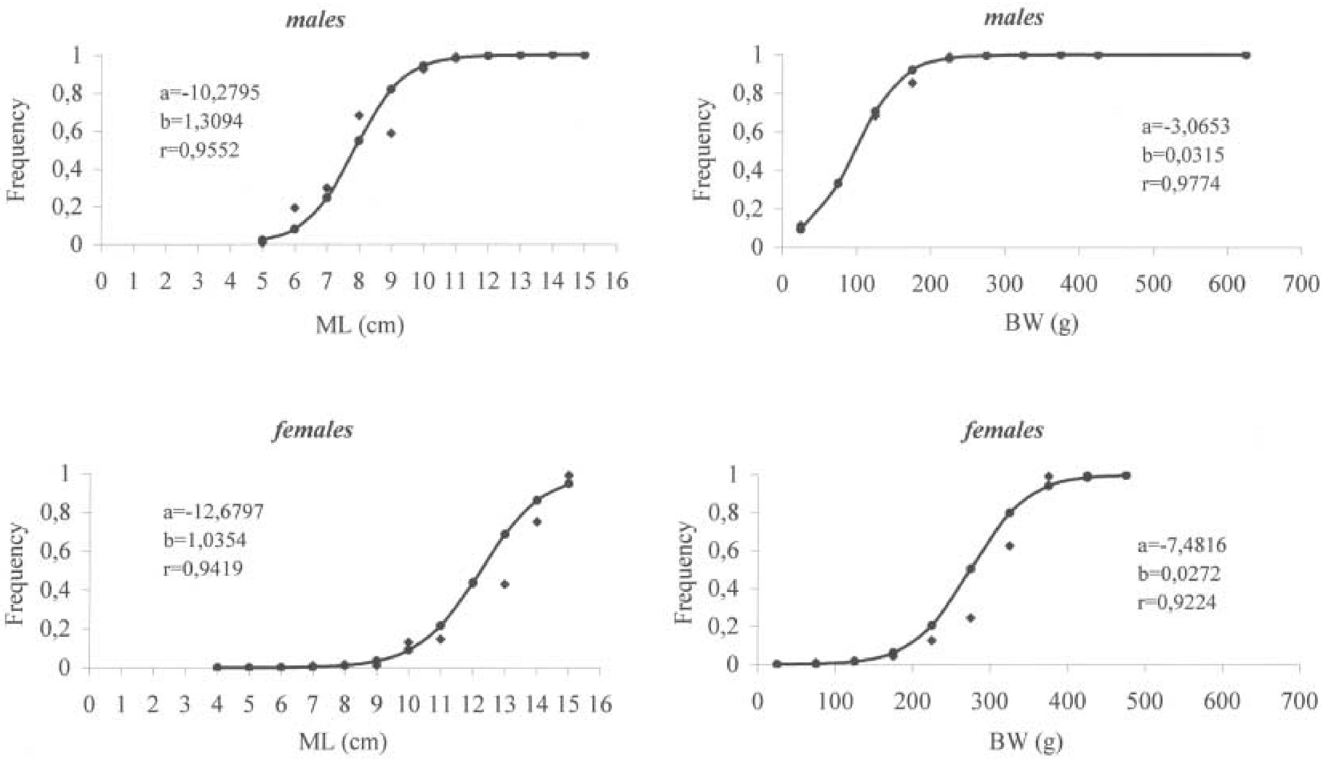

Figure 4. *Eledone moschata*. Maturity ogive corresponding to dorsal mantle length (ML) and body weight (BW) for males and females.

increasing trend until the spring of the following year. In October, a small peak is observed, and two higher ones in March and May, followed by a decrease in June. An aspect worth mentioning is that the maximum value was attained in the first month of sampling, June 1998. In the following year, this value was reasonably lower for the same month.

The same tendency can be observed in the evolution of the OGI (Figure 3), which shows a small peak in October and maximum values from March to May. The values for June in 1998 and 1999 show the same trend as the GSI.

The GI is represented in Figure 3, for males and females respectively. Females show minimum values in spring and in October, coinciding with the months of a higher proportion of mature females and higher GSI values. Conversely, males reach the highest values in the months of a greater proportion of mature males, that is, from January to May.

Size and weight at maturity

The sizes and weights of the smallest mature specimens sampled were 6.1 cm and 52 g in males, and 6.5 cm and 65 g in females. Figure 4 shows at maturity ogives corresponding to males and females, in weight and size. The size at maturity ($MLm_{50\%}$) were estimated at 7.8 cm ML in males, and 12.2 cm ML in females. With respect to weight (BW), weight at maturity ($BWm_{50\%}$) was estimated at 97 g in males and at 274 g in females. The parameters of the logistic model (a,b) and the coefficient of determinations (r) obtained for each case are included in Figure 4.

Fecundity

The minimum and maximum estimates of total fecundity were 187 and 944 oocytes in two females measuring 8.5 cm ML (130 g BW, 11.7 g OW) and 13.7 cm ML (480 g BW, 36.7 g OW), respectively. The mean total fecundity (ITF) during the spawning period was estimated to 443.48 ±154.38 oocytes per female. A number of residual oocytes were observed in the ovaries, especially in those females showing a more advanced maturity stage. The observed mean proportion of residual oocytes and its standard deviation was 6.6% ±2.91.

Average relative fecundity, defined as the number of oocytes per gram of female, was estimated at 1.43 ±0.36 when referring to the total weight of the specimen, and 1.840.49 when referring to the eviscerated weight.

The mean size of the largest oocytes was 10.90 ±1.22 mm, per female. The maximum oocyte size found was 14.8 mm. The mean size of all the sampled non-residual oocytes was 10.24 ±1.07 mm, per female.

The mean length of fully-developed spermatophores (mature spermatophores) was of 13.88 ±1.60 mm. The maximum and minimum spermatophore lengths were 17.5 and 10.9 mm, which were found in two males of 240 g and 10 cm, and 98 g and 6 cm, BW and ML respectively.

DISCUSSION

Although *Eledone moschata* is a typical species of the Mediterranean (Mangold-Wirz, 1963; Mangold, 1983), it is also relatively abundant in the Atlantic waters of the Gulf of Cádiz. The same occurs with other species due to the proximity of the Mediterranean and to the prevailing oceanographic and environmental conditions in the area. As described in the Mediterranean, in the Gulf of Cádiz this species is mainly distributed over the shelf, in depths of over 100 m. The presence of specimens beyond 200 m has also been reported along the coasts of Barcelona (Morales, 1958), Algeria (Dieuzeide & Roland, 1957) and other areas of the Mediterranean basin (Belcari et al., 2002).

In the study area, both annual and monthly sex ratios showed an overall predominance of females. These results differ from those reported for the species in the Mediterranean by Mangold (1963, 1983; south-eastern French coasts) and Ezzedine-Najai (1997; Gulf of Gabès, Tunisia), where the sex ratio was significantly in favour of males, especially during the reproductive season.

In the Mediterranean, Mangold-Wirz (1963) and Mangold (1983) detected reproductive migrations that affected the sex ratio in relation to size in particular areas and seasons. This type of phenomenon was also observed by Ezzedine-Najai (1997). This researcher indicated that the mature females leave the fishing grounds during the spawning season, hence favouring the predominance of males in the sex ratio.

In our case, this phenomenon was not observed, and contrarily, there was a major proportion of females throughout the sampled period. Furthermore, in the analysis of the results from the trawl surveys, a predominance of females by bathymetric strata was also observed. Moreover, in the comparison of the samples from November 1998 and March 1999, with those of the surveys which were carried out during the same months, the results were very similar (0.44:1 and 0.61:1, trawl surveys 11/98 and 03/99, respectively). Therefore, although the combination of a migratory behaviour related to reproduction and the fact that a non-targeted fishery could favour the deviation from a 1:1 sex ratio, the results from the trawl surveys corroborate those obtained from the fishery-based sampling programme. In conclusion, the species *E. moschata* in the Gulf of Cádiz presents a greater proportion of females, with a mean sex ratio around 0.5:1 (male:female).

The GSI of both sexes correlated noticeably with the mature fraction of the population. The diameter of the oviduct gland of the females also showed an increase with sexual maturation. The relationship between the decrease of the GI in females and the increase of this index in males was also confirmed; increasing gonad size in females and the number of spermatophores in Needham's sac, respectively, with progressive sexual maturation.

As a result of this analysis, it can be stated that the reproductive season of *E. moschata* in the Gulf of Cádiz affects practically the entire year, with the exception of August and September. However, the peak spawning mainly occurs from February to May. A less intensive peak is also detected in autumn, particularly in October, as shown by the increase of the GSI and the OGI, the decrease of the GI and the proportion of mature females in that month. These results coincide with those reported by Ezzedine-Najai (1997) in the Gulf of Gabès (Tunisia), also presenting its peak between February and May, and an extensive spawning season of approximately nine months, but slightly shifted in time, from November to July. On the other hand, these results differ from those of Mangold (1983) who established a shorter reproductive season, from January to May–June.

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With regard to the size and weight at first maturity in females, the results of this study are similar to those of Ezzedine-Najai (1997) for the Gulf of Gabès. This author reported a size of 11 cm and a weight of 274 g at first maturity, in contrast with the 12 cm and 274 g estimated in this study. Moreover, these findings differ from the study of Mangold (1983) stating that females below 400 g are not mature.

Comparing this study with others carried out in the Mediterranean, it seems that a series of environmental variables influence the reproductive biology of this species, in particular the temperature, which may influence the precocity in the sexual development and a greater temporal duration of reproduction, as maintained by Ezzedine-Najai (1997) for the Gulf of Gabès population. Similar temperature ranges are recorded at the same depths and areas (Ktari-Chakroun & Azouz, 1971; García et al., 2002).

The peak spawning and the temporal duration of the spawning period can be highly influenced annually by diverse environmental factors, as previously described for *Octopus vulgaris* in the same study area (Silva et al., 2002). This was evident from the results of the biological sampling. Thus, from the values of the different indices recorded in June 1998 it may be inferred that the spawning period in that year included this month, whereas in the following year the main spawning season finished in May.

The life cycle model of this species in the north-eastern Mediterranean is based on the alternation of short-living and long-living life cycles (Mangold, 1983). This model, which has also been observed in other Gulf of Cádiz cephalopod species such as *Sepia officinalis* (Ramos et al., 2001), seems to also occur in the Gulf of Cádiz musky octopus. A recruitment is detected in September and October. It presumably originates from the long-lived fraction of mature females that spawned at the beginning of the spawning season. Another recruitment is detected in January and February, which in this case is related to the short-lived fraction of the population that spawned at the end of the spawning season. We can assume that favourable environmental conditions, as previously mentioned, could influence a faster growth and a more rapid sexual development in the short-living fraction of the population. The smaller spawning peak observed in October could be the consequence of this phenomenon.

Internal insemination is confirmed by the presence of 6–10 sperm sacs found in the ovaries of some females, both from commercial and research samples. As in other populations, the number of females found in this state was very scarce, indicating that the number of spermatophores that reach the ovary is rather reduced and that mating does not take place long before spawning (Mangold, 1983; Ezzedine-Najai, 1997). Thus, the fact of finding fecundated females at the beginning of November in the surveys confirms the inclusion of the months of October and November in the reproductive season.

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