REPRODUCTIVE BIOLOGY OF FEMALES OF *PLESIONIKA EDWARDSII* (BRANDT, 1851) (CRUSTACEA, DECAPODA, PANDALIDAE) IN THE NORTHERN TYRRHENIAN SEA (WESTERN MEDITERRANEAN)

Abstract - Individuals of P. edwardsii were collected on soft bottoms from 300 to 450 m depth through experimental traps and samplings on commercial trawlers from December 2003 to April 2006. A total of 4919 specimens (2385 females, 1451 males and 1083 undetermined) was analysed. Sex was macroscopically determined: adult and subadult males show the masculine appendix on the second pair of pleopods. Sizes ranged from 10 to 29 mm carapace length (CL) for females, from 11 to 26 mm CL for males. Sex ratio was in favour of females along all the investigated period. Females with mature ovaries and ovigerous ones were observed all year long, except in December, even though sexual maturity peaked from March to July. Physiological size at first maturity (size where 50% of females shows mature ovaries) and the functional one (size where 50% of females is ovigerous) were 18.5 and 20.2 mm CL, respectively. Absolute fecundity ranged from about 1,000 to about 20,000 eggs for ovigerous female. A positive correlation between number of eggs and size was obtained.

Key words - *Plesionika edwardsii*, crustaceans, decapoda, reproductive biology, fecundity, resource assessment, northern Tyrrhenian Sea, Western Mediterranean.

Riassunto - Biologia riproduttiva delle femmine di Plesionika edwardsii (Brandt, 1851) (Crustacea, Decapoda, Pandalidae) nel Mar Tirreno settentrionale (Mediterraneo occidentale). I campioni mensili del gamberetto Plesionika edwardsii, raccolti da dicembre 2004 ad aprile 2006, sono stati ottenuti sia attraverso campagne di pesca con nasse sperimentali, sia tramite campionamenti effettuati a bordo di pescherecci a strascico, in zone comprese tra 300 e 450 m di profondità, caratterizzate da fondali fangosi con ripide pendenze. Sono stati analizzati 4919 esemplari (2385 femmine, 1451 maschi e 1083 indeterminati). Il sesso è stato determinato macroscopicamente: i maschi adulti e subadulti possiedono l'appendice mascolina, localizzata sul secondo paio di pleopodi. La struttura in taglie è risultata compresa tra 10 e 29 mm di lunghezza del carapace (LC) per le femmine e tra 11 e 26 mm LC per i maschi. La sex ratio è risultata spostata a favore delle femmine per tutto il periodo investigato. Femmine con ovari maturi e femmine ovigere sono state osservate in tutti i mesi dell'anno, ad eccezione di dicembre, anche se il picco di maturità sessuale è risultato compreso tra marzo e giugno. La taglia di prima maturità fisiologica (taglia alla quale il 50% delle femmine ha ovari maturi) e quella funzionale (taglia alla quale il 50% delle femmine è ovigera) sono state rispettivamente di 18,5 e 20,2 mm LC. La fecondità assoluta è risultata compresa fra poco meno di un migliaio a circa 20.000 uova per femmina ovigera. È stata individuata una correlazione positiva del numero di uova con la taglia.

Parole chiave - *Plesionika edwardsii*, crostacei, decapodi, biologia riproduttiva, fecondità, gestione delle risorse, Mar Tirreno settentrionale, Mediterraneo occidentale.

INTRODUCTION

The pandalid decapod crustacean *Plesionika edward-sii* (Brandt, 1851) is a nektobenthic species, present in a wide bathymetric range: in Mediterranean it has been recorded from 100 to 650 m depth (Colloca *et al.*, 1998). *P. edwardsii* is a gregarious species, able to perform nictemeral vertical migrations, mainly for feeding (Relini *et al.*, 1999).

P. edwardsii is one of the largest Pandalid species, the maximum size reported for the Mediterranean Sea is 30 mm CL (carapace length) (Colloca, 2002). The species, called «striped soldier shrimp» thanks to the longitudinal, dark red stripes on the abdomen (Fisher *et al.*, 1987; Relini *et al.*, 1999), is of commercial interest, and it is included in the FAO catalogue of species of interest to fisheries (FAO, 2005). Landings mainly comes from fishing with traps and trawling (De Ranieri, 2005). In spite of this, a targeted fishery for *P. edwardsii* is still absent, so that this resource can be considered as underexploited.

In expectation of a future increase of fishing pressure, it is important to deepen the information on biology and population dynamic of this species, still scarce and scattered.

This study has been focused on the reproductive biology of *P. edwardsii*, in order to provide information on reproductive period, maturity size and fecundity.

MATERIALS AND METHODS

Data collection

Sampling was performed both with experimental traps (Sartor *et al.*, 2006) and through embarks on a professional bottom trawler of Porto Ercole fleet (Grosseto, Italy). Data were collected monthly in the period 2003-2006 to the South of Giannutri Island, in the northern Tyrrhenian Sea. Muddy bottoms from 300 to 450 m depth were investigated, in a zone far more than 20 miles from the harbour of Porto Ercole, characterised by high slopes.

Traps used for sampling were made following the structure of those in use in several Spanish fleets, targeted to the exploitation of *P. edwardsii* (Gestin & Guennégan, 1989; De Ranieri, 2005). Embarks were realized on a bottom trawler of 27 m overall length and 207 kw engine power, usually exploiting for bathyal crustaceans.

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Laboratory analysis

Size was registered as carapace length (CL, to 0.1 mm) on each specimen caught, with a precision calliper, from the posterior tip of the eye socket to the end of the carapace. Sex was assigned on the basis of the presence of the male secondary sexual characters. Adult and subadult males show two sexual appendages, one of them, the «masculine appendix», is located near the endopod of the second pair of pleopods (Fig. 1); this appendix is particularly small in juveniles and difficult to recognize (King & Moffitt, 1984).

Maturity of females was determined through the macroscopic analysis of the ovaries, their size and coloration. Three maturity stages were assigned, according to Ceccaldi (1966) and Company & Sardà (1997): stage I (immature), ovary small, pale, yellow-pinkish; stage II (maturing), ovary developed, light blue-violet; stage III (mature), ovary fully developed, dark blue.

Females were also classified as ovigerous and nonovigerous; ovigerous females were further classified in stage 1 (light blue eggs, without embryo pigmentation) and stage 2 (light brown-grey eggs, with embryos in different stages or fully developed larvae).

Total fecundity was estimated on monthly samples of 24 females with eggs in the two development stages; samples were chosen according to four female size classes (15-18 mm CL, 19-22 mm CL, 23-26 mm CL, 27-30 mm CL). Totally, eggs of 267 females (137 stage 1, 130 stage 2) were counted.

Counts were performed on sampling basis. For each female, the ovary content was placed in a Dollfuss cell of 200 square boxes of 0.5 mm side each; eggs of 25 boxes (1/8 of the total), randomly chosen, were counted. A preliminary analysis (Possenti, unpublished data) showed that this sample size was large enough to keep down in \pm 10% the estimate error.

Data analysis

Sex ratio was calculated as percentage of females on the total sampled population. Differences from the expected 1:1 sex ratio were investigated by Chi square test,

applied to the samples of each month, when the catches were greater than 50 specimens.

The reproductive period was investigated monthly, tacking into account the percentage of females in the different maturity stages of gonads and eggs.

Maturity size was estimated by calculating the size at which 50% of females were mature (physiological maturity) as well as the size at which 50% of females carried eggs on pleopods (functional maturity). In both cases a logistic curve was adapted to the experimental data.

Total fecundity was calculated raising the egg counts coming from the Dollfuss sampled boxes to the total Dollfuss boxes. Relative fecundity was estimated through the linear regression between number of eggs and carapace length.

RESULTS

Sex-ratio

4919 specimens were collected, ranging from 10 to 29 mm CL; the smallest ones were caught in May (modal size at 14 mm CL), June and July, the largest in winter (modal size 20-22 mm CL).

Sex was assigned on all the specimens from 15 mm CL; under this size, sex identification was difficult, because the male secondary sexual characters were not present or little developed.

1451 males and 2385 females were collected, showing a *sex ratio* of 62% (Chi square = 167, p < 0.001). At monthly level, females always significantly outnumbered males; only in three months the Chi square test gave not significant differences. Males ranged from 15 to 26 mm CL, females from 15 to 29 mm CL; the percentage of females by size class was always more than 50% (Fig. 2).

Maturity period

Specimens in the three maturity stages were caught each month, even though in different percentages, except in December.



Fig. 1 - P. edwardsii. Detail of the second pleopod with masculine appendix (MA) of the adult male.



Fig. 2 - P. edwardsii. Percentage of females by size class. ■ = No significant differences for sex ratio 1:1 (Chi square test).

Maturation of gonads showed a clear seasonal trend (Fig. 3). Mature females were concentrated in spring, with a peak in April, decreased in the following months to disappear in winter. Immature specimens were present all year round, but prevailed in winter.

Ovigerous females

The temporal evolution of the presence of ovigerous females is shown in Figure 4; the highest percentages were observed in spring and summer (92% in April and 70% in June). Ovigerous females decreased in the following months and disappeared in December. The presence of females with eggs in advanced development stage (stage 2) did not show evident seasonal differences, even though these specimens were always less abundant than the females with eggs in stage 1.

Size at first sexual maturity

The smallest females bearing eggs were 15 mm CL, and the smallest female with mature ovaries measured 13 mm CL. All the females over 27 mm CL had mature gonads; all females over 28 mm CL were ovigerous. The logistic curve gave a good fit of the observed data on the proportion of mature females by each size (Fig. 5). Physiological size at first maturity was 18.5 mm CL (Fig. 6).

Functional size at first maturity, calculated by using the presence of eggs on pleopods like criterion, was 20.2 mm CL; also in this case the logistic curve well fitted the observed data.

Taking into account only the adult females (*i.e.* the females larger than 18.0 mm CL, the size at first maturity), the temporal evolution of maturity (Fig. 7) shows the same pattern of that of the whole sample (Fig. 3).

Fecundity

The minimum and maximum number of eggs were 920 (recorded in a female of 21 mm CL) and 19,792 (in a specimen of 25 mm CL), respectively.

The estimations of total fecundity coming from the specimens caught by trawling gave, for each size class, values considerably lower than those from the individuals caught by traps (Fig. 8). Relative fecundity was significantly positively correlated to the animal size (CL) by a linear relationship (Fig. 9). The slope of the regression line decreased of about 15% moving from the relationship for the females with eggs in stage 1 (Fig. 9A) to that for the females in stage 2 (Fig. 9B). This difference could be related to the loss of eggs during the brooding period.

DISCUSSION

The size range of the collected individuals (10-29 mm CL) was similar to that reported in other studies carried out in the Mediterranean Sea (Guennégan *et al.*, 1992; Company & Sardà, 1997; Garcia-Rodriguez *et al.*, 2000; Colloca, 2002).

On the other hand, Santana *et al.* (1997), in the southeastern Atlantic Ocean (Canary Islands), observed greater maximum sizes, both for males (34 mm CL) and females (40 mm CL).

Sex ratio was shifted in favour of females, while Guennégan et al. (1992) and Garcia-Rodriguez et al. (2000) reported a proportion of sexes of around 50%. Colloca (2002) observed a sex ratio in favour of females between 100 and 200 m, around 1:1 or in favour of males between 200 and 250 m; at depths greater than 250 m, females were predominant. Such differences were probably due to the different bathymetric ranges investigated, tacking into account the size-depth relationship showed by this species (Company & Sardà, 1997).

A different *sex ratio* by depth was also observed in other species of the genus *Plesionika*, except *P. acanthonotus*; *P. gigliolii* shows higher proportion of females between 350 and 450 m, whereas females of *P. martia* prevail at depths greater than 450 m (Company & Sardà, 1997).



Fig. 3 - P. edwardsii. Percentage of females in the three maturity stages, by month.



Fig. 4 - P. edwardsii. Monthly proportion of ovigerous females (eggs in stage 1 and in stage 2) and non-ovigerous females.



Fig. 5 - P. edwardsii. Physiological size at first maturity.



Fig. 6 - P. edwardsii. Functional size at first maturity.



Fig. 7 - P. edwardsii. Percentage of adult females ($CL \ge 18.5 \text{ mm}$) in the three maturity stages, by month.



Fig. 8 - P. edwardsii. Average fecundity by size class, according to specimens sampled with embarks on a professional trawler or with experimental traps.



Fig. 9 - P. edwardsii. Relationship between egg number and carapace length, for females with eggs in stage 1 (A) and in stage 2 (B).

The seasonal pattern of female maturity observed in this study substantially agrees with that reported by Colloca (2002) for the central Thyrrenian Sea, Guennégan *et al.* (1992) for the north-central Thyrrenian Sea and by Company & Sardà (1997) for the Catalan Sea: mature females are present all year round, but they show a peak in spring. Ovigerous females have two periods of maximum presence, one in spring and another in summer. Little differences were reported by Garcia-Rodriguez *et al.* (2000) in the Levante Sea (Murcia), where the percentage of mature females was higher in autumn. A wider spawning season characterises *P. edwardsii* in tropical and sub tropical oceanic areas (Poupin *et al.*, 1990; Santana *et al.*, 1997).

A wide reproductive period seems to be a typical feature of species distributed on deep bottoms, where environmental conditions are costant (Tyler, 1988). Temperature is the main abiotic factor wich can determine egg incubation in crustaceans (Wear, 1974; Sastry, 1983).

The estimated physiological maturity size (18.5 mm CL) is close to those reported by Guennégan *et al.* (1992) and Colloca (2002) (18 and 17.9 mm CL, respectively), while the functional one (20.2 mm CL)

is slightly different from those reported by the same authors (18.4 and 21.8 mm CL, respectively).

As concerns fecundity, Guennégan *et al.* (1992) and Company & Sardà (1997) reported values of total fecundity lower than those obtained in the present work; they observed a maximum of 13,000 eggs per ovigerous female, whereas in this study the maximum value of fecundity was around 20,000 eggs for female. This probably depends to the sample size: in the present study eggs from 267 ovaries were counted, while the other authors worked on considerably smaller samples.

Results showed that sampling gear affects the estimates of fecundity: at the same size females collected with traps showed a number of eggs significantly higher than those of the females caught by trawling. As a matter of fact, trawl fishing is in general a high «stressing» sampling gear: specimens are subjected to several injuries during trawling, hauling, sorting on board, *etc.*, which also produce the loss of eggs from the pleopods during all the fishing operations.

In other species of crustaceans carrying eggs on pleopods lower fecundity estimates were obtained. Gramitto & Froglia (1980), Morizur *et al.* (1981), Mori *et al.*

(2001) reported a maximum number of eggs of about 10,000 for Nephrops norvegicus; Company & Sardà (1997) and Maiorano et al. (2002) estimated about 3,000 eggs for P. gigliolii and P. acanthonotus, 6-7,000 for P. heterocarpus, 11,000 for P. martia. These differences could be due both to the size and the habitat of the different species. P. edwardsii is, among pandalids, one of the biggest size species and less linked to the bottom. Many decapod species show strategies to minimize the egg loss during their development: in the females of P. edwardsii, during moult that come before copula, ovigerous setae and other modifications to keep eggs appear on pleopods; these expedients disappear with the next moult; eggs are fixed on pleopods by a liquid constituting a very resistant weave, which totally breaks up when larvae are released (Ceccaldi, 1966). Fecundity of P. edwardsii was proportional to animal size; the same was reported by other authors in other decapod species (Gramitto & Froglia, 1980; Morizur et al., 1981; Abellò & Sardà, 1982; Mori et al., 2004). A decrease of about 15% of the number of eggs during their development on the pleopods has been supposed; Guennégan et al. (1992) estimated a loss of about 35%. The main causes of egg loss are aborted development, mechanical injuries due the fishing operations, maternal cannibalism, predation and parasitism (Kuris, 1991).

P. edwardsii is a species of a certain commercial interest (15-25 euro/kg at the auctions); in spite of this, a targeted fishery is still absent and the species is only a by-catch of many trawl fisheries (De Ranieri, 2005). Only in some restricted areas *P. edwardsii* is object of a specifically devoted fishery, performed with nectobenthic traps (Gonzalez *et al.*, 1992; Guennégan *et al.*, 1992).

In order to perform a more targeted exploitation of this species also in Italian seas, the striped soldier shrimp has been object of various research projects based on the experimentation of new fishing gears (Colloca, 1999; De Ranieri, 2005; Sartor *et al.*, 2006). Traps seem to be the more promising fishing gear, to achieve the best compromise between commercial yields and sustainable exploitation of the resource.

In this context, reliable and accurate estimations of the main biological parameters are the basic information to implement a future management of the fishery targeted to this species.

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