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# *In situ* method for assessing the biometric data of *Pinna rudis* Linnaeus, 1758

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RESUMO

*Pinna rudis* Linnaeus, 1758, é uma espécie de bivalve pertencente à família Pinnidae com uma distribuição Atlântico-Mediterrânica. A degradação do habitat tem sido considerada a principal causa de declínio populacional no passado recente, elevando o estatuto de conservação à categoria de vulnerável em algumas paragens. Estudos sobre a dinâmica populacional serão necessários para o delineamento de estratégias para a conservação da espécie. Além disso, é necessário o desenvolvimento de novos métodos de colheita de dados individuais, uma vez que os indivíduos são altamente sensíveis à extracção e manipulação. No presente estudo é proposto um método não invasivo para recolha *in situ* de dados morfométricos de *P. rudis*. Para isso, foram amostrados 900 m$^2$ do fundo marinho da praia de Matiota, na ilha de São Vicente, arquipélago de Cabo Verde e colhidos 18 espécimes para calcular as relações entre comprimento e largura da concha. A equação de regressão entre os parâmetros permitiu estimar o tamanho total de 59 indivíduos de *P. rudis* encontrados nessa praia. O método não invasivo adoptado permitiu determinar o tamanho total dos indivíduos sem removê-los do substrato, o que poderá permitir fazer estudos comparativos da espécie em diferentes zonas.

**Palavras-chave:** bivalvia, Cabo Verde, morfometria, método não invasivo.

ABSTRACT

*Pinna rudis* Linnaeus, 1758 (Bivalvia: Pinnidae) has an Atlantic-Mediterranean distribution. Habitat degradation is considered the main cause of population declines in the recent past, raising the conservation status of the species to the category of vulnerable in some places. Population dynamics studies of *P. rudis* are still necessary to fully understand its conservation requirements. Moreover, new methods of individual data collection should be developed as individuals are highly sensitive to extraction and manipulation. In the present study, we propose a non-invasive method to collect *P. rudis* morphometric data *in situ*. For this, we sampled 900 $m^2$ of the sea bed at Matiota Beach on São Vicente Island, Cabo Verde Archipelago, and collected 18 individuals to compute the relationship between shell length and width. The regression equation between the parameters allowed us to estimate the total size of 59 *P. rudis* individuals obtained from the beach. The non-invasive method adopted allowed determination of the total size of the individuals without removing them from the substratum and, thereby, allowing the comparative study of the species in different zones.

**Keywords:** bivalvia, Cabo Verde, morphometry, non-invasive method

## INTRODUCTION

*Pinna rudis* Linnaeus, 1758 is distributed throughout the Mediterranean and the northeast Atlantic (Azores, Madeira, Selvagens, Canaries and Cabo Verde), extending as far south as Saint Helena and the Gulf of Guinea (Poppe & Goto 1993, Barea-Azcón *et al.* 2008). It occurs on sandy, rocky and muddy substrata, at depths that can exceed 40 metres (Sempere *et al.* 2006). Its life cycle is characterized by two phases, one planktonic and the other fixed to the substratum via byssal threads (Basso *et al.* 2015). On the seabed, *P. rudis* shells frequently provide a hard substratum for other species to settle upon, a phenomenon known as epibiosis (Gómez-Rodríguez & Pérez-Sánchez 1997). These epibiotic associations give their shells an irregular appearance often full of colour. The diversity and complexity of epibiotic organisms tend to increase with the age of *Pinna* specimens (Basso *et al.* 2015). As such, numerous biofouling species of algae, molluscs, bryozoans, polychaetes, ascidians, cnidarians and sponges occur on *P. rudis* shells (García-March 2005).

Due to habitat degradation, often caused by fishing activities, anchors and alterations to the rocky bottom of coastal areas, *P. rudis* populations are declining. This has led to their inclusion in Annex II of the Bern Convention as a ‘strictly protected’ species (in synonymy with also cited *P. pernula* Chemnitz, 1785), the Barcelona Convention as a ‘Threatened or Endangered’ marine species (Nebot-Colomer *et al.* 2016), and in the *Libro Rojo de los Invertebrados de Andalucía*, as ‘Vulnerable’ (Barea-Azcón *et al.* 2008). The ecological importance of *P. rudis* has been studied mainly in the Mediterranean Sea, specifically with regard to the interaction of shell epibionts (Richardson *et al.* 1997, Wirtz & D’Udekem-d’Acroz 2001, 2008, Cosentino & Giacobbe 2007, 2008, Barreiros *et al.* 2016), differences in shell ornamentation (Cosentino & Giacobbe 2006), the capacity for induced shell repair (Dietl & Alexander 2005) and population structure and growth (Nebot-Colomer *et al.* 2016).

Except for taxonomic/ biogeographically related studies (e.g., Ávila 2000, Segers *et al.* 2009, Gómez & Pérez 2011, Sanna *et al.* 2013), no ecological studies on *P. rudis* are known for the Macaronesian archipelagos.

Unfortunately, many studies upon *P. rudis* require the removal of individuals from the substratum, putting at risk their survival since they no longer have the ability to produce new byssus threads (Ruppert *et al.* 2004). Given the protected status of *P. rudis*, it is important to develop methodologies that relate total shell length with the dimensions of its non-buried parts. Hence, the main objective of this study was to use a non-invasive methodology to determine the size structure of *P. rudis*, using individuals resident off Matiota beach, on São Vicente Island (Cabo Verde) as a model. It is expected that this methodology will contribute to the expansion of ecological studies, such as spatial distribution and population size and structure in other habitats and locations, especially marine protected areas.

## MATERIAL AND METHODS

This study was carried out on Matiota beach (16° 53' 22" N and 24° 59' 46" W) in Porto Grande Bay, São Vicente Island. This coastal area is located between Ponta de João Ribeiro to the NE and Ponta do Morro Branco to the SW. The bay is semi-circular with a diameter not exceeding 4 km, is characterized by calm waters protected from strong sea currents and with depths varying from 10 to 30 m (Almada 1993).

Data collection was performed by diving using SCUBA along strip transects, 3 m wide and 60 m long. Five transects were laid perpendicular to the shore, 15 m apart, and anchored to the seabed with removable weights. Each individual of *P. rudis* identified within each strip transect was marked with a flag connected to a bottom weight and a surface float for counting and *in situ* data collection. The depths at which individuals were located was determined with a graduated cable (to the nearest 5 cm), which also had a basal weight and a surface float, and the length and maximum width of each shell (to the nearest 1 cm) above the sediment-seawater interface was determined with a ruler (Fig. 1A and B). After data collection, all the markers and transects were removed.

A total of 18 shells were sampled haphazardly to establish relationships between observable morphometric characteristics and total shell length. Biometric data were obtained from the 18 collected shells (Fig. 1C) with a calliper. The variables measured were: (i), posterior shell length (PL), that is the height of the shell projecting above the substratum measured underwater; (ii), the height of the shell buried within the substratum (AL); (iii), maximum shell width (MW); (iv), minimum shell width (mw) and (v), maximum shell length (ML) measured in the laboratory.

The maximum shell length of each individual was estimated based on the adaptation of the method applied by García-March & Ferrer (1995) to *Pinna nobilis* Linnaeus, 1758. Maximum shell size was obtained by summing the posterior and anterior lengths (ML= PL+AL). The relationship between anterior height and minimum width (AL= a+b*mw) or maximum width (AL= a+b*MW) was established through linear regression using the PAST software version 3.15 (Hammer *et al.* 2001). The regression significance was tested using ANOVA at a confidence level of 95%. These two equations were then used to estimate the theoretical sizes (and errors) in relation to the obtained data.

This strategy aimed at allowing the non-destructive assessments of population size structure at Matiota Beach. With the size data of the entire population sampled in the study area it was possible to infer the distribution of the sizes, using 5 cm classes.

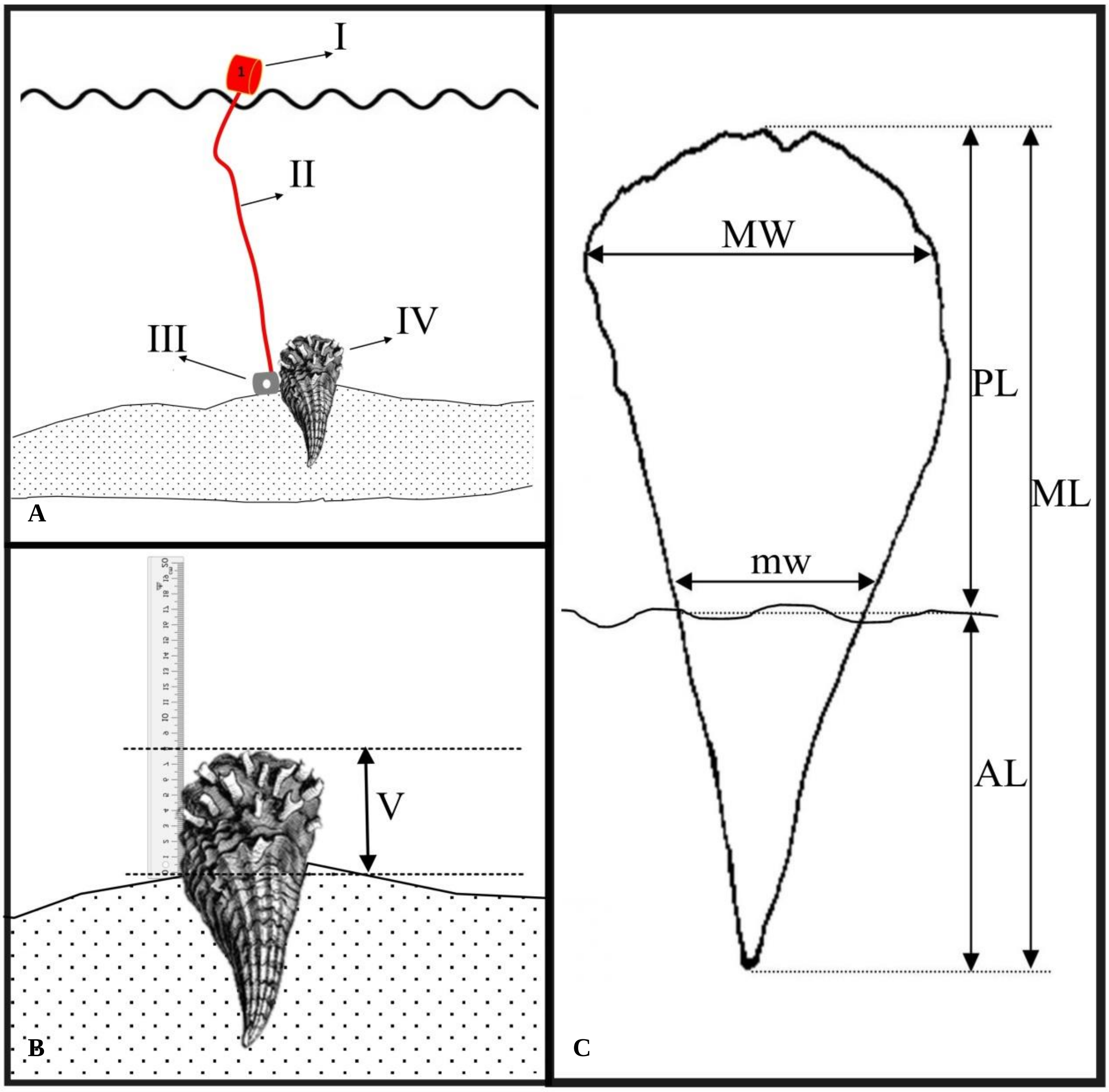


**Fig. 1.** *Pinna rudis* biometric data and sampling scheme. **A**) Sample marking scheme (I, float; II, connecting cable; III, weight; IV, test individual). **B**) Determination of the length of the shell external to the substratum (V). **C**) Illustrative chart with the biometric data used to measure *P. rudis shells* (ML, maximum length; PL, posterior length; AL, anterior length; MW, maximum width; mw, minimum width).

## RESULTS

The entire area covered by this study was 900 $m^2$ and 59 *P. rudis* individuals were collected, including 53 living and 6 dead ones. These were found at depths of between 1 and 3 metres (standard error, SE ± 25 cm) with a density distribution of between 2 and 3 m. The ANOVA results revealed that both models were statistically significant (mw-PL: $F_1$= 8.73, $p$ = 0.009, MW-PL: $F_1$= 5.32; $p$= 0.03). The best linear regression ($R^2$= 0.59) was obtained between minimum width and anterior shell height (mw-AL). The uses of MW-AL linear regression may be less precise ($R^2$= 0.50) because the values are more dispersive (Fig. 2A and B). The plotting of the residues showed a smaller dispersion in the mw-AL ratio than MW-AL, which showed a greater accuracy in terms of overall size estimation (Fig. 2C and D).

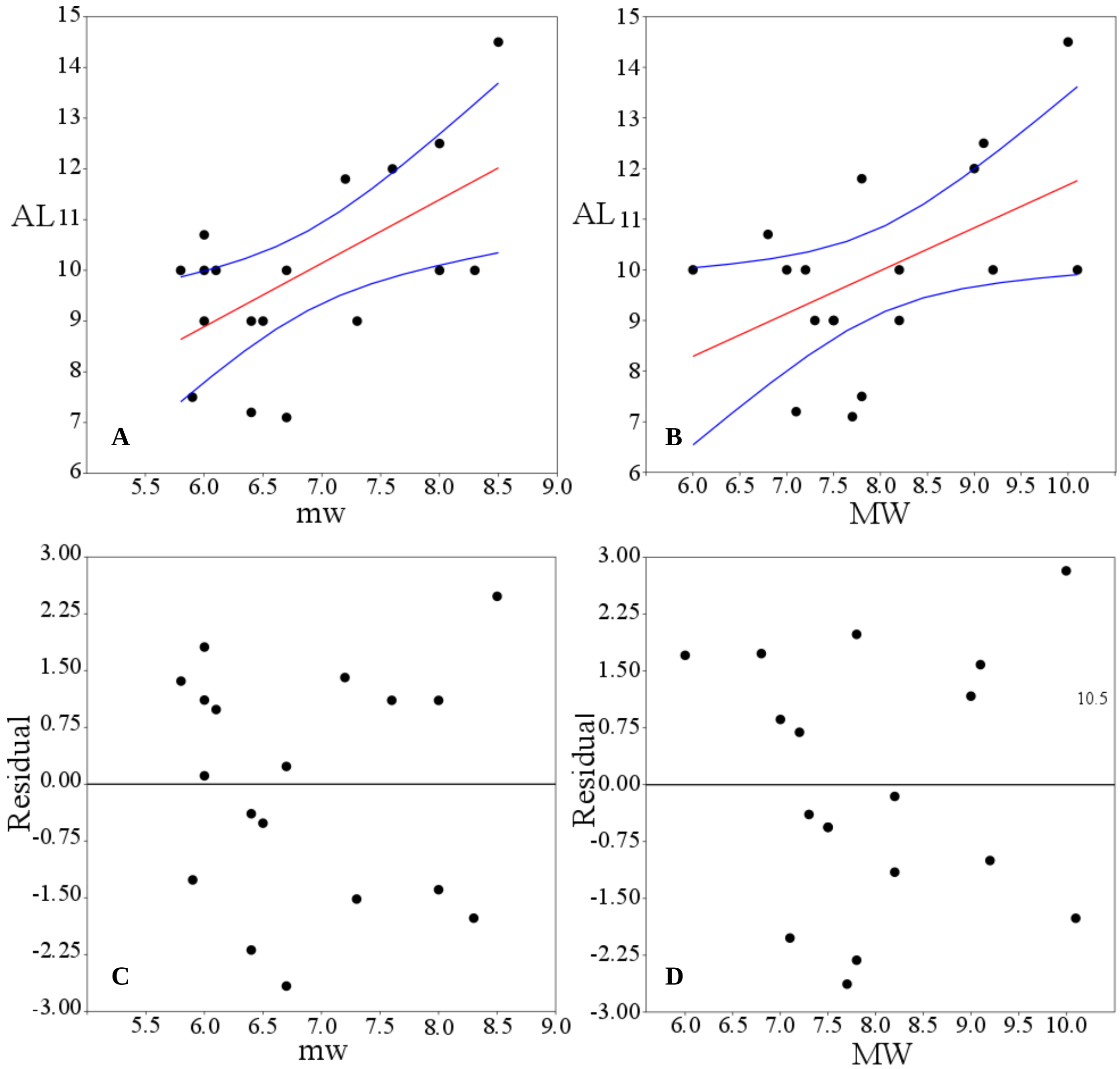


**Fig. 2.** Linear regression and residual graphs of the measurements obtained. **A)** Correlation between minimum shell width (mw) and anterior shell length (AL) (AL= 1.35* mw+ 1.38); and **B)** between maximum shell width (MW) and anterior shell length (AL) (AL= 0.85* MW+ 3.21); **C)** dispersion of the residuals obtained from the relationship between mw-AL; and **D)** dispersion of the residuals obtained from the relationship between MW-AL. The red lines represent the correlation between AL-mw and AL-MW, and the blue lines show the 95% confidence interval of the data.

The errors of the total sizes estimated for each linear relationship revealed that the correlation between MW-AL was less efficient in estimating total shell size (Table 1).

The equation for total size estimation was then established based on the relationship between mw-AL: ML= PL+1.25 mw+1.38.

The *P. rudis* average size estimated for the study area was 19.1 cm ± 0.64 cm (sample size, n= 59), and frequency analysis of the total sizes showed that 95% of the distribution was encompassed between 10 and 25 cm (Fig. 3).

**Table 1.** Measurements obtained from and estimated for 18 *Pinna rudis* individuals. ML, maximum shell length; AL, anterior length below the sediment-water interface; mw, minimum width; MW, maximum width; $AL_1$(mw), anterior length estimated from the equation based on mw ($AL_1$= 1.25mw+1.38); $AL_2$(MW), anterior length estimated from the equation based on MW ($AL_2$= 0.85MW+3.21); $ML_1(AL_1)$, maximum length estimated from $AL_1$ ($ML_1$= PL+$AL_1$); $ML_2(AL_2)$, maximum length estimated from $AL_2$($ML_2$= PL+$AL_2$); $ML_1$ - ML, $ML_1$ error; $ML_2$ - ML, $ML_2$ error.

| **Observed data** | | | | **Estimated data** | | | | **Error** | |
|---|---|---|---|---|---|---|---|---|---|
| ML | AL | mw | MW | $AL_1$(mw) | $AL_2$(MW) | $ML_1(AL_1)$ | $ML_2(AL_2)$ | $ML_1$-ML | $ML_2$-ML |
| 17.00 | 10.00 | 8.00 | 9.20 | 11.38 | 11.00 | 18.38 | 18.00 | -1.39 | -1.00 |
| 16.50 | 10.00 | 6.00 | 7.00 | 8.88 | 9.14 | 15.38 | 15.64 | 1.11 | 0.86 |
| 15.00 | 9.00 | 6.50 | 7.30 | 9.51 | 9.39 | 15.51 | 15.39 | -0.52 | -0.39 |
| 13.10 | 7.20 | 6.40 | 7.10 | 9.38 | 9.22 | 15.28 | 15.12 | -2.19 | -2.02 |
| 24.50 | 14.50 | 8.50 | 10.00 | 12.01 | 11.68 | 22.01 | 21.68 | 2.48 | 2.82 |
| 15.50 | 9.00 | 6.40 | 7.50 | 9.38 | 9.56 | 15.88 | 16.06 | -0.39 | -0.56 |
| 12.80 | 7.50 | 5.90 | 7.80 | 8.76 | 9.82 | 14.06 | 15.12 | -1.26 | -2.32 |
| 18.50 | 11.80 | 7.20 | 7.80 | 10.38 | 9.82 | 17.08 | 16.52 | 1.41 | 1.98 |
| 20.00 | 12.50 | 8.00 | 9.10 | 11.38 | 10.92 | 18.88 | 18.42 | 1.11 | 1.58 |
| 16.50 | 10.70 | 6.00 | 6.80 | 8.88 | 8.97 | 14.68 | 14.77 | 1.81 | 1.73 |
| 17.00 | 10.00 | 5.80 | 6.00 | 8.63 | 8.29 | 15.63 | 15.29 | 1.36 | 1.71 |
| 15.00 | 9.00 | 7.30 | 8.20 | 10.51 | 10.15 | 16.51 | 16.15 | -1.52 | -1.15 |
| 18.50 | 12.00 | 7.60 | 9.00 | 10.88 | 10.83 | 17.38 | 17.33 | 1.11 | 1.17 |
| 17.00 | 10.00 | 6.70 | 7.20 | 9.76 | 9.31 | 16.76 | 16.31 | 0.23 | 0.69 |
| 15.50 | 9.00 | 6.00 | 7.50 | 8.88 | 9.56 | 15.38 | 16.06 | 0.11 | -0.56 |
| 17.00 | 10.00 | 6.10 | 8.20 | 9.01 | 10.15 | 16.01 | 17.15 | 0.98 | -0.15 |
| 19.50 | 10.00 | 8.30 | 10.10 | 11.76 | 11.76 | 21.26 | 21.26 | -1.77 | -1.76 |
| 13.50 | 7.10 | 6.70 | 7.70 | 9.76 | 9.73 | 16.16 | 16.13 | -2.67 | -2.63 |

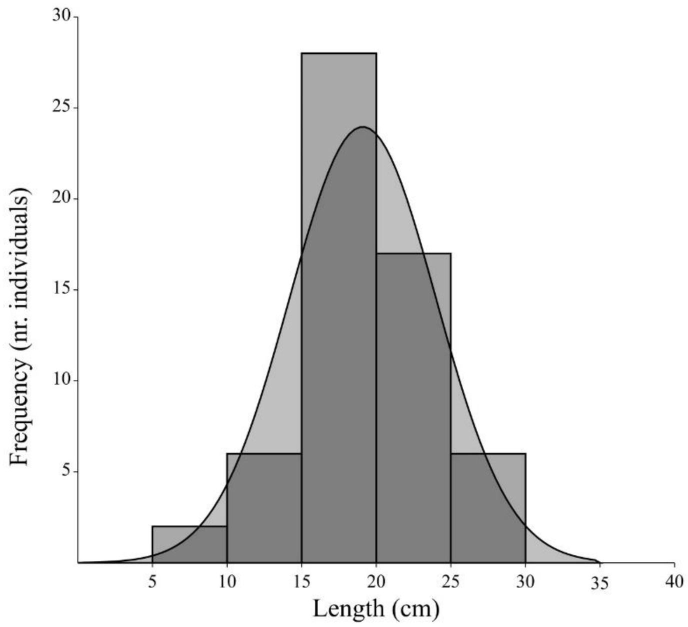


**Fig. 3.** Histogram showing the frequencies of *Pinna rudis* shell sizes in the population resident at Matiota Beach, São Vicente Island, Cabo Verde.

## DISCUSSION

The linear regression between individual total length (ML) and posterior margin length (PL) showed a relatively high determination coefficient when compared with the maximum width (MW) regression. This posterior size measurement to estimate total size is thus more accurate than width parameters, demonstrating a high efficiency that allows the extrapolation of non-destructive biometric data. This methodology is still not accurate (given that the $R^2$ value is low), due to the measurement mode (usually performed using SCUBA) that still lacks and requires an adjustment in the laboratory to minimize errors.

A similar technology has been proposed by García-March *et al.* (2002) to study the growth rate of the *P. nobilis* population from Moraira (Alicante, Western Mediterranean), giving an adjusted growth equation fitted to the measurements obtained with two devices. In spite of different species, the relation obtained for *P. rudis* presented a lower average error value (mean error = 1.30) than those obtained for *P. nobilis* by Garcia March (mean error = 1.79). In both studies this values are more imprecise in smaller pinna shell.

The results have proved the usefulness of this method when there is a need to measure many individuals of protected/cryptic species, although the error demonstrates that measurements are poorly accurate. Such an imprecision may be closely related to the irregularities of the substratum that makes it difficult to obtain an accurate estimation of minimum width. The shell size analysis revealed a wide range of mean sizes (between 15 and 25 cm), corresponding mostly to adults. Also, the minimum size recorded for *P. rudis* was 10.1 cm, and may be related to the non-breeding season (early summer), which decreases the probability of finding smaller individuals (Basso *et al.* 2015). A similar analysis of *P. rudis* shell size was performed by Nebot-Colomer *et al.* (2016), but most of these individuals had shell lengths ranging from 15 to 20 cm. This difference in values may be due to differences in environmental conditions that influence both growth and survival rates. Most individuals were found in caves and in *Posidonia oceanica* beds, at depths of between 4 to 40 metres - different from individuals collected from coral communities at depths of from 0 to 4 metres (Nebot-Colomer *et al.* 2016).

This is the first attempt to estimate the population size structure of *P. rudis* in the Cabo Verde Archipelago. This *in situ* measurement method may be a useful and non-invasive tool for obtaining more *P. rudis* biometric data based on only a single measurement that is easy to assess underwater. In very fragile ecosystems such as Cabo Verdean Marine Protected Areas, this less invasive method can be replicated widely to obtain more biometric data for *Pinna* specimens. More studies are, however, needed to better understand the ecological benefits of *P. rudis*, namely in eutrophic zones, in order to propose efficient management and conservation measures for the entire archipelago.

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# REFERENCES


Almada, E. (1993) *Caracterização Oceanológico das Zonas de Pesca da ZEE do Arquipélago de Cabo Verde*. INDP, Mindelo, Cabo Verde, 27 pp.

Ávila, S.P. (2000) Shallow-water marine molluscs of the Azores: biogeographical relationships. *Arquipélago Life and Marine Sciences*, Supplement 2 (Part A), 99–131.

Barea-Azcón, J.M., Ballesteros-Duperón, E. & Moreno, D. (2008) *Libro Rojo de los Invertebrados de Andalucía*. 4 Tomos. Consejería de Medio Ambiente, Junta de Andalucía, Sevilla, Spain, 1430 pp.

Barreiros, J.P., Pacheco, R.J. & Gonçalves, S.C. (2016) A new insight on the symbiotic association between the fan mussel *Pinna rudis* and the shrimp *Pontonia pinnophylax* in the Azores (NE Atlantic). *Global Journal of Zoology*, 1, 1–2.

Basso, L., Vázquez-Luis, M., García-March, J.R., Deudero, S., Alvarez, E., Vicente, N., Duarte, C.M. & Hendriks, I.E. (2015) The pen shell, *Pinna nobilis*: a review of population status and recommended research priorities in the Mediterranean Sea. *Advances in Marine Biology*, 71, 109–160.

Cosentino, A. & Giacobbe, S. (2006) Shell ornament in *Pinna nobilis* and *Pinna rudis* (Bivalvia: Pteriomorpha). *Journal of Conchology*, 39, 135–140.

Cosentino, A. & Giacobbe, S. (2007) Aspects of epizoobiontic mollusc assemblages on *Pinna* shells. I. Composition and structure. *Cahiers de Biologie Marine*, 2, 187–197.

Cosentino, A. & Giacobbe, S. (2008) Aspects of epizoobiontic mollusc assemblages on *Pinna* shells. II. Does the Mediterranean *P. nobilis* represent an isle of biodiversity? *Cahiers de Biologie Marine*, 49, 161–173.

Dietl, G.P. & Alexander, R.R. (2005) High frequency and severity of breakage-induced shell repair in western Atlantic Pinnidae (Bivalvia). *Journal of Molluscan Studies*, 71, 307–311.

García-March, J.R. (2005) *Aportaciones al conocimiento de la Biología de* Pinna nobilis *Linneo, 1758 (Mollusca: Bivalvia) en el litoral mediterráneo ibérico*. PhD thesis University of Valencia, Spain, 332 pp.

García-March, J.R. & Ferrer, J.F. (1995) Biometría de *Pinna nobilis* L., 1758: una revisión de la ecuación de De Gaulejac y Vicente (1990). *Boletim del Instituto Espanol de Oceanografia*, 11, 175–181.

García-March, J.R., García-Carrascosa, A.M. & Peña, Á.L. (2002) *In situ* measurement of *Pinna nobilis* shells for age and growth studies: a new device. *Marine Ecology*, 23, 207–217.

Gómez-Rodríguez, R. & Pérez-Sánchez, J.M. (1997) Moluscos Bivalvos de Canarias, Ed. Cabildo Insular de Gran Canaria. Las Palmas de Gran Canaria, Spain, 425 pp.

Gómez, R. & Pérez, J.M. (2011) Bivalvia: pp. 301-389, Part 6. In: Rolán, E. (coord). Moluscos y conchas marinas de Canarias. Conchbooks, Hackenheim & Emilio Rolán, Vigo, 716 pp.

Hammer, Ø., Harper, D.A.T. & Ryan, P.D. (2001) PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontologia Electronica*, 4, 1–9.

Nebot-Colomer, E., Vázquez-Luis, M., García-March, J.R. & Deudero, S. (2016) Population structure and growth of the threatened pen shell, *Pinna rudis* (Linnaeus, 1758) in a Western Mediterranean Marine Protected Area. *Mediterranean Marine Science*, 17, 785–793.

Poppe, G. & Goto, Y. (1993) *European seashells. Volume: 2 (Scaphopoda, Bivalvia, Cephalopoda)*. Conchbooks, Haekenheim, Germany, 221 pp.

Richardson, C.A., Kennedy, H., Duarte, C.M. & Proud, S.V. (1997) The occurrence of *Pontonia pinnophylax* (Decapoda: Natantia: Pontoniinae) in *Pinna nobilis* (Mollusca: Bivalvia: Pinnidae) from the Mediterranean. *Journal of the Marine Biological Association of the United Kingdom*, 77, 1227–1230.

Ruppert, E.E., Fox, R.S. & Barnes, R.D. (2004) Invertebrate Zoology: A functional evolutionary approach. *Systematic Biology*, 53, 662–664.

Segers, W., Swinnen, F. & de Prins, R. (2009) Marine molluscs from the Portuguese province of Madeira, Snoeck, Belgium, 611 pp.

Sempere, J.B., Jerez, P.S., Luis, M.V. & Martínez, C.O. (2006) *Evaluación de las Especies Marinas de Interés Ecológico del LIC de las Islas Columbretes*. Servicio de Conservación de la Biodiversidad – Conselleria de Território y Vivenda, Espanha, 117 pp.

Wirtz, P. & D'Udekem-d'Acroz, C. (2001) Decapoda from Antipatharia, Gorgonaria and Bivalvia at the Cape Verde Islands. *Helgoland Marine Research*, 55, 112–124.

Wirtz, P. & D'Udekem-d'Acroz, C. (2008) Crustaceans associated with Cnidaria, Bivalvia, Echinoidea and Pisces at São Tomé and Príncipe islands. *Arquipélago. Life and Marine Sciences*, 25, 63–69.

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