Growth, size class frequency and reproduction of purple sea urchin, *Paracentrotus lividus* (Lamarck, 1816) in Bistrina Bay (Adriatic Sea, Croatia)

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This paper presents research data on the growth, size class frequency and reproduction of the sea urchin Paracentrotus lividus in Bistrina Bay, eastern Adriatic Sea coast, Croatia. The population was sampled on a monthly basis and from depths ranging between 0 and 3 m from the surface. Size class frequency of the test diameter showed that the population size ranges between 10 and 65 mm in the area with the main population in the class 30-45 mm. The von Bertalanffy growth parameters were found: $TD\infty=7.07$ cm, K=0.182, $t_o=-1.403$ years. The estimated longevity was found to be 15.06 years. The gonad somatic index annual distribution showed that the peak of reproduction occurs in April. The average GSI values range between 1.33±0.96% in September and 4.83±2.02% in April.

Key words: Paracentrotus lividus, growth, size class frequency, reproduction, Adriatic Sea

INTRODUCTION

Sea urchin gonads are intensively harvested in many parts of the world as a delicacy (LAW-RENCE, 2001). In times past, the sea urchin fishery was conducted by artisanal fishermen and the product was sold locally. However, the existence of certain markets in the world, mainly Japan, which today absorb large amounts of sea urchins increase the fishing pressure (YOKOTA, 2002). As a consequence, echinoid stocks had to be strictly managed in order to be protected (LESSER & WALKER, 1998; YOKOTA *et al.*, 2002). Increasing demand for sea urchin roe has led, in the last decade, to over-fishing of natural populations (CONAND & SLOAN, 1989; LE GALL, 1990). Several possible solutions have been tested; reseeding natural habitats with farmed juveniles (AGATSUMA & MOMMA, 1988; GOMEZ *et al.*, 1995); mariculture (SPIRLET *et al.*, 2000); raising sea urchins in immersed cages, alone (SPIRLET *et al.*, 2000; ROBINSON & COLBORNE, 1998) or in polyculture, with salmon notably (KELLY *et al.*, 1998); and finally land-based, closed-system echiniculture allowing control of each phase of the echinoid biological cycle (LE GALL & BUCAILLE, 1989; LE GALL, 1990; GROSJEAN *et al.*, 1998).

The sea urchin, *Paracentrotus lividus*, is a benthic echinoid that is distributed throughout the Mediterranean Sea and in the north-eastern Atlantic, from Scotland and Ireland to southern Morocco and the Canary Islands (BOUDOUR-ESQUE & VERLAQUE, 2001). It is typically a sub-

littoral species living from the mean low-water mark up to 10 to 20 m (GAMBLE, 1965; TORTON-ESE, 1965; ALLAIN, 1975; REGIS, 1978; HARMELIN *et al.*, 1980). Isolated individuals occur at depths down to 80 m (CHERBONNIER, 1956; TORTONESE, 1965). This sea urchin occurs mainly on solid rocks, boulders and in the meadows of seagrasses *Posidonia oceanica* and *Zostera marina* (MORTENSEN, 1927; TORTONESE, 1965; EBLING *et al.*, 1966; VERLAQUE, 1987). Its reproduction in the Mediterranean extends throughout the year probably due to beneficial environmental conditions.

Aspects of distribution and biology of the sea urchin *P. lividus* have been long studied (SALA *et al.*, 1998). Somatic growth of *P. lividus* in the field appears to be related to temperature, food quality and gonadal development (FERN-ANDEZ, 1996). The relationship between test diameter and corresponding age may be interpolated from various authors (BOUDOURESQUE & VERLAQUE, 2001.) None of the resulting growth curves account for the largest individuals > 7cm in diameter, which has been attributed to incomplete or incorrect field data.

The fact of the gonads being the edible part of the sea urchin has led to a large number of papers regarding gametogenesis and reproduction both in natural and in laboratory conditions (KLINGER, 1997; LAWRENCE *et al.*, 1997; McBRIDE *et al.*, 1997; BARKER *et al.*, 1998). EMLET (2002) yet, despite the huge number of ecological studies on the role of sea urchins, studies about fisheries and ecology are scarce. The purple sea urchin is potentially important as a local fishery resource for the tourist sector in Croatia. Presently, fishing is not intensive due to a lack of demand. This species is also a promising candidate for the emerging aquaculture industry of Croatia, especially today when diversification of production is obligatory. As far as the eastern Adriatic is concerned, there are no previous studies on the biology and reproduction of this species in the area which could be used in fishery management and intensive aquaculture trials.

The present paper aims to study the size class frequency, growth and reproduction of the coastal population of the purple sea urchin in Bistrina Bay, Croatia (Adriatic Sea).

MATERIAL AND METHODS

Description of the study area

The study area is Bistrina Bay on the Croatian coast of the Adriatic Sea (Fig. 1) located 6 km north of the town of Mali Ston (42°50N;17°42E). The bay enclosure stretches 3 km north into the mainland and it makes part of Mali Ston Bay. The width across the bay is 1 km and in some places slightly more. Mali Ston Bay is ramified and is positioned between the peninsula of Pelješac and the mainland. Maximal depth in the bay area is 29 m while maximal depth in Bistrina Bay is 9 m. The substrate is composed of soft mud. Numerous fresh water springs outflow in Bistrina Bay. Fresh water originating also from the Neretva River (north



Fig. 1. Map of the sampling area in Bistrina Bay in the eastern Adriatic, Croatia

side of Mali Ston Bay) along with the freshwater springs creates a moderately eutrophic ecosystem in the Bay. The bay is abundant with marine life, there are records of over 300 plankton species, more than 250 algal species, over 250 various benthic animals and 20 fish species (ŠPAN & ŠIMUNOVIĆ, 1981; BENOVIĆ & ONOFRI, 1981; LUČIĆ & ONOFRI, 1990).

Sampling and measurements

A total number of 1133 individuals of purple sea urchin, Paracentrotus lividus were collected from the area of Bistrina Bay, south-east Croatia (Adriatic Sea). The specimens were collected on a monthly basis from August 2002 to July 2003 in almost equal numbers per month (~ 100 specimens). The specimens were collected by hand and by self-constructed tool (netted basked on an iron stick) during cold months or by scuba diving from locations close to the coastline and at depths down to 3 m. In addition, seawater temperature and salinity was measured on site using portable digital instruments (WTW). Following their removal P. lividus samples were immediately taken to the laboratory where they were measured and dissected.

In the laboratory, the maximum diameter of each individual – excluding spines - was measured using analog calliper and weighed, using a digital OHAUS balance. The specimens were then dissected and the gonads extracted and weighed in order to estimate the gonad-somatic index (G.S.I., %). The gonad-somatic index was estimated using the standard equation:

$$GSI = \frac{Gonad \ weight, g}{Somatic \ weight, g} \cdot 100$$

Growth was studied in accordance with length-based methods for the test diameter (TD, in cm) of the individuals. Diameter-frequency histograms (DFD) were produced per month and for the pooled annual sample of all individuals based on length classes of 0.25 cm. A total of 23 diameter-classes between 1 and 6.5 cm were used to create the DFD histograms. Modal progression was analysed in accordance to the Bhattacharya method (BHATTACH-ARYA, 1967), a graphical method for the analysis of length-frequency histograms into separate normal distributions. The progression analysis produced age classes in the samples which were used to fit a von Bertalanffy growth equation in accordance with the Powell-Wetherall graphical method (POWELL, 1979; WETHERALL, 1986). The method provided estimates of TD ∞ and Z/K (Z – mortality and K – growth coefficient). The estimate of TD ∞ was then used in the ELEFAN I routine (GAYANILO *et al.*, 1989) as a starting point to estimate the K value based on the Rn criterion (GAYANILO & PAULY, 1997). Finally t_o (age of the individual when the diameter equals 0) value of the VBGF equation was estimated from the equation (PAULY, 1983):

 $\log(-t_0) = -0.3922 - 0.2752 \times \log TD_{\infty} - 1.038 \log K$

In addition to this, the diameter-frequency histogram modes were analysed based on the density function (McDONALD & GREEN, 1988) using the MIX software package. The method aims to the separate a frequency histogram into individual normal distributions based on the histogram density function. Similar results for $TD\infty$ and K were also obtained using the automatic scoring routine of ELEFAN I software as from the Powell-Wetherall method.

Longevity of the sea urchins was estimated using the approximation (PAULY, 1983):

$$Longevity = \frac{2.9957}{K} + t_o$$
RESULTS

Hydrographic data

Hydrographic data was measured at a depth of 2 meters and at the surface in the sampling area. The highest value for temperature was recorded at the surface in July 2003, measuring 28.3°C, while the lowest was recorded in January 2003, also at the surface, measuring 10.1°C. The average temperature throughout the course of the experiment was calculated at 18.4°C. There were no significant oscillations in temperature observed for the two measured strata. Salinity of the water column in the course of the experiment

ranged from the highest recorded in March 2003, at 37, to the lowest in October 2002, at 26.6, both at the surface level. The average salinity was calculated at 33.5. Significant oscillations in values between the two strata were recorded for the months of October and December.

Size class frequency distributions

The monthly diameter-frequency histograms are illustrated in Fig. 2. Bhattacharya analysis of modes showed that each sample included 2 to 5 modes. The overall population showed a great concentration of lengths within the classes 3.00-4.50 cm TD. The overall totals are summarised in Fig. 3.

Growth

Bhattacharya analysis of the samples produced 5 TD distinct modes. The statistics of these diameter modes are summarised in Table 1. Based on the Bhattacharya results, the VBGF coefficients were estimated (Fig. 4). TD ∞ was found to be 7.07 cm, K was found to be 0.182 (year⁻¹) and t_o was found to be -1.403 years. Longevity of the



Fig. 2. Monthly diameter-frequency distributions of samples of Paracentrotus lividus on the eastern Adriatic coast, Croatia



Fig. 3. Overall totals of diameter frequency distribution of Paracentrotus lividus on the eastern Adriatic coast, Croatia

sea urchins based on the above estimates, was determined at 15.06 years. The density function mathematical analysis (Fig. 4) shows different results for age classes 2 and 3 (deviations do not overlap; chi-square goodness of fit = 86.135, df=13, p<0.001). In particular, the estimated average diameters for age classes 2 and 3 are shown to be lower than those obtained by Bhattacharya analysis.

Reproduction pattern

The monthly distribution of gonad-somatic index is illustrated in Fig. 5. The maturation of the gonads is gradual and peaks in April. Shows a very smooth monthly oscillation pattern in the study area. The average GSI values



Fig. 4. Von Bertalanffy (VBGF) growth curve of Paracentrotus lividus in the eastern Adriatic, Croatia

range between 1.33±0.96% in September and 4.83±2.02% in April.

According to the diameter frequency histograms (Fig. 2), the younger individuals (< 2 cm) appear in the researched area in May, June, July and August with higher numbers in June. Individuals larger than 5.75 cm are scarce and appear in the samples of April, August and September only.

 Table 1. Von Bertalanffy (VBGF) growth curve parameters and related statistics of Paracentrotus lividus in the eastern Adriatic, Croatia

Age class	Bhattacharya method			MIX method		Number of
	Average TD (cm)	±SD	Separation index	Average TD (cm)	±SD	specimens
1	1,28	0,26	-	1,06	0,15	16
2	2,55	0,21	5,40	1,72	0,27	56
3	3,80	0,39	4,17	2,57	0,38	849
4	4,56	0,32	2,14	3,76	0,55	192
5	5,52	0,12	4,36	5,37	0,81	20
					Total :	1133

Length-weight relationship

The length-weight relationship was found to be significant (Fig. 6). Statistical analysis of the equation showed that there is a high variation of the length-weight data pairs. ANCOVA analysis using the months as a categorical dummy variable (1-12) showed that there is no significant variation of the relationship between months (p = < 0.0001). The relationship was found to be:

 $Weight(g) = 1.894[Diameter, cm]^{1.849}, r^2 = 0.679, std.error = \pm 8.04g$



Fig. 5. Gonad-somatic index distribution of Paracentrotus lividus in the eastern Adriatic, Croatia

DISCUSSION

From the comparison of VBGF parameters obtained for various stocks in the Mediterranean (Table 2), it is obvious that the urchins in the eastern Adriatic reach a higher size than in other areas of the Mediterranean but they show slower growth (low K value). Both facts indicate that the fishery of the species is not developed as in other areas so that the individuals are allowed to grow to bigger sizes. It is known that the species in various habitats exhibits a large variety of diameter sizes and corresponding ages: an individual with 40 mm diameter on the Atlantic coast of Europe can be 2.5 years old while a similar sized individual in the western Mediterranean can be as old as 4 years (SELLEM et al., 2000; ALLAIN, 1978). REGIS (1979) working in Marseille (France) reported a individual of 42 mm diameter which was 11 years old while TURON et al. (1995) observed in Spain a individual of 40 mm which was over 5 years old.

A difference was found between the modal analysis of the diameter-frequency histogram between the Bhattacharya method and the density function method. The results from both methods are acceptable since the overall statistics show significant fitting to the raw data. However, from the other results and the literature regarding the TD ∞ in the various Mediterranean regions, it is obvious that the Bhattacharya results are consistent with those of other authors.



Fig. 6. Length-weight relationship of Paracentrotus lividus in the eastern Adriatic, Croatia (dashed lines: confidence limits 95%)

The density function results show that the VBGF curve reaches a maximum that is higher than the maximum obtained from the Bhattacharya method ($TD\infty=7.07$ cm). The Bhattacharya method provides results close to those reported in the literature.

Gonad somatic index shows a typical fluctuation with a single peak in April. Based on the gonad observations in the samples, it is evident that reproduction of the species occurs over a period from March to July. P. lividus is exposed to a wide range of climatic conditions and light regimes. Authors who have examined the spawning cycle of P. lividus in the Mediterranean have observed either one or two spawning events, whose timing varies among regions, at different spatial scales suggesting that, for a given site, these spawning events are closely linked to the energy sources availability (algae in particular) and, indirectly, to the prevailing hydrodynamic conditions (CHIANTORE et al., 2008). In Ireland, both single and double spawning (in Spring and late Autumn) were reported (CRAPP & WILLIS, 1975). The results of the present study support one spawning mode per year for the Adriatic Sea. It is known that gonad growth depends on food quality and availability (KLINGER et al., 1988; LAWRENCE et al., 1997; McBRIDE et al., 1997; BARKER et al., 1998) and that it is hindered by high summer temperatures (BYRNE, 1990; SPIRLET et al., 2000). It seems that, in the case where a double spawning mode is

Area	TD∞	К	t _o	Distribution (mm)	Reference
East Atlantic (UK)	61.80	0.222	-0.716	7.50-62.40	Allain, 1978
North-west Mediterranean	58.96	0.268	-0.112	20.72-51.5	Sellem <i>et al.</i> , 2000
South-west Mediterranean	52.83	0.689	-0.015	22.90-62.40	Sellem <i>et al.</i> , 2000
Adriatic Sea	70.70	0.182	-1.403	32.50-45.00	Present study

Table 2. Comparison of growth curve parameters of Paracentrotus lividus in various geographic regions

observed, this should be expected in eutrophic geographic areas with low water temperatures (LOZANO *et al.*, 1995). In the case of Bistrina lagoon, feed for the urchins is not limited due to the eutrophic nature of the water and therefore, the one spawning mode observed for this population could be explained by the high summer temperatures ($\geq 28^{\circ}$ C) which do not allow for the ripening of the gonads in time for a late fall reproduction. Regardless of the number of annual peaks, spawning can occur nearly yearround, although usually at very low levels (BOUDOURESQUE & VERLAQUE, 2007).

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Rast, učestalost dužinskih klasa promjerskih veličina i reprodukcijski ciklus hridinskog ježinca (*Paracentrotus lividus*) (Lamarck, 1816) iz uvale Bistrina, Južni Jadran

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SAŽETAK

U ovom radu prikazani su rezultati istraživanja populacije hridinskog ježinca, *Paracentrotus lividus* iz uvale Bistrina, Malostonski zaljev. Istraživani parametri su: rast, učestalost dužinskih klasa i reprodukcijske značajke. Uzorkovanje je obavljano jednom mjesečno, ronjenjem do dubine od 3 metra. Učestlost dužinskih klasa kućice kolebala je između 10 i 65 mm, gdje je glavnina populacije unutar razreda, 30-45 mm. Izračunati su Von Bertalanfijevi parametri rasta: TD ∞ =7.07 cm, K=0.182, t_o=-1.403 godine. Procijenjena najveća starost je 15.06 godina. Godišnja raspodjela gonadosomatskog indeksa pokazala je maksimalnu vrijednost u travnju, a GSI je kolebao između 1.33±0.96% u rujnu do 4.83±2.02% u travnju.

Ključne riječi: *Paracentrotus lividus,* rast, učestalost dužinskih klasa, reprodukcijski ciklus, Jadransko more