Monitoring of a Posidonia oceanica bed (Punta Manara, Eastern Ligurian Sea, Italy) and the associated molluscs twenty years after: What's new?

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Monitoring of a *Posidonia oceanica* Bed (Punta Manara, Eastern Ligurian Sea, Italy): un confronto twenty years after. What’s new?

Nicola Nurra, Fabio Belci, Rocco Mussat Sartor, Daniela Pessani

Laboratorio di Zoologia e Biologia Marina, Dipartimento di Biologia Animale e dell’Uomo, Università di Torino, Via Accademia Albertina 13, 10123 Torino, Italy

Abstract

The degree of conservation and the extension of the *P. oceanica* bed off P.ta Manara (Eastern Ligurian Sea, Italy) was examined in 1973. It was deeply studied in 1985-86, extending the research to the molluscs community of foliar stratum. We update the investigation in 2006. Therefore our project has spanned a long time interval, something unique and completely missing in *P. oceanica* literature. This research pointed out the structure, dynamics and organization of malacofauna of foliar stratum, determined according to the most important space-time variables: the bathimetric gradient and the periods (seasons). The main aim was to evaluate any change in the *P. oceanica* bed and molluscs community dynamics after 20 years. Samplings were undertaken using an experimental design equivalent to the one used in the past (’85-’86), during 3 periods (Autumn, Early Spring and Late Spring) and across 7 different depths (-5, -9, -13, -17, -19, -21 m). We sampled using the standardized hand-towed net method, using 60 stroke for each sample. Samples yielded 8972 specimens, belonging to 127 taxa (species). The multivariate analyses was applied to both the “historical” and recent data set. The new data showed that the structure of the assemblages significantly differed among depths with a clear distribution gradient from shallow to deep stands. However the composition of the malacological stocks also strictly depends on the sampling period (ANOSIM *R*=0.532, *P*<0.001). The species contributing most to characterize depth at each level and each period (season) were identified. As observed 20 years
before *B. latreillii* remains the dominant species and interestingly the core stock species (sensu Russo) is highly unchanged in terms of both the presence and the abundance of species. In spite of different ecological conditions of the *P. oceanica* bed. Some potential ecological causes of the observed similarities and differences are discussed.

Keywords:

*Posidonia oceanica*, Historical comparison, *Bittium latreillii*, Malacofauna, core stock

1. Introduction

*Posidonia oceanica* (L.) Delile, endemic seagrass of the Mediterranean Sea, play an ecological and multifunctional key role in the coastal ecosystem, because of its vast distribution along the infralittoral bottoms (Peres & Picard, 1964). Hence it has been termed a “priority habitat type” (EEC, 1992). The vagile fauna within the *P. oceanica* meadow constitutes one of the most important components of the ecosystem (Kikuchi & Peres, 1977; Mazzella *et al.*, 1992; Russo *et al.*, 1991b; Gambi *et al.*, 1992, 1995) formed by this seagrass and a relevant fraction of these vagile organisms, is composed of forms linked to the leaf surface (Ledoyer, 1968; Gambi *et al.*, 1991). The molluscs associated to the leaf stratum represent one of the most important component and the most diversified macro-faunistic taxa both in terms of abundance and species richness (Russo & Terlizzi, 1998). The *P. oceanica* meadow off P.ta Manara (SCI - Natura 2000 Code: 1333371) has been studying since 35 years (See Relini *et al.*, 1973; Pessani *et al.*, 1987; Bianchi and Peirano, 1995; Nurra *et al.*, 2005; Diviacco and Coppo, 2007; Vetere *et al.*, 2006, Nurra, 2008; Nurra et *al.*, 2009) and the LBMT has been monitoring since 25 years. The size, the main features, and the typology of the lower limit of the meadow have been known since more than 20 years (Pessani *et al.*, 1987). Recent studies defined the new morphological
outline of the lower limit as erosive-regressive. We drew up a new cartography. Our results confirm the progressive and continuous process of site regression and depletion. This progressive regression and the suffering of the meadow in every investigated sector, confirmed to the main physical, physiografical and phenological parameters (Nurra et al., 2005; Vetere et al., 2006; Nurra, 2008), togheter with hystorical data availability about malacofauna distribution, suggests to verify whether this phenomenon may have influenced tha malacological assemblage along both the bathimetric gradient and during the periods (seasons). We made structure and composition analyses of the malacological community to build a model of malacofauna zonation that was defined by hydrodynamic variations in the site that are strictly associated with the various depths (Idato et al., 1983). Using the concept of core-stock or fundamental stock (Russo et al., 1984b; Russo et al., 1991b), we associated specific-species stock along the bathymetric slope theoretically, stock characterized by high levels of quantitative dominance. Similarly, we defined the typical malacological assemblage for each zone. In the mid-1980s Vetere devised and executed a sampling design (hereafter called past samples) that proved to be accurate tool for understanding the time and space dynamics of the meadow’s foliar malacocoenosis. Over the years his research broadened our knowledge of the foliar-stratum malacofauna in the Punta Manara P. oceanica bed (Vetere, 2005). Our project mainly uses his recently published research results (Vetere et al., 2006) as a base. Hence we were able to compare the malacogical populations in the same site after the passing of 20 years. This gave us a rare opportunity, probably the only one related to taxon and P. oceanica. Considering the great amount of historical data available, the main objectives of the present study are: to verify the current conservation status of the meadow; to define the qualitative and quantitative features of foliar-stratum molluskan community and analyze the assemblages according to space-time variables (depths and sampling
periods) and to carry out a historical comparison between the present malacogical community and the one observed 20 years ago.

2. Materials and Methods

Punta Manara Promontory (44° 15’ 18” N - 9° 24’ 42” E) (Fig. 1) divides the Gulf of Riva Trigoso from the Gulf of Sestri Levante. In the Gulf of Riva Trigoso, there are two principal currents that alternate – the western current (summer) and the eastern (winter). These move vast masses of sediment during the year.

After a preliminary investigation, we identified the meadow and divided it into three belts – the superficial, the intermediate and the deep. As previous studies have demonstrated, these belts represent the three different potential (structural) discontinuity zones of the *P. oceanica* ecosystem. The term is referred to the morpho-structural-discontinuity belts of the *P. oceanica* bed (defined by the bathymetric slope), hydrodynamism, and the substratum typology (defined by the main structural describers).

The three belts have been further divided into sampling stations: 2 in the superficial belt, 2 in the deeper, and 3 in the intermediate one, as randomly chosen in the past samplings.

Samples were collected in 2005-2006 (hereafter called new samples) with the standard semi-quantitative technique of the hand-towed net, as recommended by Russo and co-workers (Russo et al. 1985; Russo and Vinci, 1991) and permanently adopted in 2004 by Gambi and Dappiano.

In order to obtain comparable results, we used the depth curve for sampling stations from the study of Vetere et al. (2006): - 5 m, - 9 m (shallower belt), - 13 m, - 15 m, - 17 m (intermediate belt), and - 19 m, - 21 m (lower belt).

A sample and its replicate were collected at each of the seven stations. There were 3 sampling periods, as in Vetere et al (2006), periods corresponding to significant changes in the annual biological cycle of the plants in the northern part of the Mediterranean Sea: ES: *early spring*; LS: *late spring*; AT: *autumn*.

Overall 42 samples were obtained. Samples were treated with a 7% solution of MgCl₂ in sea water,
and then fixed in 1:10 formaldehyde solution in sea water.

The species-abundance matrices were subjected to multivariate analysis by means of the program PRIMER 6 (*Plymouth Routines in Multivariate Ecological Research*). The data was analyzed independently in each sampling period. The data was also analyzed in the combination of the different periods in order to investigate how time (period) and spatial factors (depth) affect the distribution pattern of assemblages.

Results were achieved through Bray-Curtis (1957) similarity-coefficient-analysis among samples. Data were processed through *Cluster analysis* and *n-MDS* (*non metric Multi-Dimensional Scaling*) (See Warwick and Clark, 1991; Clarke and Warwick, 1994; Clarke and Gorley, 2001), using matrices with the “group average” mode on data converted in square root.

The species exhibiting the best performances during the characterizing and the separating of the various sample clusters were further investigated through the *SIMPER* (*SIMilarity PERcentage analysis*) procedure (*sample discrimination*), using the standard *cut-off* of 90% for the values of similarity/difference among samples (Clarke, 1993; Clarke and Warwick, 1994). The multivariate analysis test *ANOSIM* (*hypothesis testing*) (Clarke, 1993) was given in order to define the importance and the influence of the investigated variables in relation to the samples examined. All the stations and grouping sampling stations in the three main structural discontinuity zones were compared.

3. Results

The meadow forms an irregular zone, approximately 1 km long and 200 m wide and it extends over an area of about 16 hectares. The mean density values (shoots / m²) for each of the examined belts are 542 ± 5.8 in the superficial belt, 247 ± 7.2 in the intermediate and 235 ± 4.2 in the lowest one. We noted a regression of the meadow’s lower limit: 2 - 7 m less than that reported in 1973 (Relini et al., 1973).

3.1 Malacological assemblages - qualitative and quantitative analysis.
We collected a total of 8,972 mollusk specimens belonging to 127 species (96 Gastropods Prosobranchs, 7 Gastropods Heterobranchs, 3 Gastropods Opisthobranchs and 21 Bivalves) in three sampling campaigns. The malacological samples, analyzed by period and depth, were more abundant in autumn (AT), especially in the superficial zone of the meadow (-5, -9 m). At a greater depth, the quantitative data during the different periods was definitely more uniform. There was a slight abundance of the late spring community (LS).

Several families were found, including (from most to least represented) the *Rissoidea* with 27 taxa (4,099 specimens, 45.68%), the *Cerithiidae* with 7 species (3,902, 43.49%), the *Trochidae* with 5 species (129, 1.43%), the *Turbinidae* with 4 species (169, 1.88%) and the *Triphoridae* with 2 species (200, 2.22%).

In the overall sample, the most common species were *Bittium latreillii* (Payraudeau, 1826) (Cerithiidae) (3,724 specimens, 41.5 %), *Rissoa auriscalpium* (Linnaeus, 1758) (Rissoidae) (1,071 specimens, 11.93%) and *Pusillina philippi* (Aradas & Maggiore, 1844) (Rissoidae) (715, 7.96%).

A small portion of the sample included bivalves, including these species: *Hiatella arctica* (Linnaeus, 1767) (Hiatellidae) (23 specimens, 0.25%), *Musculus costulatus* (Risso, 1826) (Mytilidae) (21 specimens, 0.23%) and *Striarca lactea* (Linnaeus, 1758) (Noetiidae) (18 specimens, 0.2%).

Among the 127 recognized taxa, there were only 15 species that reached values of dominance ≥ 1%. These 15 species made up 91.08% of the whole sample’s dominance for the three periods AT - ES – LS. They consisted in the malacological association characterizing the meadow foliar strata in Punta Manara, considered also as the main biocenotic stock.

10 of the 15 species belong to the *Rissoidea* family (43.51%), here represented by 3 genera: *Rissoa* (4 species, 21.47%), *Pusillina* (4 species, 19.91%) and *Alvania* (2 species, 2.14%).

There are 2 species of *Cerithiidae* belonging to the genus *Bittium*, equal to 42.87% of the dominance: *Bittium latreillii* and *B. reticulatum* (da Costa, 1778). This main malacological stock of 15 species is completed by a species of each of the genera *Monophorus*, (2.08%) (Triphoridae), *Jujubinus* (1.36%) (Trochidae) and *Tricola* (1.26%) (Turbinidae).

3.2 Malacological assemblages - structural analysis.
The species-abundance matrix of the sampling periods was subjected to cluster analysis. Through this analysis we described the malacogical association structures in relation to their depths and to the three different zones in the *P. oceanica* bed (Fig. 2).

Through *SIMPER* analysis (*Similarity Percentage - two-way analysis*) we identified the assemblages, described the most important group in each bathymetric zone, and monitored all this in each of the seasons.

We made a characterization of depths. According to this characterization, the values for each species (≥ 3%) consist in the percentage-contributions to similarity – i.e. the results calculated in each of the depths and averaged over the three periods. (The species that are most important are those that contribute the most to Bray-Curtis similarity at each depth averaged across the three periods.) Similarly, we made a specific characterization of periods. According to this characterization, the values consist in the percentage-contributions to similarity, which were recorded for each sampled species in each period and averaged across the various depths.

The species that never reach similarity values > 3% was not included. (Tab. 1)

*SIMPER* has also identified 14 species that contributed the most to the dissimilarity among the three main depths and the three periods. Results are summarized in table 2. Through *ANOSIM* tests we evaluated the significance of the variables that were investigated. In particular, we were able to conduct a cross analysis of the data on the various depths and on the various sampling periods (Tab. 3).

4. Discussion

This research project determined how well conserved the meadow was. It did this through the use of structural parameters – i.e. density and percentage coverage – as well as through the examination of the typology of the lower limit. The meadow has maintained its high index of ecological diversity for the last 20 years, as the monitoring has illustrated. Nevertheless, its lower limit have shrunken more and more. Its central and deepest belts have been covered with more and more mud, thus suffocating some of the plants. Its number of shoots per surface unit (Table 4) has been reduced, as evidenced in the meadow’s most recent density values (Table 4). These
phenomena have caused a progressive fragmentation of the ecosystem and an opening in the ecological niches, those that are specifically more selective under conditions of stability and equilibrium.

A number of anthropogenic factors worked together to disrupt the P. Manara meadow (Nurra et al., 2005, 2009). The shipbuilder Fincantieri’s shipyard and the ship-launching basin, which was dredged periodically, caused an increase in the turbidity of the water and a partial shelving in the meadow’s northern sector.

Illegal trawling and summer boating damaged the southern sector. The intensive anchoring of boats damaged the meadow physically and made it more likely to shrink. All these factors worked towards shrinking the deepest borders of the meadow. The massive exploitation of the intermediate belt greatly increased the entrance of ubiquist and accidental species from contiguous bio-cenosis, species that enriched the biocenotic stock established in these belts of the meadow as a consequence of the so-called "edge effect" (Bell et al., 2001).

Despite these factors, special assemblages of foliar-stratum mollusks persisted, according to semi-quantitative results. These assemblages were made up of a limited number of species (the core stock), those that can reach quantitative dominance. There were 13 species in the core stock during the AT and ES periods and 10 species during the LS period. There were 9 species in each of the assemblages that were found in all 3 periods. The recurrence of these 9 species is evidence that the groups sampled during different periods were strikingly similar in their compositions and that the foliar stratum were highly selective and that they imposed strong qualitative selection on the host mollusks. These nine species were made up of B. latreillii, R. auriscalpium, Rissoa violacea, Rissoa vari-abilis, Rissoa guerinii, P. philippi, Pusillina radiata, Pusillina lineolata and Pusillina incospicua.

The primary aspect emerging from the analysis of the variables is that the mollusk assemblages were significantly different in make up (P < 0.001) at different depths, illustrating that there was a clear pattern (a gradient) of the differences between shallower and deeper belts. Thus the most obvious distribution pattern in sub-tidal mollusk association in P. oceanica beds and hard substrata was a vertical one. The variations among the depths imply that the significant variations in
the stock constitutions depend on the vertical zones (evident for every single period) as well as on the different phases of the biological cycle of the plants (Russo et al., 1984b). These spatial variations — these varying depths — determine that there is a greater qualitative “selection” of the sample. They dis- criminate specific associations according to different depths (global $R = 0.532$, $P < 0.001$) and confirm the zonal pattern.

However, vertical zoning alone is not a complete explanation for all the variability in the patterns of spatial distribution. There is also some variability from site to site (Terlizzi et al., 2003). Within the same sampling site, the principal variations in the distribution of communities depend on the season or the time that the samples were taken — i.e. temporal variations. Temporal variables mainly influence the quantitative aspects of the community — i.e. the differences recorded over the different periods (global $R = 0.579$, $P < 0.001$). The influence of spatial and temporal variables is evidence that there is a characteristic core stock, where the dominant malacological association is made up of fewer species than those in the overall sample. This is a qualitative factor, a finding that seemingly contrasts with the high quantitative dominance of the dominant malacological association.

The data of our most recent research project is purely descriptive. Therefore we obviously cannot explain the similarities and differences we have observed in exclusively ecological terms that would provide an explanation for totally specific assemblages. However, there are significant differences that emerge from the composition of the malacological stocks and from a semi-quantitative comparison between past and present data. The total number of specimens went down drastically from 15,030 in past samples to the current 8972 specimens. The total number of taxa went up 46% — from 87 taxa to the current 127. These are differences that cannot be attributed to the sampling method, which was the same in the past and the present, even though performed by two different divers. As a consequence, this data confirms the hypothesis that the ecological deterioration of the meadow influenced the assemblages in each belt investigated. This deterioration is evidenced by an increase in the number of singleton, accidental species,
up 63% in the new samples. These are species that generally do not thrive in meadows with good structural integrity. Likewise, the total number of species that formed the core stock during the AT period went up from 10 in past samples to 13 in new ones. 8 species were found in both the past and the new assemblages (Table 5).

B. latreillii was the dominant species, wielding a strong influence on all the assemblages, especially in the shallower belt. This dominance was not pointed out in past research (Russo et al., 1984a) because this species was confused with Bittium reticulatum and hence its real spatial distribution was obscured. Despite the progressive deterioration of the meadow, its most remarkable feature is the high degree of conservation in the AT sample over more than twenty years.

Despite its semi-quantitative variations, the species core-stock still displays a certain integrity that can be attributed to the crucial presence of P. philippi, P. radiata, P. lineolata, R. auriscalpium, R. violacea, R. guerinii and Alvania lineata in the characteristic species assemblages. There are many similarities among the core stocks in the ES and LS periods. In the ES period, 9 of the 11 species forming the characteristic association of March 1986, are also found in the core stock found by Nurra in new samples (Table 5). In the LS period, 10 species appear in both past and new malacological groups (Table 5). Each of the two assemblages confirms the high dominance of the two main families, Cerithiidae and Rissoidae. Furthermore, the stocks are generally preserved in these two periods, regardless of the structural variations of the meadow.

Although there are quantitative fluctuations that are hard to interpret ecologically in other than purely descriptive terms, the variability in the past and new coenosis confirms that there is a specific nucleus of the foliar stratum that has basically remained unchanged from a qualitative point of view for the past 20 years. At present, stock conservation is still being maintained, at least partially, in spite of the proven deterioration of the meadow and habitat fragmentation that results from this, despite the opening of new ecological and trophic niches, and despite the massive entrance of ubiquist species.
There is something important to be learned from the main stocks. It is true that extended environmental perturbation of a directly or indirectly anthropogenic nature provokes quantitative effects on the malacofauna foliar stratum (Russo et al., 2002). Despite this, our research brings us to two conclusions: (1) the meadow hosts a faithful community, highly conserved and resistant to the environmental alterations; and (2) foliar stratum mollusks are species with significant stress-tolerant features.

The temporal variable may very well be the most important source of variability within the foliar stratum malacological association, as Vetere’s past structural analysis pointed out. Even so, he noted that there was a strong correlation with the bathymetric gradient within each period. Within this gradient, he interpreted the variations in specific assemblages as they went from the shallower to the deeper belts. Vetere’s interpretation clashes with earlier ones – Russo et al., 1983, 1984b, 1991b; Gambi et al., 1992. The earlier researchers had maintained that the distribution of species is not affected by seasonality, site-exposition and the main edaphic features of the meadow. The results of our study of the P. Manara meadow confirm that the variations within the assemblages along the bathymetric gradient are the greatest source of qualitative variability. The study also revealed that the specific association, defined as the core stock, resists the periodical-seasonal variations although these variations have a meaningful influence (P < 0.001, Table 5) on the main stock’s quantitative features.

One of our aims was to investigate the overall malacological sample in order to explain the role of period-seasonality as a source of variability. To do so, we observed the distribution of the depth points in the n-MDS plot (Fig. 4) as they varied according to season. Consequently, we tried to find out if there were any tendencies in what was happening among the depth-points within the mollusk assemblages over the three periods. We found two tendencies: one tendency corresponded to the period-season and the other that corresponded, at least partially, to the bathymetric gradient. We found these tendencies both in the past samplings and the new ones.

In past samplings Vetere (Fig. 3) found that there was a tendency depending on the depths and the seasons according to which there was a structural organization in the assemblages that was stronger in ES and LS than in AT. This happened because two
intermediate stations (−13 and −15) were separated during AT. There is an influence of the temporal variable that is evident in the depth-order in the n-MDS plot of the new samples (Fig. 4). However, the spatial variable needs to be taken into account too: there is a tendency in the coenosis that corresponds partially to the bathymetric gradient that yields the same data in each period.

According to our recent research, the samples from AT (autumn), unlike those from ES and LS, are influenced both by the bathymetric gradient and the season, a characteristic that helps distinguish this group from the other two. We concluded that there were specific associations tied in with the three periods in both studies. These associations make up 90% of the dominance value and therefore should be defined as the specific core stocks. These core stocks of the three annual periods in the new samples are similar qualitatively to those in the past samples. Thus the core stocks have been conserved significantly over the years. The Cerithiidae and the Rissoidae gastropod families (hence, B. latreillii) dominate.

This distribution of the malacological-associations is one that is consistent with the vertical zoning model of the malaco fauna foliar stratum proposed by Idato et al. (1983). There are three easily recognizable assemblages that are ordered along the three main belts of the meadow. This is an effect that is produced by the influence of the spatial variable (depth).

The temporal variable also influenced the composition of the assemblages in the historical and the recent studies and thus the temporal variable should be considered an important source of semi-quantitative variability. This is an influence on the quantitative and structural variability of the malacological associations of the foliar stratum that is possibly linked to the core-stock species’ cycles of settlement and reproduction cycles.

This research project can help clarify the key role that mollusks play in the P. oceanica meadows as indexes of the changes caused by the pressures made by human beings. The meadow contains a malacological association that has maintained its stability and equilibrium over the years in the face of a partial deterioration in the ecological parameters of the meadow. Nevertheless, the fact that this association has endured does not rule out the chances that human-caused disturbances may bring on radical changes in the malacological stocks, allow alien species to enter, and change
the real population of the foliar stratum once and for all. Further research based on the comparison of historical and current data will help clarify many aspects of this research project.

References


Figure 1.
Punta Manara promontory (Sestri Levante, Eastern Ligurian Sea). Grey points: sampling points.

Figure 2.
Cluster analysis of sampling depths according to the main sectors in all the sampling periods. Up, upper sector; Mid, middle sector; Low, lower sector; AT, autumn; ES, early spring; LS, late spring.

Figure 3.
Non-metric multidimensional scaling (n-MDS) Plot of sampling depths of P.ta Manara bed in 1985–86. AT, autumn; ES, early spring; LS, late spring.

Figure 4.

Non-metric multidimensional scaling (n-MDS) Plot of sampling depths of P.ta Manara bed in 2006. AT, autumn; ES, early spring; LS, late spring.

Table 1.

SIMPER analysis (two way crossed): characterization of sampling depths and periods. For each species of the core-stock is reported the percentage contribution to similarity (in bold, values ≥ 3%) averaged over the periods for the depth characterization and over the sampling depths for the periods characterization. In brackets it is reported the total similarity expressed for each depth and period. AT, autumn; ES, early spring; LS, late spring.

Table 2.

SIMPER analysis (two way crossed): dissimilarity values between depths and periods (in brackets) and percentage contribution to dissimilarity (in bold, values ≥ 3%) of the main biocoenotic stock species. AT, autumn; ES, early spring; LS, late spring.

Table 3.

ANOSIM test (two-way crossed analysis) to define differences between depths (mediated through periods) and between sampling periods (mediated through depths). In bold minimum and maximum values. AT, autumn; ES, early spring; LS, late spring.

Table 4.

Comparison between Manara ’85–86 and Manara 2006 with number of sampled species and P. oceanica foliar density (shoot/m²) in the three sectors of the bed.

Table 5.

Biocoenotic core-stock comparison between Manara 2006 and Manara ’85–86 among the sample periods. n, number of specimens, D%, quantitative dominance. AT, autumn; ES, early spring; LS, late spring.

<table>
<thead>
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<th>Period characterization</th>
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<td></td>
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Table 2.

Tests for differences between depth groups
(across all period groups)
Significance level of sample statistic: 0.1%
Global R: 0.532

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<td>- 5 vs 15</td>
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Table 3.

Tests for differences between period groups
(across all depth groups)
Significance level of sample statistic: 0.1%
Global R: 0.579

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Table 4.

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<th>Density/m² 2006</th>
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Table 5.

AT Period

P.t.a Manara 2006       P.t.a Manara 1985-'86

<table>
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<th>Species</th>
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Total D% 89.6

ES Period

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Total D% 91.60

LS Period

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Total D%                        Total D%