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Geographical implications of seasonal reproduction in the bat star *Asterina stellifera*

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ABSTRACT

The reproductive cycle and environmental cues that regulate gonad production in *Asterina stellifera* were studied from April 2009 to April 2011 in a rocky subtidal habitat at the southernmost limit of its distribution (Mar del Plata, Argentina). The geographic variation in reproductive traits between latitudinal range limits was analyzed. The gonadal and pyloric caeca weight varied with sea star size and time in both sexes. Despite a previous study which suggested the absence of recruitment in a 4 year period, our data of the same period demonstrated that spawning happens from early spring to early summer. The gonad and pyloric caeca weight did not show an inverse relationship, this suggested that there is no dependence on energy transfer between the organs and that the bat star presented a good nutritional state. Seawater temperature appears to be the variable explaining gonad proliferation at the range limits of *A. stellifera* distribution. Furthermore, differences in sex ratio, oocyte production, oogenesis duration and capability of energy transformation into ova were found between range limits.

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1. Introduction

Seasonal patterns of gamete production followed by synchronized spawning periods have been documented for many asteroid species (Byrne et al., 1997; Chen and Chen, 1992; Chia and Walker, 1991). At high latitudes, sea stars show a marked seasonality in reproduction (Stanwell-Smith and Clarke, 1998) that fades in mid-latitudes (Rubilar et al., 2005; Ventura et al., 1997) and is not evident in tropical species (Guzmán and Guevara, 2002).

Many studies provide evidence about the relationship between reproductive cycles of sea stars and different environmental factors such as seawater temperature, day-length, pluvial precipitation and food supply (Benítez-Villalobos and Martínez-García, 2012; Benítez-Villalobos et al., 2007; Stanwell-Smith and Clarke, 1998; Tyler and Pain, 1982a, 1982b; Tyler et al., 1984). Understanding the fluctuation of these factors is important to elucidate the coupling between reproductive patterns and the environment (Mercier and Hamel, 2009).

Asterina stellifera (Möbius, 1859) occurs in shallow waters from Cabo Frio, Brazil (23°S, 42°W) to Mar del Plata, Argentina (35°S, 56°W) in the southwest Atlantic (Clark and Downey, 1992). This bat star is one of the few asteroid species in the rocky coast of the South Atlantic Ocean and there is little published information on the life history of this species. *A. stellifera* is an omnivorous generalist predator, and modifies the abundance of other invertebrates and algae in subtidal marine communities (Farias et al., 2012).

A declining density has been reported for this species at the northern and central areas of its western latitudinal distribution (Calil et al., 2009) without an evident cause. For this reason, it was included in the Brazilian Red List of Endangered Species (MMA, 2004). The construction of coastal break waters along the coast of Argentina led to a decrease in abundance of intertidal populations in recent decades (Roux, 2004). In contrast, Farias et al. (2012) reported that the subtidal population of *A. stellifera* at Mar del Plata recently reached a high unexpected abundance despite the absence of recruitment during their four year study. Studies dealing with latitudinal interpopulation differences in abundance and life history traits of marine benthic invertebrates are very scarce (Fenberg and Rivadeneira, 2011; Rivadeneira et al., 2010). Numerous studies have proposed several factors limiting geographic ranges of species, e.g., resource availabilities and physiological tolerances. Those tolerances may be forced in the limit of the geographic range; thus species may not persist in areas where environmental demands exceed these (Spicer and Gaston, 1999). Combinations of abiotic and biotic factors probably modify population traits such as abundance, reproduction cycles, fertility, larval survival and recruitment.

Understanding the population dynamics and the reproductive biology of the endangered bat star *A. stellifera* is important to elucidate potential causes of the scarce recruitment that was reported at range limits (Farias et al., 2012; MMA, 2004). In this study, we evaluate whether the Argentinian population of *A. stellifera* is sexually active and the geographic variation in reproductive traits at the distribution

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limits. For this purpose, we analyze the reproductive cycle and gametogenesis of *A. stellifera* at the southern limit of its distribution and we re-analyzed those data of reproduction previously published (Carvalho and Ventura, 2002) at the northern limit of its geographical distribution. Furthermore, we analyze the coupling of reproductive parameters with environmental factors at distribution range limits.

2. Materials and methods

2.1. Study area and environmental parameters

From April 2009 to April 2011, about 15 specimens of *A. stellifera* were randomly collected each month by SCUBA diving inside Mar del Plata port, Argentina (38°02′S, 57°31′30″W) (Fig. 1). The habitat is a rocky sublittoral consisting of walls of big orthoquartzite blocks limiting the harbor, and boulders of orthoquartzite rocks surrounding internal and external breakwaters. The breakwaters are surrounded by a fine-grained muddy bottom that limits individual's dispersion. Depth in the sampling area ranged between 6 and 8 m. Environmental factors that potentially influence the gonad periodicity (seawater temperature, salinity, day-length and cumulative precipitation) were obtained from official agencies. Seawater temperature and salinity from the sampling area were provided by the National Institute of Fisheries Research and Development (INIDEP). Average day-length was obtained from the web page of the Naval Hydrographic Service (http://www.hidro.gov.ar/observatorio/sol.asp), and precipitation was taken from the Mar del Plata Aerodrome database. Monthly means were calculated for all these parameters.

2.2. Reproductive cycle

Specimens were preserved in 10% formalin for at least 72 h. The largest radius of each specimen (center of disk to arm tip) was measured to the nearest 1 mm using a digital caliper. Water was blotted off by placing each individual on paper toweling for approximately 20 min and then weighted to the nearest 1 mg (drained wet weight). Gonads and pyloric caeca of the five arms were dissected out, weighted to the nearest 0.001 mg on a digital scale and stored in 70% ethanol.

Gonads were dehydrated in ascendant ethanol dilutions, cleared with xylol and embedded in paraffin wax. Tissues were sectioned at 7 μm and stained with hematoxylin and eosin. The gonad cycle was divided into five stages based on the frequency of cell types and size and shape of acinus, based on an adaptation of the scale by Byrne (1992), Byrne et al. (1997) and Carvalho and Ventura (2002). Individual sex was determined by examining histological gonad sections of each animal.

In order to analyze gametic growth and proliferation, oocyte size frequency distribution was constructed by measuring the diameter of at least 100 oocytes per individual to the nearest 1 μm. Only oocytes with visible nucleus were measured. The relative oocyte area (area of all oocytes present in an ocular field of optical microscope as a percentage of the field area) was estimated for each sample as: [mean number of oocytes per field/total area of a field] × 100.

2.3. Data analysis

Sex ratio was estimated considering the total sample over the study period. Statistically significant difference from the expected 1:1 sex ratio was tested using goodness of fit (G) test (Sokal and Rohlf, 1995).

A power function ($y = ax^b$) was fitted for organ wet weight (gonads and pyloric caeca) and sea star radius. To evaluate differences in gonad (GW) and pyloric caeca (PCW) wet weight between Brazilian and Argentinian populations, an ANCOVA analyses was performed after logarithmic transformation of both variables. Brazilian data was recorded by Carvalho and Ventura (2002) from Cabo Frio (Brazil) population.

Both *A. stellifera* GW and PCW are allometrically related (Fig. 2), i.e. the variables do not vary as a fixed proportion of the measure of body size (Packard and Boardman, 1988, 1999). Several studies had demonstrated that the use of a gonadosomatic index does not eliminate the relationship between individual's organ weights and body

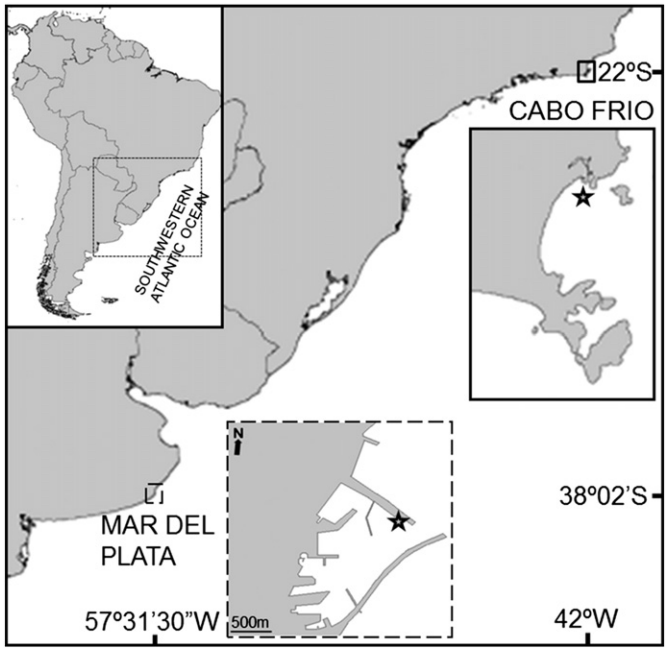

Fig. 1. Map showing *Asterina stellifera* western geographic range and sampling site in Mar del Plata (Argentina) and Cabo Frio (Brazil).

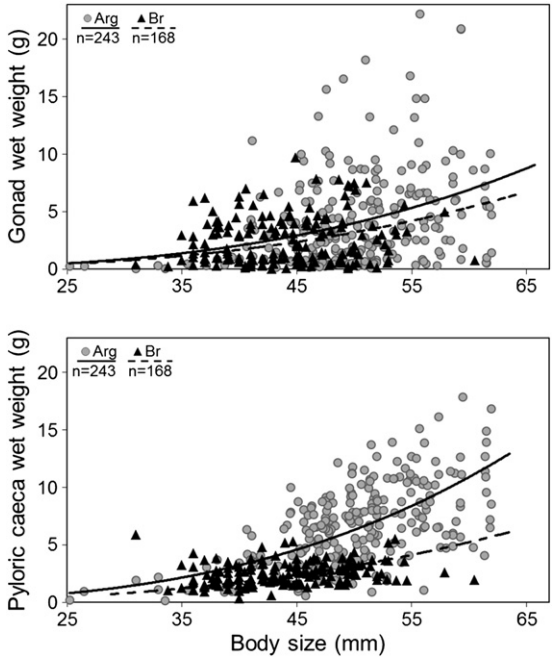

Fig. 2. *Asterina stellifera.* Relationship of gonad wet weight (g) and pyloric caeca wet weight (g) with radius (mm) between populations at the distribution range limits, Argentina (Arg) and Brazil (Br). The lines correspond to adjusted power function. Sample sizes of each data set are given.

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