



## ORIGINAL ARTICLE

# Environmental effects on the reproduction and fecundity of the introduced calcareous sponge *Paraleucilla magna* in Rio de Janeiro, Brazil

Emilio Lanna<sup>1,2</sup>, Rodolfo Paranhos<sup>3</sup>, Paulo C. Paiva<sup>2</sup> & Michelle Klautau<sup>2</sup>

1 Departamento de Biologia Geral, Instituto de Biologia, Universidade Federal da Bahia, Salvador, BA, Brazil

2 Departamento de Zoologia, Instituto de Biologia, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

3 Departamento de Biologia Marinha, Instituto de Biologia, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

**Keywords**

Calcarea; life history evolution; population biology; Porifera; temperature.

**Correspondence**

Michelle Klautau, Universidade Federal do Rio de Janeiro, Instituto de Biologia, Departamento de Zoologia – 21941-902, Rio de Janeiro, Brazil.  
E-mail: mklautau@biologia.ufrj.br

Accepted: 24 June 2014

doi: 10.1111/maec.12202

**Abstract**

The calcareous sponge *Paraleucilla magna* (Porifera, Calcarea) has been the subject of several studies in the last decade. It was first described along the Brazilian coast, where it is considered cryptogenic, and was subsequently found in the Mediterranean, where it is considered invasive. The wide artificial distribution of this species allows us to compare different aspects of the biology of an introduced species in different locations. Here, we analysed the effects of selected environmental parameters on the reproductive dynamics of *P. magna* in Rio de Janeiro (Brazil) over 18 months and compared our results with those obtained for the same species in the Mediterranean Sea. Specimens were collected monthly and analysed through histological methods. The density of reproductive elements in each month was calculated, and the effects of environmental parameters (photoperiod, precipitation, temperature, phytoplankton and bacterioplankton) were analysed using a regression tree analysis. *Paraleucilla magna* was reproductive throughout the study period. The densities of the reproductive elements (oocytes, embryos and larvae) showed no seasonality, and this species presented one of the highest reproductive efforts documented to date in the phylum Porifera ( $99.0$  oocytes  $\cdot$  mm<sup>-3</sup>;  $89.0$  embryos  $\cdot$  mm<sup>-3</sup>;  $319.0$  larvae  $\cdot$  mm<sup>-3</sup>). The main environmental parameters related to the reproduction of *P. magna* were temperature, photoperiod and bacterioplankton. Temperature was the main driver associated with the densities of oocytes and embryos, while bacterioplankton was the main driver of larvae (positive relationships). In Rio de Janeiro, larvae were present and continuously released. This strategy is different from that observed in the Mediterranean, where a larger larval output was observed but only during the summer months. Our results show that *P. magna* is a species with a strong invasive potential, considering its high and continuous reproductive effort. This high fecundity stimulated by high temperatures may be a key factor contributing to the growth of *P. magna* populations and its invasion of new areas.

**Introduction**

The calcareous sponge *Paraleucilla magna* Klautau *et al.* 2004; was first observed in Brazil in the late 1980s and very rapidly became one of the most abundant sponges in Rio de Janeiro (Klautau *et al.* 2004). As this sponge

was a species new to science, its type locality became Rio de Janeiro. However, soon after it was described, *P. magna* was recognized in the Mediterranean Sea, where it had been present since the 1970s according to fishermen (Zammit *et al.* 2009; Gravili *et al.* 2010; Pierri *et al.* 2010; Occhipinti-Ambrogi *et al.* 2011; Guardiola

*et al.* 2012; Cvitković *et al.* 2013), and then in the Eastern Atlantic Ocean (Agell *et al.* 2012). In Brazil, in addition to Rio de Janeiro state, this species has been found in São Paulo and Santa Catarina states.

The abundance, continuous spreading and unknown origin of this species have made it a model for several studies addressing ecology (Monteiro & Muricy 2004; Pierrri *et al.* 2010; Cavalcanti *et al.* 2013; Padua *et al.* 2013a,b; Batista *et al.* 2014), symbiosis (Turque *et al.* 2010), biomineralization (Rossi *et al.* 2014) and population genetics (Guardiola *et al.* 2012). The reproduction and embryology of *P. magna* are of particular interest (Lanna *et al.* 2007; Lanna & Klautau 2010, 2012; Longo *et al.* 2012; Padua *et al.* 2013a). This species is a hermaphrodite, producing oocytes and sperm cells simultaneously in the choanosome (Lanna & Klautau 2010). The embryos develop in the mesohyl, and in later stages (after inversion), the larvae are maintained in an epilarval trophocyte epithelium until ripening and subsequent spawning (Lanna & Klautau 2012). The periodicity of the reproduction of *P. magna* appears to vary according to the locality and even the year. Lanna *et al.* (2007) and Guardiola *et al.* (2012) showed that the reproduction of this species is restricted to the warmer months of the year in Rio de Janeiro and in Spain, respectively, while Longo *et al.* (2012) and Padua *et al.* (2013a) observed continuous reproduction throughout the year in Italy and Rio de Janeiro, respectively, with peaks during summer.

The discontinuous distribution of this species, its presence mainly near port areas and its continuous and fast spreading have called attention to the possibility of bioinvasion. In the Mediterranean Sea, *P. magna* is considered invasive, as it causes negative impacts for mollusk farmers (Longo *et al.* 2007), and new records are continuously being reported (see Cvitković *et al.* 2013). In Brazil, following the classification of introduced species presented by Geller *et al.* (2010), *P. magna* is cryptogenic, as its origin remains unknown. Along this coast, the populations of *P. magna* do not appear to cause any problems to the environment or to the economy (Cavalcanti *et al.* 2013; Padua *et al.* 2013a).

Although biological introductions can irreversibly alter the invaded community, leading to extinction of native species and economic losses in some cases (Geller *et al.* 2010), these invasions represent a unique opportunity for studying a number of aspects of species biology, such as the influence of the environment. Several hypotheses have been proposed to explain the success or failure of bioinvaders, but three factors have been repeatedly indicated: (i) the life history of the species; (ii) environmental health; and (iii) latitude (Ruiz & Hewitt 2002). The presence of the opportunistic species *P. magna* (r-strategist, Longo *et al.* 2012) in the Mediterranean (Italy – 40 °N)

and in Rio de Janeiro (Brazil – 23 °S) provides an excellent opportunity to increase our knowledge of the behavior of a species in different environments.

In sponges, the onset of the reproductive period has been related mainly to seawater temperature (*e.g.* Ereskovsky 2000; Maldonado & Riesgo 2008; Riesgo & Maldonado 2008), suggesting that each species has an optimal temperature at which to reproduce. Unfortunately, as few species of sponges exhibit wide spatial distributions, little is known about the influence of temperature and other environmental parameters on the reproduction of the great majority of species (Fell 1976). For this type of study, introduced species represent good models. In fact, there is an urgent need to improve our understanding of the species-specific relationships among reproduction, temperature and other environmental parameters because of the increasing number of introduced species (Olive 1995; Przeslawski *et al.* 2008; Riesgo & Maldonado 2008; Mercier & Hamel 2009).

Therefore, in the present work, we investigated the reproductive dynamics of a population of *P. magna* in Rio de Janeiro and the possible effects of different environmental parameters on the fecundity of this species. Based on previous results, we hypothesized that there is seasonality in the reproduction of this calcareous sponge in Rio de Janeiro and that the environment drives this seasonality and the reproductive output. Moreover, we compared our results with those of Longo *et al.* (2012) for a population of *P. magna* in the Mediterranean Sea with the aim of understanding the patterns and processes of the bioinvasion of this species in marine ecosystems.

## Material and Methods

### Study area

This study was carried out at Vermelha Beach, Rio de Janeiro, Brazil (22°57' S, 43°09' W). This beach is limited by two large rocky shores, located close to the mouth of the eutrophic Guanabara Bay (Fig. 1). The freshwater input is low, being limited to precipitation and water run-off from surrounding mounts (Lanna *et al.* 2007). The climate in the region is humid tropical, with a seasonal pattern of heavy summer rains (December to March) and a comparatively dry winter (July–August) (INMET 2008). The superficial seawater temperature varies between 19 and 29 °C, with an annual average of 23.1 (±2.9) °C (Albuquerque & Genofre 1999).

### Histology and quantification of reproductive elements

In this study, we collected fragments measuring 5 mm<sup>3</sup> from the base and top (Fig. 2a) of 10 specimens of



**Fig. 1.** The sampling site of *P. magna*, Vermelha Beach (arrow), is found close to the mouth of the eutrophized Guanabara Bay in the Rio de Janeiro State, Brazil.

*Paraleucilla magna* chosen at random on a monthly basis from September 2006 to March 2008. To perform the identification and quantification of reproductive elements [oocytes, embryos (a category comprising all stages between the first cleavage and the stomoblastula) and larvae], all fragments were fixed in Bouin's fixative solution, decalcified and embedded in paraffin. Thin sections ( $5\ \mu\text{m}$ ) were mounted on glass slides and then stained with Harris' hematoxylin and eosin [see Lanna & Klautau (2010) for more details on the histological procedures].

To quantify reproductive elements and establish the reproductive dynamics of this species, a reticulated coverslip with  $4\text{-mm}^2$  squares was positioned above the sections, and the reproductive elements were visually counted (at  $100\times$  magnification). We counted three squares per fragment (total area of  $12\ \text{mm}^2$ ), examining a total of six squares (total area of  $24\ \text{mm}^2$ ) per specimen.

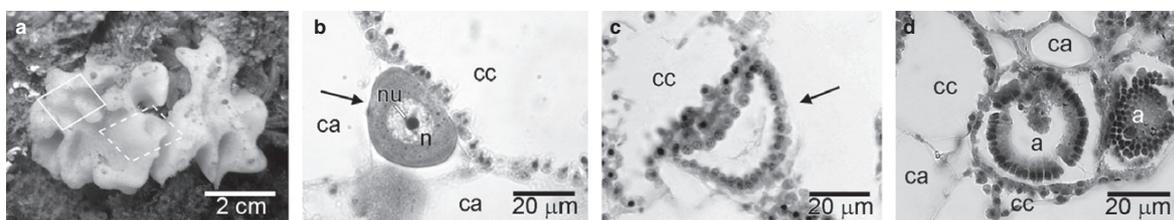
We then calculated the average reproductive elements in the squares per specimen, followed by the population average for each month as the average of the values obtained for all specimens collected in each month. Unless otherwise indicated, the data obtained in this study are presented as averages  $\pm$  SDs.

#### Sampling of environmental parameter data

Data on the investigated environmental parameters [photoperiod, precipitation, superficial seawater temperature (hereafter referred to as 'temperature'), phytoplankton abundance and bacterioplankton abundance] were obtained at or adjacent to Vermelha Beach. All of the data were collected monthly from September 2006 to March 2008, except for temperature, which was obtained only after October 2006.

Photoperiod values were generated by calculating the average daily difference between sunrise and sunset obtained from the Brazilian National Institute for Space Research (CPTEC-INPE 2008) for all days of the month. Monthly precipitation data were obtained from the website of AlertaRio (2008) and are expressed as accumulated millimeters of rain per month. Temperature was measured using a calibrated thermometer three times per week in the vicinity of the sampling site (2 km southward), and the average value was calculated for each month.

Soon after collecting the sponge tissue, a sample of water from Vermelha Beach was collected with a 500 ml polypropylene bottle. It was kept refrigerated ( $4\ ^\circ\text{C}$ ) and promptly brought back to the laboratory. Phytoplankton abundance (chl *a* and its degenerated form, phaeophytin) analysis was performed *via* vacuum filtration through cellulose membrane filters (Millipore HAWP  $0.45\ \mu\text{m}$ ). The filters were extracted with 90% acetone, and spectrophotometer readings and calculations were performed according to Parsons *et al.* (1984). Bacterioplankton abundance was determined after nucleic acid staining with the fluorochrome SYTO13 (Invitrogen, ref. S-7575)



**Fig. 2.** General aspects of the reproduction of *Paraleucilla magna*. (a) Adult specimen of *P. magna* on the rock shore of Vermelha Beach indicating the top (dashed line) and base (solid line). (b) Mature oocyte of *P. magna* (arrow) with a large nucleolated (nu) nucleus (n). (c) After successive cleavages the embryo became a stomoblastula (arrow), close to a choanocyte chamber. (d) After pass through an inversion, the embryo becomes an amphiblastula larva (a). (ca – canal, cc – choanocyte chamber).

at 2.5  $\mu\text{M}$  in samples fixed with 2% paraformaldehyde, according to Gasol & del Giorgio (2000) and Andrade *et al.* (2003). To each 0.5 mL triplicate sample, 25  $\mu\text{L}$  of a working bead solution was added (sampled after intense sonication), and the content was homogenized *via* vortexing for 15 s. Immediately thereafter, inside a fume hood under dim light, 20  $\mu\text{L}$  of a SYTO 13 working solution was added to each tube (final concentration, 2.5  $\mu\text{M}$ ), followed by mixing in a vortex apparatus for 15 s. The tubes were kept in the dark until cytometer readings were taken (between 30 and 90 min) using a FACSCalibur flow cytometer (BectonDikson, CA), equipped with a 20 mW blue laser with a standard optical filter configuration. Autotrophic bacteria were also analysed *via* flow cytometry, based on their autofluorescence properties (Gasol & del Giorgio 2000); however, their abundance and biomass were one- to twofold lower compared with the heterotrophic bacteria and were therefore not considered here.

#### Statistical analysis

All of the data sets were tested for normality and homoscedasticity prior to analysis. To evaluate whether the dynamics of the reproductive elements of *Paraleucilla magna* presented seasonality, a permutational multivariate analysis of variance (PERMANOVA, Anderson 2001) was applied to a Euclidean distance matrix for the data on all *P. magna* reproductive elements to evaluate whether they presented seasonal differences. PERMANOVA was performed using the 'vegan' package (Oksanen *et al.* 2013) in the R environment (R Development Core Team 2014).

Owing to the complex relationships among the explanatory environmental variables and their role in the reproductive response, we constructed regression trees providing intuitive insights regarding high-order interactions among variables (De'ath & Fabricius 2000). The procedure is based on repeated binary splitting of each response variable (Crawley 2007) expressed graphically in hierarchical trees of the main environmental drivers, with the root node on the top representing non-split data and the branches and leaves representing the groups partitioned by the explanatory variables. Each split is based on finding the one predictor variable (and a given threshold of that variable) that results in the greatest change in the explained deviance (for Gaussian error, this is equivalent to maximizing the between-group sum of squares). Tree functions carry out this process through an exhaustive search of all possible threshold values for each predictor. Once a split is made, the routine is repeated for each group separately until all deviance (or some low threshold) is explained, or there are too few samples in the remaining subset (Fridley 2010). In this analysis, we had

to exclude data for 3 months (September and December 2006, and June 2007) because of missing data for at least one of the three variables (temperature, phytoplankton and bacterioplankton). All statistical analyses were carried out in R (R Core Team 2012), with the regression tree analyses performed using the 'tree' package (Ripley 2014).

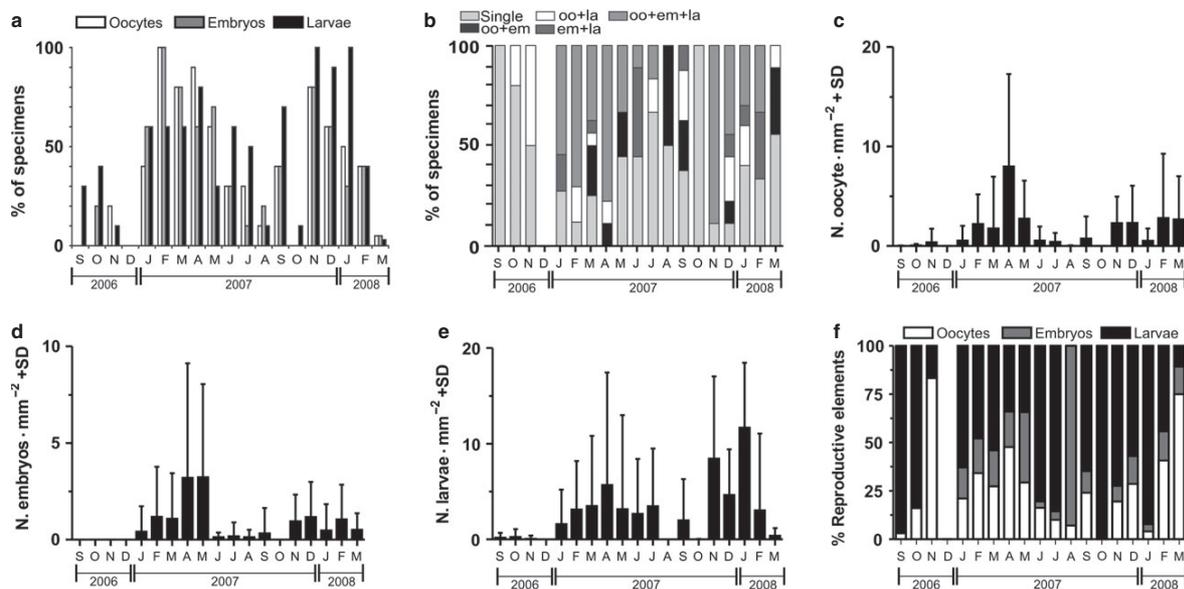
## Results

### Reproduction pattern

As previously described (Lanna & Klautau 2010), *Paraleucilla magna* is a simultaneous hermaphrodite and is viviparous, incubating embryos and spawning swimming larvae. Of the 225 specimens of this species analysed during this study, 140 (62%) showed at least one of the three female reproductive elements; 99 (44%) presented oocytes; 86 (38%) presented embryos; and 105 (47%) presented larvae (Fig. 3a). The three types of reproductive elements could either be present at the same time or not in the choanosome of the studied sponges. They were observed as one, two or three simultaneous elements in approximately the same proportions (33.3, 29.1, and 37.6%, respectively) during the studied period (Fig. 3b).

### Dynamics of the reproductive elements

With the exception of December 2006, the investigated population of *Paraleucilla magna* was always reproductively active (*i.e.* at least one of the reproductive elements was present in one of the sampled specimens, Fig. 3a). However, there was strong inter-individual and inter-annual variation in the densities of the reproductive elements. Mature oocytes (Fig. 2b) were found in all months, except for September and December 2006 and October 2007 (Fig. 3c). Higher densities of this reproductive element were first observed in the summer–fall period of 2007 and then from spring 2007–summer 2008 (Fig. 3c), demonstrating a lack of seasonality in gamete production. Embryos (Fig. 2c) were recorded only after December 2006, and their densities increased until reaching a peak in April and May 2007. The density values subsequently dropped during winter, then increased again just before summer 2008, but showed lower values than in the previous year (Fig. 3d). Amphiblastula larvae (Fig. 2d) were found year round, except for in December 2006 and August 2007, and usually displayed an average density higher than 3 larvae  $\cdot \text{mm}^{-2}$ . Higher densities occurred in April and November 2007 ( $5.7 \pm 11.7$  larvae  $\cdot \text{mm}^{-2}$  and  $8.4 \pm 8.5$  larvae  $\cdot \text{mm}^{-2}$ , respectively) and January 2008 ( $11.6 \pm 6.7$  larvae  $\cdot \text{mm}^{-2}$ ) (Fig. 3e), indicating that fertilization and embryonic development occurred without clear seasonality. Larvae were the most



**Fig. 3.** Dynamics of the reproductive elements of *P. magna*. (a) Monthly percentage of specimens bearing the reproductive elements (oocytes, embryos and larvae). (b) Percentage of specimens bearing simultaneously one, two or three of the reproductive elements. (c–e) Dynamics of the mean density of the reproductive elements (with standard deviations - SD) per mm<sup>2</sup> along the studied period: (c) Oocytes; (d) Embryos; (e) Larvae. (f) Proportional density of the reproductive elements (oocytes, embryos, and larvae) per month.

common reproductive element in the choanosome of *P. magna* throughout the studied period (Fig. 3f). Although seasonality was unclear for the reproductive elements (Fig. 3c–e), there was great variation in oocytes, embryos and larvae among the studied months, as demonstrated *via* PERMANOVA [Pseudo-F (17,197) = 5.9743;  $P < 0.001$ ].

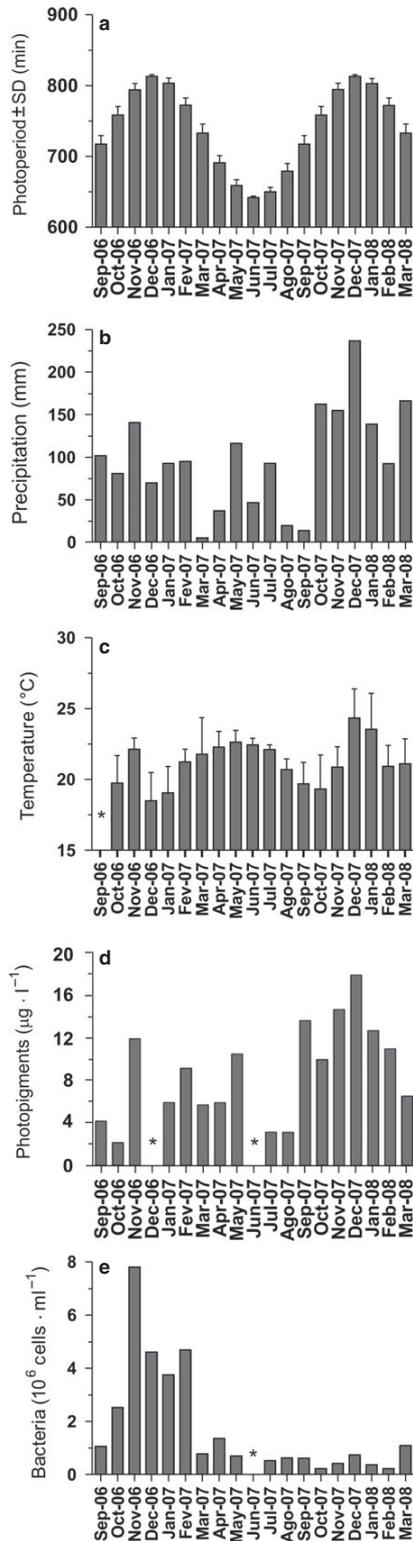
#### Dynamics of environmental parameters

The photoperiod in Rio de Janeiro varied from 641 min in June to 814 min in December and, as expected, presented a clear seasonal pattern, showing higher values in summer and lower photoperiods during winter (Fig. 4a). Lower precipitation was observed during fall and winter [although the lowest value was found in March (summer) 2007: 5.2 mm], and higher values were recorded in spring and summer (highest value found in December 2007: 237 mm) (Fig. 4b). Close to Vermelha Beach, the temperature varied between 16 and 28 °C. The warmest monthly average ( $24.3 \pm 2.0$  °C) occurred in December 2007, while the coldest waters were observed during December 2006 ( $18.5 \pm 2.0$  °C); these low temperatures observed during the summer season are due to the influence of upwelling waters on coastal regions around Rio de Janeiro, whose intensity can vary depending on the year (Yoneshigue-Valentin & Valentin 1992; Fernandes

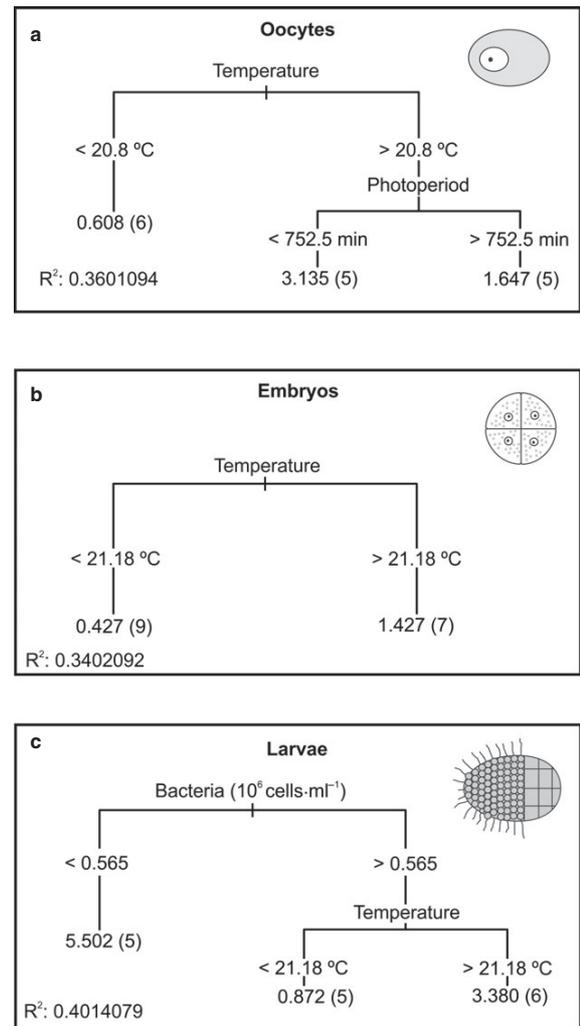
*et al.* 2012; Fig. 4c). The phytoplankton abundance also presented large variation during the studied period, showing an average value of  $8.7 \mu\text{g} \cdot \text{l}^{-1}$ . The highest phytoplankton abundance was observed during summer in December 2007 ( $17.9 \mu\text{g} \cdot \text{l}^{-1}$ ), while lower concentrations were observed during spring and winter ( $2.14 \mu\text{g} \cdot \text{l}^{-1}$  in October 2006 and  $3.12 \mu\text{g} \cdot \text{l}^{-1}$  in July 2007) (Fig. 4d). The abundance of bacterioplankton was higher during the first studied months (September 2006 to February 2007), after which it fell to values fluctuating around  $1.0 \times 10^6 \text{ cells} \cdot \text{ml}^{-1}$ . The highest abundance of bacteria was observed in November 2006 ( $7.8 \times 10^6 \text{ cells} \cdot \text{ml}^{-1}$ ; Fig. 4e). This environmental parameter did not present a seasonal pattern. In fact, with the exception of the photoperiod, all of the investigated environmental parameters lacked seasonal or monthly patterns.

#### Environmental drivers of reproductive dynamics

The regression tree analysis indicated (with moderate to high support, according to Cohen 1988) that among all of the possible explanatory variables, the main environmental parameters related to the reproduction of *Paraleucilla magna* were temperature, photoperiod and bacterioplankton (Fig. 5). Temperature was the main driver related to the density of oocytes and embryos, while bacterioplankton was the main driver of larval densities



**Fig. 4.** Dynamics of the environmental parameters investigated at Vermelha Beach from September 2006 to March 2008. (a) Average ( $\pm$ SD) values of photoperiod (min). (b) Precipitation accumulated during the whole month (mm). (c) Average ( $\pm$ SD) values of superficial seawater temperature in a location 2 km apart from Vermelha Beach. (d) Variation of phytoplankton abundance evidenced by the concentration of chlorophyll a and pheophytin (a degenerate stage of chlorophyll) (photopigments  $\mu\text{g}\cdot\text{L}^{-1}$ ) of the water mass; (e) Abundance of bacterioplankton ( $10^6 \text{ cells}\cdot\text{mL}^{-1}$ ) in the water mass of Vermelha Beach. (\* missing data).



**Fig. 5.** Regression tree for predicting the density of the reproductive elements of *P. magna* [(a) Oocytes, (b) Embryos, and (c) Larvae] from the investigated environmental parameters in Rio de Janeiro. Values present in the leaves are the mean values of the density (number·mm<sup>-2</sup>) of the reproductive elements and the number of months classified in each leaf (parentheses).

(Fig. 5). Oocytes were more abundant when the temperature was higher than 20.8 °C, mainly when the photoperiod was less than 752.5 min. These splits resulted in the highest fit with oocyte density ( $R^2 = 0.360$ ) among all of the trees produced (Fig. 5a). The only environmental parameter that was able to explain the dynamics of embryo density was temperature. When the temperature was higher than 21.18 °C, a higher density of embryos was observed. This model produced the best fit with embryo density ( $R^2 = 0.340$ ) among all of the generated trees (Fig. 5b). The density of larvae was higher when the concentration of bacteria was lower than  $0.565 \times 10^6$  cells · ml<sup>-1</sup>. When bacterioplankton was present above this concentration (which was more usual), a higher larval density was reached depending on higher temperatures (Fig. 5c). This was the model that best explained larval variation ( $R^2 = 0.401$ ) among all of the produced trees.

## Discussion

The Vermelha Beach population of *Paraleucilla magna* reproduced continuously throughout the studied period (September 2006 to March 2008) but presented strong inter-individual and inter-annual variation in its densities of reproductive elements (Fig. 3). Most calcareous sponges exhibit only one breeding season per year (see Lanna *et al.* 2007), although some are able to reproduce twice a year (Orton 1914, 1920). However, some species, including *P. magna* in the Mediterranean Sea, are capable of reproducing throughout the year (Sarà & Orsi 1975; Franzen 1988; Longo *et al.* 2012). In Rio de Janeiro, *P. magna* was reported to reproduce once a year from 2004 to early 2006 (Lanna *et al.* 2007). By contrast, in the present study, the examined population at the same beach (Vermelha Beach) reproduced throughout the year. Nevertheless, we cannot state that each individual was able to reproduce more than once a year, as we collected individuals randomly to study the population. However, Lanna *et al.* (2007) used the same criteria and found seasonality at the time of their study. These contrasting reproductive periodicities could be related to natural fluctuations in different environmental parameters.

During this study, *P. magna* presented one of the highest reproductive efforts documented to date in the phylum Porifera, especially if only viviparous species are considered (an exception is the viviparous species *Thoosa mismalolli*; Table 1). Larvae were the most abundant element throughout the studied period (see Fig. 3f), which may indicate that oogenesis and embryogenesis take place quickly, while larval ripening appears to require a longer period to be completed (*i.e.* larvae are incubated for a long time inside the epilarval trophocyte epithelium

before hatching; Lanna & Klautau 2012). In addition, the constant presence of a large number of larvae in *P. magna* tissue is indicative of continuous larval release at the population level, which is corroborated by the continuous recruitment of the species observed in the same area (Padua *et al.* 2013a). Constant larval release can be considered a reproductive strategy, as it reduces the risk of all larvae being lost due to unfavorable conditions (Strathmann 1985; Ettinger-Epstein *et al.* 2007) and may be one of the keys to the success of *P. magna* in colonizing new areas, as observed in other marine invertebrate invaders (Johnston *et al.* 2009).

In the Mediterranean population of *P. magna*, reproduction is also continuous, and fecundity can reach levels even higher than those observed in Brazil (Table 1; Longo *et al.* 2012). However, the Mediterranean population of this sponge presents a different reproductive strategy: although oocytes and embryos were observed continuously throughout the year, larvae occurred only from September to December in 2006 (Longo *et al.* 2012). If we consider only larvae as a proxy for fecundity, a contrasting scenario is observed between these two populations of *P. magna*. In the Brazilian population, larvae are continuously produced throughout the year, while in the Mediterranean, there is only a short period of larval production, although with a peak production that is higher than any density observed in Brazil. This may indicate that at higher latitudes, the reproductive effort must be concentrated and stronger to guarantee population success than in lower latitudes. This characteristic is most likely related to the different fluctuations of environmental parameters in the two regions (*i.e.* the densities of larvae may depend on environmental conditions; see below). In any case, the reproductive effort presented by this species is so high in both Rio de Janeiro and the Mediterranean Sea that it might promote the death of the parental organisms after larval release, indicating that individuals of *P. magna* are semelparous.

Many studies have indicated a strong influence of temperature on sponge reproduction, especially in temperate regions (*e.g.* Riesgo *et al.* 2007; Riesgo & Maldonado 2008; Maldonado & Riesgo 2009; Chung *et al.* 2010; Ereskovsky *et al.* 2013) and even in tropical areas (Fromont 1994a,b; Ettinger-Epstein *et al.* 2007). Temperature is usually implicated in either controlling gamete production or larval release (Fell 1974, 1993; Maldonado & Riesgo 2008). In several species of Demospongiae, for example, a seasonal increase in seawater temperature triggers the production of oocytes and spermatozoa (*e.g.* Fromont 1994a,b; Fromont & Bergquist 1994; Ettinger-Epstein *et al.* 2007; Mercurio *et al.* 2007; Whalan *et al.* 2007; Abdo *et al.* 2008; Riesgo & Maldonado 2008; Chung *et al.* 2010), although a decrease in water temperature has also been

**Table 1.** Comparison of the largest values of the densities of the reproductive elements of *Paraleucilla magna* and different species of Porifera. The densities of *Paraleucilla magna*'s elements are presented both in  $\text{mm}^{-2}$  and in  $\text{mm}^{-3}$  (calculated by the Abercrombie equation present in Elvin 1976) in order to facilitate the comparison. Studies in which the reproductive effort of the sponge is presented only as the reproductive output index (percentage of tissue occupied by reproductive elements) were excluded from this list (modified from Whalan *et al.* 2007).

species	reproductive elements			mode of development	reference
	oocytes	embryo	larvae		
<i>Paraleucilla magna</i> Brazil	8.0 $\text{mm}^{-2}$	3.4 $\text{mm}^{-2}$	11.7 $\text{mm}^{-2}$	V	
– Southwest Atlantic	(299 $\text{mm}^{-3}$ )	(89 $\text{mm}^{-3}$ )	(319 $\text{mm}^{-3}$ )		
Italy – Mediterranean	553.6 $\text{mm}^{-3}$	~300.0 $\text{mm}^{-3}$	121.5 $\text{mm}^{-3}$	V	20
<i>Haliclona loosanoffi</i>	–	–	–	V	1
<i>Stylopus</i> sp.	–	0.2 $\text{mm}^{-3}$	–	V	2
<i>Anchinoe</i> sp.	–	2.0 $\text{mm}^{-3}$	–	V	2
<i>Chondropsis</i> sp.	–	0.1 $\text{mm}^{-3}$	–	V	2
<i>Ancorina alata</i>	50.0 $\text{mm}^{-3}$	–	–	O	2
<i>Polymastia</i> sp.	130.0 $\text{mm}^{-3}$	–	–	O	2
<i>Polymastia hirsuta</i>	30.0 $\text{mm}^{-3}$	–	–	O	2
<i>Aaptos aaptos</i>	150.0 $\text{mm}^{-3}$	–	–	?	2
<i>Polymastia</i> sp.	220.0 $\text{mm}^{-3}$	–	–	?	2
<i>Raspaillia topsenti</i>	320.0 $\text{mm}^{-3}$	–	–	?	2
<i>Haliclona amboinensis</i> <sup>1</sup>	2.4 $\text{mm}^{-2}$	–	–	V	3
<i>Niphates nitida</i> <sup>1</sup>	2.9 $\text{mm}^{-2}$	–	–	V	3
<i>Haliclona cymiformis</i> <sup>1</sup>	0.8 $\text{mm}^{-2}$	–	–	V	3
<i>Xestospongia bergquistia</i> <sup>1</sup>	3.1 $\text{mm}^{-2}$	–	–	O	3
<i>Xestospongia testudinaria</i> <sup>1</sup>	1.8 $\text{mm}^{-2}$	–	–	O	3
<i>Xestospongia exigua</i> <sup>1</sup>	10.2 $\text{mm}^{-2}$	–	–	O	3
<i>Tethya aurantium</i>	507.3 $\text{mm}^{-3}$	–	–	O	4
<i>Tethya citrina</i>	689.7 $\text{mm}^{-3}$	–	–		4
<i>Mycale contarenii</i>	–	–	–	V	5
<i>Crambe crambe</i>	–	–	–	V	6
<i>Scopalina lophyropoda</i>	–	–	–	V	6
<i>Petrosia</i> sp.	~200.0 $\text{cm}^{-2}$	~60.0 $\text{cm}^{-2}$	~30.0 $\text{cm}^{-2}$	V	7
<i>Tetilla</i> sp. <sup>2</sup> shallow	–	–	–	O	8
deep	–	–	–		
<i>Spongia officinalis</i>	6.0 $\text{mm}^{-3}$	1.3 $\text{mm}^{-3}$	1.0 $\text{mm}^{-3}$	V	9
<i>Luffariela variabilis</i>	1.5 $\text{mm}^{-2}$	1.3 $\text{mm}^{-2}$	1.2 $\text{mm}^{-2}$	V	10
<i>Corticium candelabrum</i>	7.0 $\text{mm}^{-2}$	4.0 $\text{mm}^{-2}$ (21.3 $\text{mm}^{-3}$ )	4.2 $\text{mm}^{-2}$	V	11
<i>Rhopaloeides odorabile</i>	–	–	–	V	12
<i>Axinella damicornis</i>	<2.0 $\text{mm}^{-2}$	–	–	O	13
<i>Raspaciona aculeata</i>	~4.0 $\text{mm}^{-2}$	–	–	O	13
<i>Chondrosia reniformis</i>	~7.2 $\text{mm}^{-2}$	–	–	O	13
Spain – Mediterranean					
Italy – Adriatic Sea	44.6 $\text{mm}^{-3}$	–	–	O	19
<i>Haliclona</i> sp. 1 (green)	<0.1 $\text{mm}^{-2}$	0.2 $\text{mm}^{-2}$	<0.1 $\text{mm}^{-2}$	V	14
<i>Haliclona</i> sp. 2 (brown)	<0.1 $\text{mm}^{-2}$	0.1 $\text{mm}^{-2}$	<0.1 $\text{mm}^{-2}$	V	14
<i>Petrosia ficiformis</i>	~11.5 $\text{cm}^{-3}$	–	–	O	15
<i>Spongia ceylonensis</i>	–	~10 $\text{mm}^{-3}$	–	V	16
<i>Thoosa mismalolli</i> <sup>1</sup>	18.0 $\text{mm}^{-2}$	54.0 $\text{mm}^{-2}$	–	V	17
<i>Cliona viridis</i>	~12.0 $\text{mm}^{-2}$	–	–	O	18
<i>Cliona celata</i>	~12.0 $\text{mm}^{-2}$	–	–	O	18
<i>Tedania anhelans</i>	8.5 $\text{mm}^{-3}$	18.5 $\text{mm}^{-3}$	1.2 $\text{mm}^{-3}$	V	19
<i>Hemimycale columella</i>	4.0 $\text{mm}^{-2}$	0.43 $\text{mm}^{-2}$	0.07 $\text{mm}^{-2}$	V	20
<i>Crella elegans</i>	0.17 $\text{mm}^{-2}$	0.33 $\text{mm}^{-2}$	1.6 $\text{mm}^{-2}$	V	20
<i>Sarcotragus spinosulus</i> <sup>2</sup>	8.1 $\text{mm}^{-3}$	1.5 $\text{mm}^{-3}$	0.8 $\text{mm}^{-3}$	V	21

V, viviparous; O, oviparous.

References: 1, Fell (1976); 2, Ayling (1980); 3, Fromont (1994b); 4, Corriero *et al.* (1996); 5, Corriero *et al.* (1998); 6, Uriz *et al.* (1998); 7, Asa *et al.* (2000); 8, Meroz-Fine *et al.* (2005); 9, Baldacconi *et al.* (2007); 10, Ettinger-Epstein *et al.* (2007); 11, Riesgo *et al.* (2007); 12, Whalan *et al.* (2007); 13, Riesgo & Maldonado (2008); 14, Abdo *et al.* (2008); 15, Maldonado & Riesgo (2009); 16, Chung *et al.* (2010); 17, Bautista-Guerrero *et al.* (2010); 18, Piscitelli *et al.* (2011); 19, Longo *et al.* (2012); 20, Perez-Porro *et al.* (2012); 21, Mercurio *et al.* (2013).

<sup>1</sup>There is no differentiation between embryos and larvae in the quantification.

<sup>2</sup>Data provided for different locations.

? means unknown.

related to the onset of reproduction in some species (Fell 1993; Riesgo *et al.* 2007).

Few works have investigated the possible role of temperature in the reproduction of calcareous sponges. The Brazilian population of *P. magna* showed higher production of oocytes and embryos when the temperature was elevated (Fig. 5), which is also observed in the Mediterranean population (Longo *et al.* 2012). However, while a higher investment in oocytes and embryos was observed in Brazil when the temperature was above 20.8 and 21.18 °C, respectively, in the Mediterranean this increment was observed only at temperatures over 26–28 °C (Longo *et al.* 2012). The calcareous sponge *Sycon scaldense* was found to reproduce in the Netherlands during two periods of the year: August (when the temperature was 18 °C) and November–December (when the temperature varied between 4 and 9 °C), indicating that that this species can maintain its reproductive activity at very disparate temperatures (van Koolwijk 1982; Fell 1993). By contrast, reproduction of *Petrobiona massiliana* and *Clathrina blanca* is most likely triggered by an increase in water temperature in the Mediterranean and along the Pacific coast of the USA, respectively (Vacelet 1964; Johnson 1978). In both studies, in years when high temperatures were maintained for a longer period, the reproductive period of these species was also longer (Vacelet 1964; Johnson 1978).

It is has been demonstrated that the thermal regime in a given region can trigger the reproduction of different species in different periods (Riesgo & Maldonado 2008; Mercurio *et al.* 2013) and that the same species will reproduce in different periods depending on the latitude where it is found (Fell 1993; Witte & Barthel 1994), indicating that the temperature effect on sponge reproduction is species specific. The Mediterranean and Brazilian populations of *P. magna* both reproduce year round (Longo *et al.* 2012), independent of the thermal regimes experienced in these different latitudes. Moreover, at both locations, a rise in temperature apparently increases the fecundity of individuals. This finding indicates that rather than triggering the initiation of reproduction, temperature may influence the rate and quantity of reproductive elements being produced.

In addition to temperature, photoperiod and bacterioplankton were also implicated as drivers of the reproductive dynamics of the Brazilian population of *P. magna* (Fig. 5). Marine invertebrate reproduction is strongly associated with photoperiod (Olive 1995). This environmental parameter is generally connected to temperature, but while temperature is related to the rate of reproduction in marine invertebrates, photoperiod is implicated as a timing mechanism (Olive 1995). Although there is little information on the influence of this parameter in sponge repro-

duction, photoperiod has been implicated in the onset of gametogenesis in *Haliclona permolis* (Elvin 1976). Maldonado & Riesgo (2008) suggested that in areas showing low temperature variation throughout the year (which is the case for Vermelha Beach in Rio de Janeiro), gametogenesis should rely on stimuli undergoing more intense changes, such as photoperiod. This appears to be the case for *P. magna* oogenesis in Rio de Janeiro, as this species generally presented an increase in density associated with an increase in day length (see Figs 3 and 4).

Bacterioplankton was implicated as the main driver of larval production. We have previously observed that the amphiblastula of *P. magna* appears to feed on bacteria, which are transferred to the larval cavity, while the larvae are brooded inside the parental sponge (Lanna & Klautau 2012). Bacteria have been considered one of the main sources of energy for sponges (Reiswig 1974; Ribes *et al.* 1999; Stabili *et al.* 2006). Ribes *et al.* (1999) observed that *Dysidea avara* could feed on a broad size spectrum of food sources, allowing the species to maintain a constant level of food uptake throughout the year. This species releases larvae during summer (Mariani *et al.* 2005), when the sponge diet is heavily dependent on bacteria (Ribes *et al.* 1999).

A large difference in the period of the major reproductive output of the *P. magna* population was observed between the years 2007 and 2008 (Fig. 3). Additionally, the reproductive activity of this population was continuous during the studied period, constituting a completely different situation from a previous report (Lanna *et al.* 2007), when in the same locality, this species was found to be reproductively active only during summer. The fluctuation in the length of the reproductive period from year to year observed in *P. magna* is most likely related to different thermal regimes in Vermelha Beach. Albuquerque & Genofre (1999) showed that the temperature at Vermelha Beach was higher during the summer period and decreased during the fall and winter months. This pattern was not recovered during the present study: the temperature was constantly high from January through November 2007 (Fig. 4c). Therefore, the high temperatures found during this study may explain the different reproduction patterns found for the same population of *P. magna* here and in a previous study, when this species reproduced only during summer (Lanna *et al.* 2007). The same explanation may be given for the different timing of the strongest reproductive output observed between 2007 and 2008 (Fig. 3). Changes in the reproductive period of marine species related to modifications of thermal regimes have been observed in various groups of marine invertebrates in different regions of the globe (Olive 1995; Lawrence & Soame 2004 and references therein).

## Conclusions

In contrast to our hypothesis, our results showed that the Vermelha Beach population of *Paraleucilla magna* was constantly reproducing. However, we confirmed that some of the environmental parameters investigated here had effects on the reproductive effort of this species. *Paraleucilla magna* is a species with a strong invasive potential, considering its reproductive effort and continuous reproduction, and apparently requires a much higher reproductive effort to colonize higher than lower latitudes (Longo *et al.* 2012). Fast growth (Cavalcanti *et al.* 2013), a young reproductive age (Lanna *et al.* 2007), high recruitment rates (Padua *et al.* 2013b) and high fecundity (Longo *et al.* 2012; present study), stimulated by higher temperatures, may be key factors contributing to the growth of *P. magna* populations and its invasion of new areas. Our findings highlight the complex interactions between the environment and this sponge that must be taken into account to understand and monitor the spreading of this species in new areas.

## Acknowledgements

We would like to thank the staff of the Laboratório de Biologia de Porifera (LaBiPor) for helping in the collection of specimens, and the staff of the Laboratório de Hidrobiologia for the analyses of different environmental parameters. We thank PPGDA (Programa de Pós-Graduação em Diversidade Animal and PROPCI/UFBA (Pró-Reitoria de Pesquisa, Criação e Inovação da Universidade Federal da Bahia) for funding the revision of the English language. Sponge specimens were sampled with permission of the Brazilian authorities (Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis – IBAMA and Instituto Chico Mendes de Conservação da Biodiversidade – ICMBIO). This study was funded by the Rio de Janeiro State Research Foundation (Fundação Carlos Chagas Filho de Amparo à Pesquisa do Estado do Rio de Janeiro – FAPERJ) and the Brazilian National Council of Technological and Scientific Development (CNPq). M.K., P.C.P. and R.P. received research fellowships from CNPq. E.L. received a scholarship from the Coordination of Superior Level Staff Improvement (CAPES). This work was part of the M.Sc. thesis of E.L. presented to the Graduation Program in Zoology of the Museu Nacional do Rio de Janeiro/UFRJ.

## References

- Abdo D.A., Fromont J., McDonald J.I. (2008) Strategies, patterns and environmental cues for reproduction in two temperate haliclونid sponges. *Aquatic Biology*, **1**, 291–302.
- Agell G., Frotscher J., Guardiola M., Pascual M., Uriz M.J. (2012) Characterization of nine polymorphic microsatellite loci for the calcareous sponge *Paraleucilla magna* Klautau *et al.* 2004 introduced to the Mediterranean Sea. *Conservation Genetic Resources*, **4**, 403–405.
- Albuquerque E.F., Genofre G.C. (1999) Flutuação da população de *Microcerberus ramosae* (Crustacea: Isopoda) da fauna intersticial da Praia Vermelha, Rio de Janeiro, Brasil. In: Silva S.H.G., Lavrado H.P. (Eds), *Ecologia dos Ambientes Costeiros do Estado do Rio de Janeiro. Série Oecologia Brasiliensis*. PPGE-UFRJ, Rio de Janeiro, Brasil: 229–243.
- AlertaRio (2008) Rio de Janeiro City Government. <http://www2.rio.rj.gov.br/georio/site/alerta/alerta.htm>. Accessed from September 2006 to March 2008.
- Anderson M.J. (2001) A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, **26**, 32–46.
- Andrade L., Gonzales A.M., Araujo F.V., Paranhos R. (2003) Flow cytometry assessment of bacterioplankton in tropical marine environments. *Journal of Microbiological Methods*, **55**, 841–850.
- Asa S., Yeemin T., Chaitanawisuti N., Kritsanapuntu A. (2000) Sexual reproduction of a marine sponge, *Petrosia* sp. from coral communities in the Gulf of Thailand. *Proceedings 9th International Coral Reef Symposium*, **1**, 421–424.
- Ayling A.L. (1980) Patterns of sexuality, asexual reproduction and recruitment in some subtidal marine demospongiae. *Biological Bulletin*, **158**, 271–282.
- Baldacconi R., Nonnis-Marzano C., Gaino E., Corriero G. (2007) Sexual reproduction, larval development and release in *Spongia officinalis* L. (Porifera, Demospongiae) from the Apulian coast. *Marine Biology*, **152**, 969–979.
- Batista D., Muricy G., Rocha R., Miekeley N. (2014) Marine sponges with contrasting life histories can be complementary biomonitors of heavy metal pollution in coastal ecosystems. *Environmental Science and Pollution Research*, **21**, 5785–5794.
- Bautista-Guerrero E., Carballo J.L., Maldonado M. (2010) Reproductive cycle of the coral-excavating sponge *Thoosa mismalolli* (Clionaidae) from Mexican Pacific coral reefs. *Invertebrate Biology*, **129**, 285–296.
- Cavalcanti F.F., Skinner L.F., Klautau M. (2013) Population dynamics of cryptogenic calcarean sponges (Porifera, Calcarea) in Southeastern Brazil. *Marine Ecology*, **34**, 280–288.
- Chung I.F., Huang Y.M., Lee T.H., Liu L.L. (2010) Reproduction of the bath sponge *Spongia ceylonensis* (Dictyoceratida: Spongiidae) from Penghu, Taiwan. *Zoological Studies*, **49**, 601–607.
- Cohen J. (1988) *Statistical Power Analysis for the Behavioral Sciences* (2nd edn). Lawrence Erlbaum Associates, New Jersey: 567.
- Corriero G., Sarà M., Vaccaro P. (1996) Sexual and asexual reproduction in two species of *Tethya* (Porifera: Demospongiae) from a Mediterranean coastal lagoon. *Marine Biology*, **126**, 175–181.

- Corriero G., Liaci-Scalera L., Marzano C.N., Gaino E. (1998) Reproductive strategies of *Mycale contarenii* (Porifera: Demospongiae). *Marine Biology*, **131**, 319–327.
- CPTEC-INPE (2008) *Brazilian National Institute for Space Research*. <http://tempo1.cptec.inpe.br/>. Accessed from September 2006 to March 2008.
- Crawley M. (2007) *The R Book*. John Wiley & Sons, Chichester.
- Cvitković I., Despalatović M., Grubelić I., Nikolić V., Pleše B., Žuljević A. (2013) Occurrence of *Paraleucilla magna* (Porifera: Calcarea) in the eastern Adriatic Sea. *Acta Adriatica*, **54**, 93–99.
- De'ath G., Fabricius K.E. (2000) Classification and regression trees: a powerful yet simple technique for ecological data analysis. *Ecology*, **81**, 3178–3192.
- Elvin D.W. (1976) Seasonal growth and reproduction of an intertidal sponge, *Haliclona permollis* (Bowerbank). *Biological Bulletin*, **151**, 108–125.
- Ereskovsky A.V. (2000) Reproduction cycles and strategies of the cold-water sponges *Halisarca dujardini* (Demospongiae, Halisarcida), *Myxilla incrustans* and *Iophon piceus* (Demospongiae, Poecilosclerida) from the White Sea. *Biological Bulletin*, **198**, 77–87.
- Ereskovsky A., Dubois M., Ivanišević J., Gazave E., Lapebie P., Tokina D., Pérez T. (2013) Pluri-annual study of the reproduction of two Mediterranean *Oscarella* species (Porifera, Homoscleromorpha): cycle, sex-ratio, reproductive effort and phenology. *Marine Biology*, **160**, 423–438.
- Ettinger-Epstein P., Whalan S.W., Battershill C.N., de Nys R. (2007) Temperature cues gametogenesis and larval release in a tropical sponge. *Marine Biology*, **153**, 171–178.
- Fell P.E. (1974) Porifera. In: Giese A.C., Pearse J.S. (Eds), *Reproduction of Marine Invertebrates. Volume I: Acoelomate and Pseudocoelomate Metazoans*. Academic Press, New York and London: 51–132.
- Fell P.E. (1976) The reproduction of *Haliclona loosanoffi* and its apparent relationship to water temperature. *Biological Bulletin*, **150**, 200–210.
- Fell P.E. (1993) Porifera. In: Adiyodi K.G., Adiyodi R.G. (Eds), *Reproductive Biology of Invertebrates. Volume VI, Part A: Asexual Propagation and Reproductive Strategies*. John Wiley & Sons, Chichester, UK: 1–44.
- Fernandes L.D.D.A., Quintanilha J., Monteiro-Ribas W., Gonzalez-Rodriguez E., Coutinho R. (2012) Seasonal and interannual coupling between sea surface temperature, phytoplankton and meroplankton in the subtropical south-western Atlantic Ocean. *Journal of Plankton Research*, **34**, 236–244.
- Franzen W. (1988) Oogenesis and larval development of *Scypha ciliata* (Porifera, Calcarea). *Zoomorphology*, **107**, 349–357.
- Fridley J. (2010) Lecture notes from Bio793: Plant Ecology/ Niche modeling/Modeling in R. <http://plantecology.syr.edu/fridley/bio793/cart.html>. Accessed 23 April 2014.
- Fromont J. (1994a) The reproductive biology of tropical species of Haplosclerida and Petrosida on the Great Barrier Reef. In: Van Soest R.W.M., van Kempen T.M.G., Braekman J.C. (Eds), *Sponges in Time and Space*. Balkema, Rotterdam: 307–311.
- Fromont J. (1994b) Reproductive development and timing of tropical sponges (Order Haplosclerida) from the Great-Barrier-Reef, Australia. *Coral Reefs*, **13**, 127–133.
- Fromont J., Bergquist P.R. (1994) Reproductive biology of three sponge species of the genus *Xestospongia* (Porifera: Demospongiae: Petrosida) from the Great Barrier Reef. *Coral Reefs*, **13**, 119–126.
- Gasol J.M., del Giorgio P.A. (2000) Using flow cytometry for counting natural planktonic bacteria and understanding the structure of planktonic bacterial communities. *Scientia Marina*, **64**, 197–224.
- Geller J.B., Darling J.A., Carlton J.T. (2010) Genetic perspectives on marine biological invasions. *Annual Review of Marine Science*, **2**, 367–393.
- Gravili C., Belmonte G., Cecere E., Denitto F., Giangrande A., Guidetti P., Longo C., Mastrototaro F., Moscatello S., Petrocelli A., Piraino S., Terlizzi A., Boero F. (2010) Nonindigenous species along the Apulian coast, Italy. *Chemistry and Ecology*, **26**(suppl 1), 121–142.
- Guardiola M., Frotscher J., Uriz M. (2012) Genetic structure and differentiation at a short-time scale of the introduced calcarean sponge *Paraleucilla magna* to the western Mediterranean. *Hydrobiologia*, **687**, 71–84.
- INMET (2008) *Instituto Nacional de Meteorologia*. <http://www.inmet.gov.br>. Accessed from September 2006 to March.
- Johnson M.F. (1978) Studies on the reproductive cycles of the calcareous sponges *Clathrina coriacea* and *C. blanca*. *Marine Biology*, **50**, 73–79.
- Johnston E.L., Piola R.F., Clark G.F. (2009) The role of propagule pressure in invasion Success. In: Rilov G., Crooks J.A. (Eds), *Biological Invasions in Marine Ecosystems*. Springer, Berlin: 133–151.
- Klautau M., Monteiro L., Borojevic R. (2004) First occurrence of the genus *Paraleucilla* (Calcarea, Porifera) in the Atlantic Ocean: *P. magna* sp. nov. *Zootaxa*, **710**, 1–8.
- van Koolwijk T. (1982) Calcareous sponges of the Netherlands (Porifera, Calcarea). *Bulletin Zoologisch Museum Universiteit van Amsterdam*, **8**, 89–98.
- Lanna E., Klautau M. (2010) Oogenesis and spermatogenesis in *Paraleucilla magna* (Porifera, Calcarea). *Zoomorphology*, **129**, 249–261.
- Lanna E., Klautau M. (2012) Embryogenesis and larval ultrastructure in *Paraleucilla magna* (Calcarea, Calcarea), with remarks on the epilarval trophocyte epithelium (“placental membrane”). *Zoomorphology*, **131**, 277–292.
- Lanna E., Monteiro L.C., Klautau M. (2007) Life cycle of *Paraleucilla magna* Klautau, Monteiro and Borojevic, 2004 (Porifera, Calcarea). In: Custódio M.R., Lôbo-Hajdu G., Hajdu E., Muricy G. (Eds), *Porifera Research - Biodiversity*,

- Innovation and Sustainability*. Museu Nacional - Série Livros, Rio de Janeiro: 413–418.
- Lawrence A.J., Soame J.M. (2004) The effects of climate change on the reproduction of coastal invertebrates. *IBIS*, **146**, 29–39.
- Longo C., Mastrototaro F., Corriero G. (2007) Occurrence of *Paraleucilla magna* (Porifera: Calcarea) in the Mediterranean sea. *Journal of the Marine Biological Association of the United Kingdom*, **87**, 1749–1755.
- Longo C., Pontassuglia C., Corriero G., Gaino E. (2012) Life-cycle traits of *Paraleucilla magna*, a calcareous sponge invasive in a coastal Mediterranean Basin. *PLoS ONE*, **7**, e42392.
- Maldonado M., Riesgo A. (2008) Reproduction in Porifera: a synoptic overview. *Treballs de la Societat Catalana de Biologia*, **59**, 29–49.
- Maldonado M., Riesgo A. (2009) Gametogenesis, embryogenesis, and larval features of the oviparous sponge *Petrosia ficiformis* (Haplosclerida, Demospongiae). *Marine Biology*, **156**, 2181–2197.
- Mariani S., Uriz M.J., Turon X. (2005) The dynamics of sponge larvae assemblages from northwestern Mediterranean nearshore bottoms. *Journal of Plankton Research*, **27**, 249–262.
- Mercier A., Hamel J.-F. (2009) Endogenous and exogenous control of gametogenesis and spawning in echinoderms. *Advances in Marine Biology*, **55**, 1–302.
- Mercurio M., Corriero G., Gaino E. (2007) A 3-year investigation of sexual reproduction in *Geodia cydonium* (Jameson 1811) (Porifera, Demospongiae) from a semi-enclosed Mediterranean bay. *Marine Biology*, **151**, 1491–1500.
- Mercurio M., Corriero G., Gherardi M., Baldacconi R., Gaino E. (2013) Sexual reproduction in *Sarcotragus spinosulus* from two different shallow environments. *Marine Ecology*, **34**, 394–408.
- Meroz-Fine E., Shefer S., Ilan M. (2005) Changes in morphology and physiology of an East Mediterranean sponge in different habitats. *Marine Biology*, **147**, 243–250.
- Monteiro L.C., Muricy G. (2004) Patterns of sponge distribution in Cagarras Archipelago, Rio de Janeiro, Brazil. *Journal of the Marine Biological Association of the United Kingdom*, **84**, 681–687.
- Occhipinti-Ambrogi A., Marchini A., Cantone G., Castelli A., Chimenz C., Cormaci M., Froggia C., Furnari G., Gambi M., Giaccone G., Giangrande A., Gravili C., Mastrototaro F., Mazziotti C., Orsi-Relini L., Piraino S. (2011) Alien species along the Italian coasts: an overview. *Biological Invasions*, **13**, 215–237.
- Oksanen J., Blanchet F.G., Kindt R., Legendre P., Minchin P.R., O'Hara R.B., Simpson G.L., Solymos P., Stevens M.H.H., Wagner H. (2013). *Vegan: Community Ecology Package*. R package version 2.0-10, <http://CRAN.R-project.org/package=vegan>
- Olive P.J.W. (1995) Annual breeding cycles in marine invertebrates and environmental-temperature - Probing the proximate and ultimate causes of reproductive synchrony. *Journal of Thermal Biology*, **20**, 79–90.
- Orton J.H. (1914) Preliminary account of a contribution to an evaluation of the Sea. *Journal of the Marine Biological Association of the United Kingdom*, **10**, 312–326.
- Orton J.H. (1920) Sea temperature, breeding and distribution in marine animals. *Journal of the Marine Biological Association of the United Kingdom*, **12**, 312–326.
- Padua A., Lanna E., Zilberberg C., Paiva P.C., Klautau M. (2013a) Recruitment, habitat selection and larval photoresponse of *Paraleucilla magna* (Porifera, Calcarea) in Rio de Janeiro, Brazil. *Marine Ecology*, **34**, 56–61.
- Padua A. Q., Lanna E., Klautau M. (2013b) Macrofauna inhabiting the sponge *Paraleucilla magna* (Porifera: Calcarea) in Rio de Janeiro, Brazil. *Journal of the Marine Biological Association of the United Kingdom*, **93**, 889–898.
- Parsons T.R., Maita Y., Lalli C.M. (1984) *A Manual of Chemical and Biological Methods for Seawater Analysis*. Pergamon Press, Oxford: 184.
- Perez-Porro A.R., Gonzalez J., Uriz M.J. (2012) Reproductive traits explain contrasting ecological features in sponges: the sympatric poecilosclerids *Hemimycale columella* and *Crella elegans* as examples. *Hydrobiologia*, **687**, 315–330.
- Pierrri C., Longo C., Giangrande A. (2010) Variability of fouling communities in the Mar Piccolo of Taranto (Northern Ionian Sea, Mediterranean Sea). *Journal of the Marine Biological Association of the United Kingdom*, **90**, 159–167.
- Piscitelli M., Corriero G., Gaino E., Uriz M.J. (2011) Reproductive cycles of the sympatric excavating sponges *Cliona celata* and *Cliona viridis* in the Mediterranean Sea. *Invertebrate Biology*, **130**, 1–10.
- Przeslawski R., Ahyong S., Byrne M., Wörheide G., Hutchings P.A.T. (2008) Beyond corals and fish: the effects of climate change on noncoral benthic invertebrates of tropical reefs. *Global Change Biology*, **14**, 2773–2795.
- R Core Team (2012) *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org>. ISBN 3-900051-07-0.
- R Development Core Team (2014) *A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org>
- Reiswig H.M. (1974) Water transport, respiration and energetics of three tropical marine sponges. *Journal of Experimental Marine Biology and Ecology*, **14**, 231–249.
- Ribes M., Coma R., Gili J.M. (1999) Natural diet and grazing rate of the temperate sponge *Dysidea avara* (Demospongiae, Dendroceratida) throughout an annual cycle. *Marine Ecology Progress Series*, **176**, 179–190.
- Riesgo A., Maldonado M. (2008) Differences in reproductive timing among sponges sharing habitat and thermal regime. *Invertebrate Biology*, **127**, 357–367.
- Riesgo A., Maldonado M., Durfort M. (2007) Dynamics of gametogenesis, embryogenesis, and larval release in a

- Mediterranean homosclerophorid demosponge. *Marine and Freshwater Research*, **58**, 398–417.
- Ripley B.. (2014) *Tree: Classification and regression trees*. R package- version 1.0-35. <http://CRAN.R-project.org/package=tree>
- Rossi A.L., Campos A.P.C., Barroso M.M.S., Klautau M., Archanjo B.S., Borojevic R., Farina M., Werckmann J. (2014) Long-range crystalline order in spicules from the calcareous sponge *Paraleucilla magna* (Porifera, Calcarea). *Acta Biomaterialia*, **1**, 529–547.
- Ruiz G.M., Hewitt C.L. (2002) Toward understanding patterns of coastal marine invasions: a prospectus. In: E. Leppäkoski, S. Gollasch, S. Olenin (Eds). *Invasive Aquatic Species of Europe. Distribution, Impacts and Management*. Springer, the Netherlands: 529–547.
- Sarà M., Orsi L.R. (1975) Sex differentiation in *Sycon* (Porifera Calcispongiae). *Pubblicazioni della Stazione Zoologica di Napoli*, **39**, 618–634.
- Stabili L., Licciano M., Giangrande A., Longo C., Mercurio M., Marzano C.N., Corriero G. (2006) Filtering activity of *Spongia officinalis* var. *adriatica* (Schmidt) (Porifera, Demospongiae) on bacterioplankton: implications for bioremediation of polluted seawater. *Water Research*, **40**, 3083–3090.
- Strathmann R.R. (1985) Feeding and nonfeeding larval development and life-history evolution in marine-invertebrates. *Annual Review of Ecology and Systematics*, **16**, 339–361.
- Turque A.S., Batista D., Silveira C.B., Cardoso A.M., Vieira R.P., Moraes F.C., Clementino M.M., Albano R.M., Paranhos R., Martins O.B., Muricy G. (2010) Environmental shaping of sponge associated archaeal communities. *PLoS ONE*, **5**, 10.
- Uriz M.J., Maldonado M., Turon X., Martí R. (1998) How do reproductive output, larval behaviour, and recruitment contribute to adult spatial patterns in Mediterranean encrusting sponges? *Marine Ecology Progress Series*, **167**, 137–148.
- Vacelet J. (1964) Etude monographique de l'éponge calcaire pharétronide de Méditerranée, *Petrobiona massiliana* Vacelet et Lévi. Les pharétronides actuelles et fossiles. *Recueil des Travaux de la Station Marine d'Endoume*, **34**, 1–125.
- Whalan S., Battershill C., de Nys R. (2007) Sexual reproduction of the brooding sponge *Rhopaloeides odorabile*. *Coral Reefs*, **26**, 655–663.
- Witte U., Barthel D. (1994) Reproductive cycle and oogenesis of *Halichondria panicea* (Pallas) in Kiel Bight. In: vanSoest R.W.M., Kempen V., Braekman J.C. (Eds). *Sponges in Time and Space*. Balkeman, Rotterdam: 297–305.
- Yoneshigue-Valentin Y., Valentin J.L. (1992) Macroalgae of the Cabo Frio upwelling region, Brazil: ordination of communities. In: Seeliger U. (Ed), *Coastal Plants of Latin America*. Academic Press, San Diego: 31–49.
- Zammit P.P., Longo C., Schembri P.J. (2009) Occurrence of *Paraleucilla magna* Klautau *et al.*, 2004 (Porifera: Calcarea) in Malta. *Mediterranean Marine Science*, **10**, 135–138.