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Temporal variability of spawning in the sea urchin *Paracentrotus lividus* from northern Spain

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ABSTRACT

Reproductive cycle of the sea urchin *Paracentrotus lividus* was investigated from an intertidal population of Asturias, northern Spain. Sea urchins were collected monthly from March 2006 to August 2008, and during spring 2009 (March–June). In order to assess the influence of environmental factors on temporal pattern of sea urchin reproduction, photoperiod, sea temperature and phytoplankton abundance were monitored. Gonad index and histological examination of the gonads revealed a well-defined annual pattern with a single spawning period, which begins between May and June. Higher GI in the first months of the year corresponds to gonads in growing (winter) and maturation stage (spring). The index drop indicated the onset of spawning stage (late spring–summer) that ended with the minimum values of the year in August. In September, the GI began to increase again with gonads in spent and recovery stages (autumn). An advance from the minimum temperatures, with a colder autumn and a warmer winter, resulted in a slight advancement of gametogenesis during a breeding season. Nevertheless, the pattern of gonadal development was similar between years. Minimum day length indicated the onset of gametogenesis that lasted the winter months. Meanwhile, maximum day length marked the beginning of spawning that extended during the summer. Inter-annual variability in the onset of spawning was observed. In Asturias, environmental conditions that triggered spawning of the sea urchin were temperatures above 15 °C, around the longest days of the year and the spring phytoplankton bloom. The results provide a baseline for resource managers to evaluate and predict differences in reproduction, which could be useful in the management of the fishery.

1. Introduction

The purple sea urchin *Paracentrotus lividus* (Lamarck 1816) is widely distributed throughout the Mediterranean Sea and the north-eastern Atlantic, from Scotland and Ireland to southern Morocco, including the Azores and Canary Islands (Boudouresque and Verlaque, 2013). This species is the most important commercial echinoid in Europe and it is, or has been, intensively harvested in countries such as Ireland, France, Italy, Greece, Portugal and Spain (Andrew et al., 2002). Consequently, the sea urchin stocks have decreased drastically and various cases of overexploitation and collapse have been documented (Byrne, 1990; Andrew et al., 2002; Fernández-Boán et al., 2012; Couvray et al., 2015). Its economic importance is due to the gonads that are considered a highly valued product. Considering that sea urchin fisheries target the extraction and marketing of the gonads, it is essential to understand sea urchin reproductive processes to determine the best season for harvesting as well as the potential productivity of the fishery (Ouréns et al., 2011).

Several studies on this species have investigated its reproductive cycle in a wide range of regions, determining parameters such as the spawning period and the effect of environmental variables on the gonad development (Byrne, 1990; Catoira, 1995; Guettaf et al., 2000; Shpigel et al., 2004; González-Irusta et al., 2010; Tenuzzo et al., 2012; Loi et al., 2015; de Casamajor et al., 2017). The reproductive cycle *of P. lividus* is controlled by endogenous and exogenous factors. Food availability and quality (Lozano et al., 1995; Sánchez-España et al., 2004), hydrodynamism (Sellem and Guillou, 2007; Gianguzza et al., 2013), phytoplankton blooms (López et al., 1998; González-Irusta et al., 2010), water temperature and photoperiod (Byrne, 1990; Spirlet et al., 1998, 2000) are some of the most important environmental factors responsible for gonad growth.

Analysis of temporal changes in gonad index (GI) is typically used to study the reproductive cycle and the spawning period (Ouréns et al., 2011). But as sea urchin gonads serve also as organs of nutrient storage, stressful situations (starving, diseases,

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storms…) can also affect gonad mass. The energy accumulated in the gonads can be allocated to other compartments, which leads to variations in the GI that are not related to reproduction (Lozano et al., 1995). Therefore, histology is the most reliable tool for determining the reproductive cycle of this species.

Ouréns et al. (2011) synthetized the data from 52 publications on the gonad growth and spawning seasonality of *P. lividus* throughout its geographic range (excluding the Canary Islands). The results revealed clear differences between the Atlantic and the Mediterranean populations. In the Mediterranean, the GI trend is very irregular and it is difficult to identify the reproductive pattern. The authors suggested that the higher temperatures of the Mediterranean Sea could favor successive spawnings throughout the year, which would explain the lower gonad production in that basin. However, the Atlantic populations showed a clear GI trend and a higher gonad production. Lower temperatures could favor the storage of nutrients and the spawning period would occur when the conditions are most appropriate. In addition, in the Atlantic, the gonad production exhibits a latitudinal gradient which increase toward the north in both intertidal and subtidal populations.

On the northern coast of Spain, *P. lividus* constitutes an important fishery resource and the western regions of Galicia and Asturias support intensive commercial activity (González-Irusta, 2009). Nowadays, Galicia is the main fishery for this species of sea urchin, landing annually about 700 tons (Ouréns et al., 2015). The management legislation is developed and includes annual exploitation plans for this specific resource. Harvesting occurs mainly in subtidal areas, although the exploitation has expanded to the intertidal in some locations. In Asturias, there is a strong tradition of consumption and a high demand for sea urchins, often satisfied with urchins from the neighboring region. However, the sea urchin fishery in our region has not been as developed as in Galicia and professional and recreational fishers operate under an open access scheme. Harvesting occurs only in intertidal areas, where gonadal production is lower (Ouréns et al., 2011), and the maximum annual production was about 70 tons in the last decade (https://tematico.asturias.es/dgpesca/). However, a decrease in the subtidal populations was observed, especially on the west coast of Asturias (Álvarez Raboso, 2006). Thus, a closed season (from April 15th to December 15th) was included in the regulatory measures since 2013. Despite of this measure, the worrying situation of the *P. lividus* populations has led the regional government to implement in 2016 a total ban on sea urchin harvesting, during at least two years.

Despite the importance of this species in Asturias, no published recent studies on the reproductive cycle are available (Haya de la Sierra, 1990). The main objective of this study was to determine the temporal variability of the reproductive cycle of *P. lividus* from an intertidal population on the west coast of Asturias. The state of gonad development was assessed through gonad index and histological examination of the gonads. The influence of environmental variables on temporal variability of reproduction and spawning was also examined during four breeding seasons. The aim of this work is to contribute to the knowledge of this species in our region, in order to develop sustainable management strategies.

2. Materials and methods

The study area is located on the western coast of Asturias, northern Spain (Fig. 1). Samplings were performed on *P. lividus* adults from an intertidal population, La Punta de la Cruz (43°33′25″ N, 7°01′43″ W). In this area, the shore is rocky and exposed, and the pools are inundated daily. The pools are covered with calcareous algae and the sea urchins in the tide pools were found in dense aggregates.

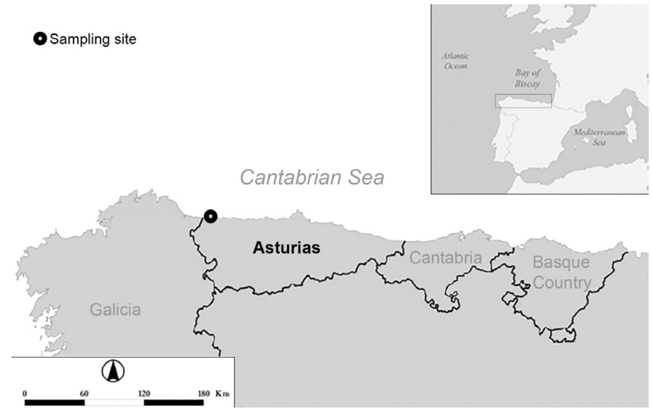

Fig. 1. Location of the sea urchin sampling site on the western coast of Asturias (northern Spain).

Twenty–thirty individuals with a diameter larger than 40 mm were collected monthly at low tide from March 2006 to August 2008, and during spring 2009 (March–June). The sea urchins were transported alive and immediately processed in the laboratory to avoid spawning. Test diameter and height (excluding spines) of each specimen were measured using a Vernier calliper (±0.1 mm). After 10 min of drainage on a filter paper, sea urchins were wet weighed (±0.1 g) and dissected and the five gonads were extracted and wet weighed (±0.1 g) separately. Sex was determined by examining the fresh gonads. The gonad index (GI) was calculated as the ratio of the gonad wet weight to the whole-body wet weight given as a percentage, which is the most common form to express this parameter found in the literature (Ouréns et al., 2011).

$$GI = [\text{gonad wet weight (g)}/\text{whole animal wet weight (g)}] * 100.$$

On each sampling date, ten gonads (from five females and five males) were collected and preserved in Davidson fluid for histological examination. Histological analysis was conducted on the middle section of the gonad since the gametogenic condition is homogeneous throughout the organ (Byrne, 1990). Samples were dehydrated in ethanol, embedded in paraffin wax, sectioned (7 μm) and stained with haematoxylin and eosin. The reproductive state of the urchins was then assessed following the descriptions of Byrne (1990) who established six stages of ovarian and testis growth: recovery (I), growing (II), premature (III), mature (IV), partly spawned (V) and spent (VI).

In addition, a monthly record of the sea surface temperature and phytoplankton abundance was obtained during the sampling period. Sea temperature and chlorophyll-a concentration data were measured from the surface layer of the water column and was provided by Autoridad Portuaria de Gijón. The sampling site of these parameters is located east of the sea urchin population (43°37′19″ N, 5°44′53″ W).

Statistical analyses were performed mainly by one-way ANOVA and Dunnett's T3 test. Student's *t*-test was also used to compare different groups with only two levels. Prior to performing parametric tests, the assumption of normality was tested by Kolmogorov–Smirnov test.

3. Results

A total of 943 sea urchins were sampled throughout the period of study, being 457 females, 468 males and 18 specimens of uncertain sex (most of them occurred during spent and recovery stages, between October and December). The sexual ratio was close to 1:1, being slightly displaced towards the males (1.02). All the

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