

# SHELL ORNAMENT IN *PINNA NOBILIS* AND *PINNA RUDIS* (BIVALVIA: PTERIOMORPHA)

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*Abstract* Shell sculpture of the Mediterranean bivalves *Pinna rudis* (L.) and *Pinna nobilis* (L.) have been compared by means of parametric and non-parametric univariate statistics, as well as techniques of joint multivariate classification and ordination. Furthermore, a quantitative definition of shell rugosity has been proposed, as an assessable factor potentially affecting anti-predatory defence and epizoic community settlement. Spine density, together with some derived measurements, which describe shell roughness well, best discriminated the two shell typologies and their variation as a function of individual growth. In this respect, *P. nobilis* showed greater individual variability, mainly due to progressive erosion in the largest specimens, in comparison with the noticeable morphologic uniformity of *P. rudis*.

*Key words* Pinnidae, molluscs, morphometry, Mediterranean Sea.

## INTRODUCTION

In bivalve molluscs, both on infaunal and epifaunal species, shell ornament plays several important functions, (cf. Wilson, 1979; Stanley, 1981; Watters, 1993) including a possible anti-predatory effect (Stanley, 1970; Logan, 1974; Feifarek, 1987; Stone, 1998). The family Pinnidae includes semi-infaunal large-sized species (Vicente, Moreteau & Ecoubet, 1980; Combelles, Moreteau & Vicente 1986; Butler, Vicente & De Gaulejiac, 1993; Warwick, McEvoy & Thrush, 1997; Richardson, Kennedy, Duarte, Kennedy & Proud, 1999; Katsanevakis, 2005), in which the shell provides additional space for the settlement of hard bottom assemblages, thus enhancing local biodiversity (Kay & Keough, 1981; Corriero & Pronzato, 1987; Giacobbe, 2002; Giacobbe & Leonardi, 1987). In this respect, remarkable differences in shell ornament of the two Mediterranean *Pinna* species (*P. rudis* and *P. nobilis*) could influence the epibiota and anti-predatory defence.

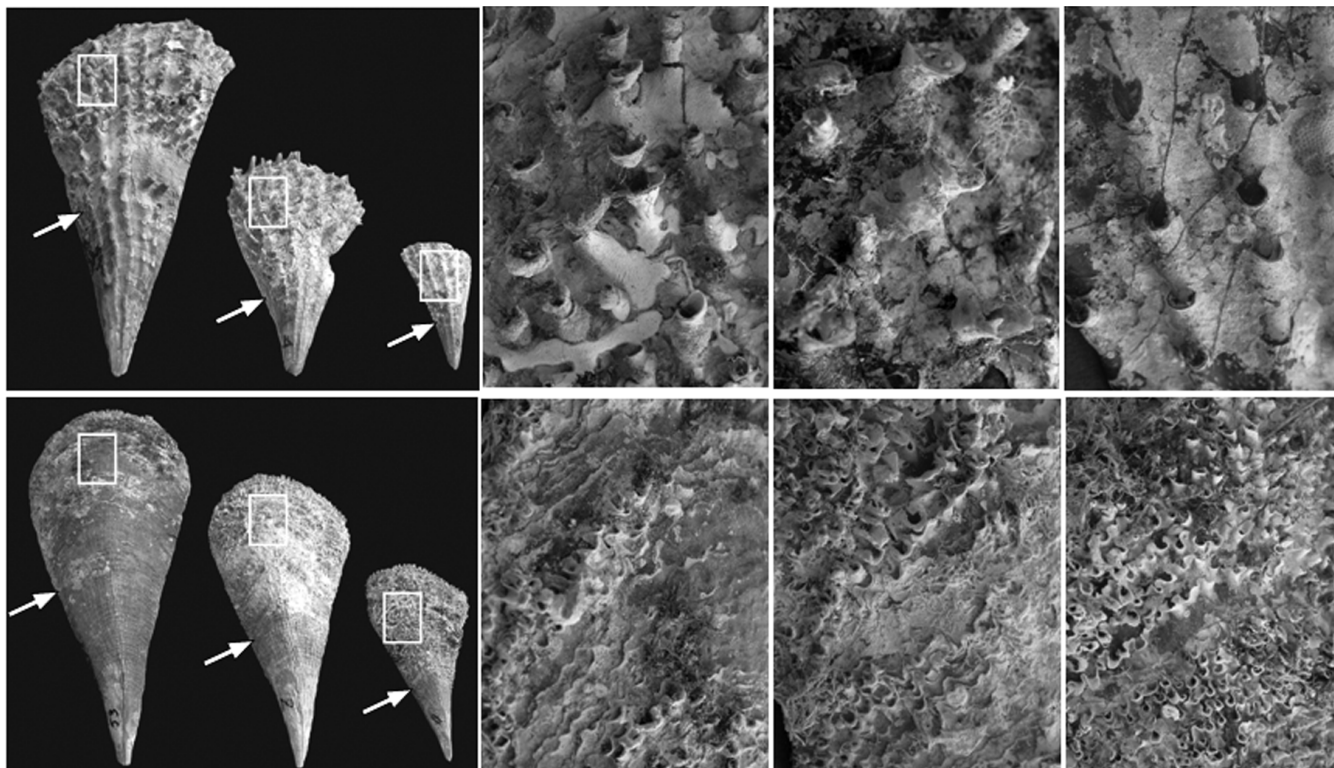
In this paper, a simple methodology to quantify spine density and shell surface roughness is proposed. This will aid the evaluation of variations in spatial complexity within and between species, and support discussion on the possible influences on colonizing biota at the small-scale level.

## MATERIALS AND METHODS

Specimens of *Pinna nobilis* and *P. rudis* were collected from several Mediterranean regions

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(Western and Eastern basins, Adriatic sea, North Africa) and their shell sculpture was examined. From the populations living in the Strait of Messina (Central Mediterranean Sea), 11 specimens per species were selected, representing all shell-sizes and shell erosion grades. The occurrence of shell scars due to mechanical damage was also evaluated. The main axis length (L, mm), maximum width (W, mm) and length of shell portion above the sea floor (UL, according to García March, García-Carrascos & Peña, 2002) were measured by means of a calliper ( $\pm 0.5$  mm), for all twenty-two specimens. The emergent plane surface (EPS, mm<sup>2</sup>) was estimated using a flexible net, cutting out the concave side of one valve and drawing the contours on millimetric paper (see also Morales Alamo, 1990). The shell surface covered by spines (sensu Stone, 1998) in respect to the "bare" part was also evaluated. For each specimen, a mean spine surface (MSS, mm<sup>2</sup>) was assessed by the average height and width of ten spines (five complete and five eroded), approximated to a hollow cylinder, whose sum of inner and outer surfaces was calculated by the formula:  $MSS = 2h_{av}w_{av}$  ( $h_{av}$  mean spine height;  $w_{av}$  mean spine width). Spine density was assessed on a 25 cm<sup>2</sup> surface through a stiff 1 cm<sup>2</sup> grid mesh; the product 2 x spine density x spiny surface furnished the estimated spine number (ESN) for both the valves, expressed as  $\log_{10}$ . The product ESN x MSS provided the total surface due to spines only (TSS, mm<sup>2</sup>). The sum TSS + EPS provided the total emergent surface (TES, mm<sup>2</sup>). Relative roughness (RR = TSS/EPS) was performed in order to quantify the extent to which the secondary order space increases the



**Figure 1** Photographs displaying two different shell morphologies of the *Pinna* species. In the left main pane, three diverse growth stages (large, medium, small size) for each *Pinna* species are shown, with particular attention to the evolution of conch sculpture. White arrows indicate the mark left by sediment on the shell surface, indicating the level of the buried portion. In the three secondary right panes, successive magnifications of conch sculpture (referred to the white frames) detail the different spine morphologies of the two bivalve species. Top row: *Pinna rudis* (L.). Bottom row: *Pinna nobilis* (L.).

plane surface, whereas roughness percentage ( $R\% = TSS/TES * 100$ ) assessed the breadth of the spiny part in respect to the bare part.

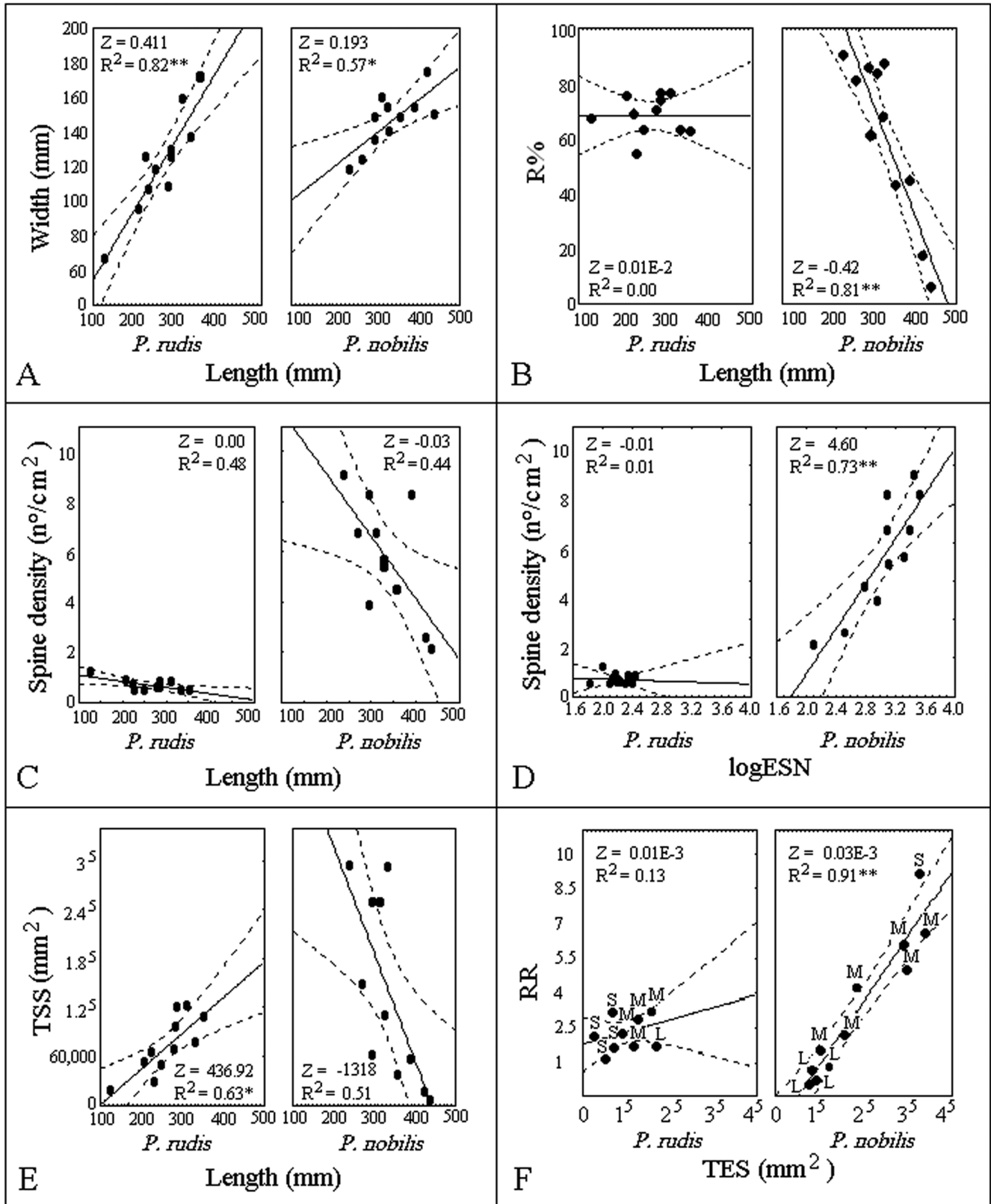
All these parameters were tested by means of conventional univariate statistics for small-group samples (Sokal & Rohlf, 1994). Shapiro-Wilks' W-test for normality and Brown-Forsythe's F-statistic for homogeneity of variance were applied, and a t-test for small independent samples was carried out. Not normally-distributed parameters were tested by means of non-parametric, rank-based comparisons (Mann-Whitney's U-test). In addition, multiple correlations (Pearson's  $r$  coefficient) were performed within each species-group.

Multivariate analysis was performed with PRIMER v5 statistical package (Clarke & Gorley, 2001; Clarke & Warwick, 2001). Number of scars (not transformed), RR, spine density (double-root transformed data), length, width, TSS, TES, R%, ESN (log-transformed data) were computed; these measurements were also normalized in order to turn them into an a-dimensional

scale. The similarity matrix was computed by the Euclidean distance method (group average linkage); cluster analysis and non-metric MDS were performed. Randomisation tests (ANOSIM 1-way and 2-way) were also employed to assess clustering factors considered alone (number of scars) or joined (species and size-class).

## RESULTS

Some remarkable differences in the morphology of the two *Pinna* species are recognizable in the series of three size-classes of both species shown in Fig.1. In our samples, sizes were from 128 mm to 354 mm for *P. rudis* and from 241.5 mm to 438.5 mm for *P. nobilis*; specimens smaller than 250 mm were considered small (S), up to 350 mm medium (M), and greater than 350 mm large (L). Apart from shell shape, some sculptural differences were evident, in *P. rudis* the spines are large and regularly arranged, whilst they appear minute and much closer in *P. nobilis*. In both species, a



**Figure 2** Multiple scatter plots of some measured and estimated morphometric parameters. Interpolation line (least square method) and 95% confidence limit (dashed lines) are displayed. Determination coefficients ( $R^2$ ) and slope ( $z$ ) are also indicated (\* $p < 0.01$ ; \*\* $p < 0.001$ ).



mark left by the sediment on the shell surface distinguishes the portion of the shell emerging from the bottom. The most significant differences in shell shape were seen in the width/length ratio (Fig. 2A), which indicated a more allometric growth in *P. rudis* ( $r = 0.91$ ;  $z = 0.411$ ) with respect to *P. nobilis* ( $r = 0.75$ ;  $z = 0.193$ ), and with greater variability in the latter. Variations in shell ornament were related to specimen size and concerned the roughness percentage (Fig. 2B), which decreased in relation to growth in *P. nobilis* ( $r = -0.90$ ), whilst no significant variation occurred in *P. rudis* ( $r = 0.00$ ). The observed decrease of spine density in larger specimens (Fig. 2C) was more evident in *P. nobilis* ( $r = -0.67$ ;  $z = -0.025$ ) than in *P. rudis* ( $r = -0.69$ ;  $z = -0.002$ ). Nevertheless, spine density linearly increased in respect to the expected spine number (Fig. 2D) in *P. nobilis* ( $r = 0.86$ ), while it did not change in *P. rudis* ( $r = -0.08$ ). Trends of total spine surface in comparison with shell size (Fig. 2E) were opposite in the two species, increasing in *P. rudis* ( $r = 0.79$ ) and decreasing in *P. nobilis* ( $r = -0.72$ ), and showing a greater variance in the latter species. The comparison of the 2<sup>nd</sup> order surface (RR) compared to the total emergent surface (Fig. 2F) displayed a significant correlation only in *P. nobilis* ( $r = 0.95$ ), where small and medium-sized individuals showed a higher relative surface compared to the large individuals. This ratio could provide a quantification of shell roughness, which in general was greater in *P. nobilis* compared to *P. rudis* ( $r = 0.36$ ), although an evident reduction of shell ornament according to growth occurred in the former species.

Cluster analysis and nm-MDS distinguished the two species at three units of Euclidean distance, although *P. nobilis* samples were grouped in two clusters, compared to the *P. rudis* specimens, which grouped in one cluster (Fig. 2A). These three clusters did not include three specimens (the two largest *P. nobilis* and the smallest *P. rudis*), each separated at a greater distance (five units). Clusters agreed perfectly with the 2d-MDS (stress 0.09), where two different morphometric trends were recognizable in accordance with species (Fig. 3B). *P. rudis* small specimens best discriminated with respect to the medium and large sized ones, whilst the largest *P. nobilis* are clearly separated from the medium and small specimens, thus stressing the wider size-dependent variability of *P. nobilis* shell in

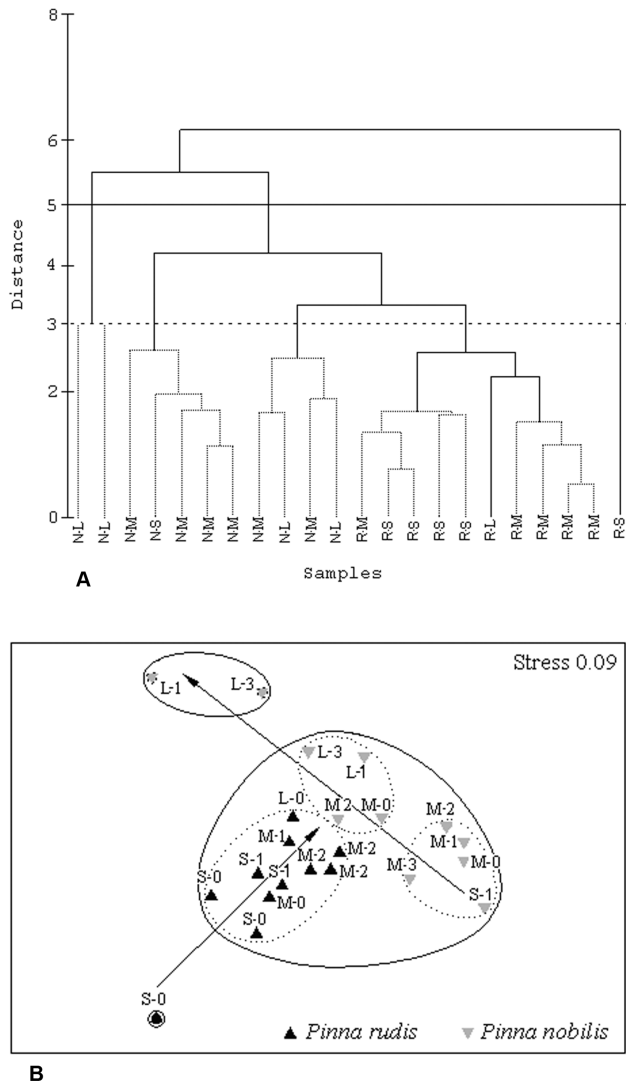
respect to the co-generic species.

The two-way permutation test (crossed with replicates) confirms the species factor as being more discriminating (Global  $R = 0.73$ ,  $p < 0.1\%$ ; permuted statistics  $\geq R: 0$ ) than the size-class factor (Global  $R = 0.49$ ,  $p < 0.1\%$ ; permuted statistics  $\geq R: 0$ ). Pairwise comparisons best discriminate small and large specimens, though with a low significance level due to the reduced number of juveniles in respect to old individuals and, therefore, the number of possible permutations (Global  $R = 0.67$ ,  $p = 6.7\%$ ); medium and large specimens differ significantly (Global  $R = 0.64$ ,  $p = 0.2\%$ ). Finally, small and medium specimens show a higher multivariate morphometric similarity (Global  $R = 0.26$ ,  $p = 3.4\%$ ). In addition, the impact of damage and shell fractures does not seem to influence growth and shell morphology significantly (Global  $R, 0.067$ ;  $p$ -level n.s.; permuted statistics  $\geq R: 182$ ).

## DISCUSSION

Some evident differences in shell ornament of the two *Pinna* species, such as the number and dimension of spines, have been quantified in this paper through a statistical approach. Two different patterns of sculpture and related shell roughness have been highlighted. The *P. nobilis*-type, which is best described by measures such as logESN, spine density, RR and R%, offers a high number of hollows per surface unity. Here, however, shell roughness was greatly variable, mainly due to a progressive reduction of the spiny surface and spine erosion during growth. On the contrary, the *P. rudis*-type showed a more uniform and stable sculpture with significantly lower numbers and lower hollow density, but more lasting in time. Assessments of logESN, RR and R% do not show significant trends according to specimen growth in the *P. rudis*-type. Unexpectedly, although these two patterns involved a different three-dimensional organization of shell-surface, the increase in the total spine surface and the total emergent surface as a function of the growth stage was not significantly different.

In both species, shell-damage substantially did not affect shape and sculpture in the adult shell (cf. Rumohr & Krost, 1991), in accordance with the proven ability of *Pinna* shells to repair (De Gaulejac & Vicente, 1990). Similar to other



**Figure 3** **A** Hierarchical cluster plot of two *Pinna* species samples, according to the Euclidean distance (dissimilarity); slice at 3.07 distance (dotted line) distinguishes group samples in relation to species and, at a greater distance of 5.0 (continuous line), separates some extreme size-class morphologies. Branches and nodes with no remaining structure are highlighted through dotted segments. **B** Non-metric MDS with superimposed clusters shows the high variance of multiple morphometric variables in *P. nobilis*, in contrast to the whole homogeneity (similarity) of *P. rudis*. Arrows indicate the differential progressive trend of morphometric variation in both species from small to large specimens. Capital letters refer to the size-classes; subsequent numbers indicate the shock-marks which occurred.

**R**, *Pinna rudis*; **N**, *P. nobilis*; **S**, small; **M**, medium; **L**, large specimen.

bivalve species (Vance, 1978; Feifarek, 1987), the role of ornamentation in encouraging the settlement of a protective epizoa biota could be suggested, particularly for the young, thickly sculptured, *P. nobilis*. In the largest, smoothed specimens, epibiota covering should be sufficiently structured so as not to need sculpture maintenance. In *P. rudis*, the prominent and sparse spines, which persist in the largest-sized specimens, could have a direct dissuasive function towards predators. On the other hand, the role played by the different shell sculptures in facilitating epibiota recruitment and succession is not yet clear, and warrants further field and experimental investigations.

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