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MACROZOOBENTHIC ASSEMBLAGES AND LEAF EPIPHYTES ASSOCIATED WITH A *POSIDONIA OCEANICA* MEADOW RESTORED

POPOLAMENTI MACROZOOBENTONICI ED EPIFITI FOGLIARI ASSOCIATI A UNA PRATERIA DI POSIDONIA OCEANICA *RESTAURATA*

Abstract - *The restoration of marine phanerogams is one of the key points of the Italian Green Deal to promote the recovery of associated biodiversity and ecosystem services. However, long term studies on the effectiveness of restored meadows to recover the pristine conditions and to sustain the associated biological communities are still scarce. Macrozoobenthic assemblages (polychaetes, molluscs, crustaceans, and echinoderms) and leaf epiphytes (encrusting algae, erected algae, bryozoans, hydroids, foraminifers, spirobids, and ascidians) were investigated in a* Posidonia oceanica *meadow restored in 2009 at Ischia Island (Southern Tyrrhenian Sea). The samplings were performed in 2018 to compare associated organisms of the meadow restored with those of the neighbouring natural* P. oceanica *meadow. Lower values of species richness and abundance were observed in the macrozoobenthic assemblages inhabiting restored meadow if compared to the assemblages of natural meadow. No differences of leaf epiphytes of* P. oceanica *were highlighted between restored and natural meadow.*

Keywords: Posidonia oceanica*, restoration, macrozoobenthos, leaf epiphytes, monitoring.*

Introduction - *Posidonia oceanica* (L.) Delile meadows are facing severe decline across the Mediterranean Basin, particularly in heavily urbanized coastal areas (Duarte *et al.,* 2008). Over the past 50 years, coastal human activities have led to the loss of between 11% and 52% of the originally documented area occupied by *P. oceanica* (Marbà *et al*., 2014; Telesca *et al*., 2015), even if, according to some authors, this decline occurs exclusively in a number of Mediterranean areas (de los Santos *et al.*, 2019).
The restoration of marine phanerogams is one of the key points of the Italian Green Deal to promote the recovery of associated biodiversity and ecosystem services. However, long term studies on the effectiveness of restored meadows to recover the pristine conditions and to sustain the associated biological communities are still scarce. In this regard, macrozoobenthic assemblages and leaf epiphytes were investigated in a *P. oceanica* meadow actively restored through transplantation in the Southern Tyrrhenian Sea. Our research aims to explore the long-term ecological response of transplanted *P. oceanica*, comparing the associated benthic organisms of the meadow restored with those of the neighboring natural *P. oceanica* meadow.

Materials and methods - Sampling was carried out in September 2018 at Ischia Porto (Italy, 40.746° N, 13.950° E) in a transplanted area (named R3) as well as in the neighboring natural *P. oceanica* meadow at 9 meters depth. Sampling was performed after nine years from the end of the restoration activities, which were carried out in 2008-2009 as a compensatory measure within an Environmental Impact Assessment (EIA) procedure. Transplantation was carried out on sand using the cement frames technique (La Porta *et al*., 2023). Macrozoobenthic assemblages were sampled using an air lift over a standardized area of 400 cm^2, collecting three replicates in both restored and natural meadow. In addition, five orthotropic shoots were sampled in two sub-areas in both restored and natural meadow for the laboratory analyses (Buia *et al*., 2003). Macrozoobenthic samples were sorted (mesh 0.5 mm) into polychaetes,

molluscs, crustaceans, and echinoderms and the organisms collected were counted and classified to the lowest possible taxonomic level (i.e. species). In order to describe the structure of assemblages of leaf epiphytes, groups defined as encrusting algae, erected algae, bryozoans, hydroids, foraminifers, spirobis, and ascidians were analyzed. In this regard, the percentage covered by algae and animal groups was estimated on the apical portion (last 15 cm) of the internal side of the two outer leaves of each shoot (Piazzi *et al.,* 2015). Data were analyzed by univariate and multivariate statistical methods using PRIMER 6 (Plymouth Routines in Multivariate Ecological Research) and PAST (PAlaeontological STatistics) 1.97.

Results - The macrozoobenthic assemblages accounted 918 specimens belonging to 175 taxa. Species richness of macrozoobenthic assemblages in restored meadow (45.3 ± 9.0 SD, S as taxa · sample^{-1}) showed significative statistically difference (Kruskal-Wallis test $p<0.05$) if compared with the natural meadow, characterized by higher values (59.3 ± 3.5). Differently, abundance didn't show significative statistically difference between restored meadow (140.7 ± 53.0 SD, N as ind. · sample^{-1}) and natural meadow (165.3 ± 24.1). Likewise, Shannon index in restored meadow (3.26 ± 0.19 SD, H') showed significative statistically difference (Kruskal-Wallis test $p<0.05$) if compared with the natural meadow, characterized by higher values (3.74 ± 0.08). Standard deviation calculated for species richness, abundance and Shannon index showed highest values in restored meadow (Fig. 1).

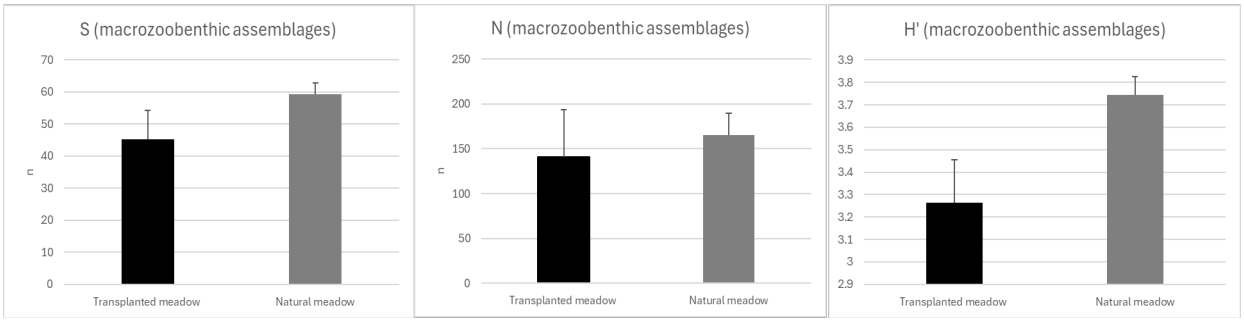

Fig. 1 – Species richness (S), abundance (N) and Shannon index (H') of macrozoobenthic assemblages in *P. oceanica* restored and natural meadow.
Ricchezza di specie (S), abbondanza (N) e indice di Shannon (H') del popolamento macrozoobentonico nella prateria di P. oceanica *ripristinata e nella prateria naturale.*

In particular, crustaceans were the group with the highest species richness in both the restored meadow (23 ± 1.0 SD, S as taxa · sample^{-1}) and the natural meadow (26.7 ± 5.5), followed by the group of polychaetes (restored meadow: 12.7 ± 1.1; natural meadow: 16.7±9.1) and molluscs (restored meadow: 8.7 ± 6.1; natural meadow: 14.3 ± 9.4). Echinoderms were the group with the lowest species richness (restored meadow: 1.0 ± 1.0; natural meadow: 0.7 ± 1.1).
Otherwise, crustaceans were the group with the highest number of specimens (restored meadow: 81.0 ± 18.3 SD, N as ind. · sample^{-1}; natural meadow: 90.7 ± 16.9) and echinoderms were the group with the lowest number of specimens (restored meadow: 1.7 ± 1.5; natural meadow: 2.7 ± 4.6). Intermediate values of abundance were observed by the group of molluscs (restored meadow: 37.6 ± 34.3; natural meadow: 37.6 ± 40.2) and polychaetes (restored meadow: 20.3 ± 7.6; natural meadow: 34.0 ± 14.5).
Cluster analysis on macrozoobenthic assemblages highlighted statistically significant evidence of genuine clusters, in all nodes of the dendrogram (SIMPROF test $p<0.05$), which reflects differences between restored and natural meadow. Samples collected in

the restored meadow are grouped in the dendrogram respect to the samples collected in the natural meadow (Fig. 2).

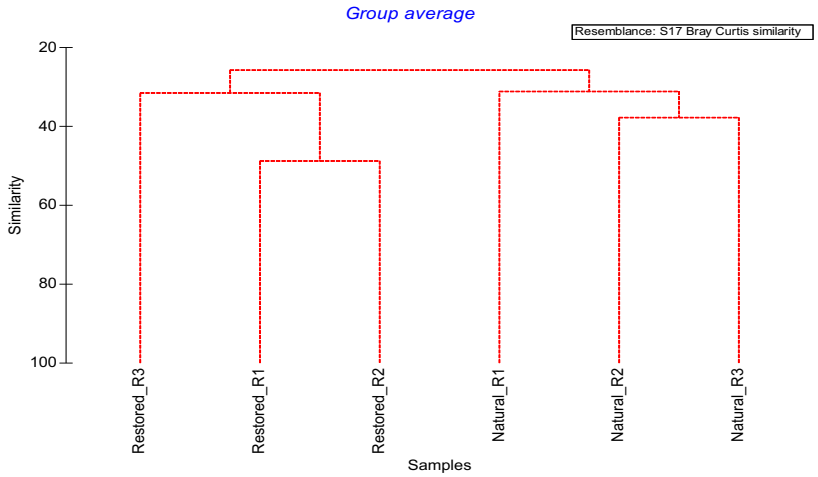

Fig. 2 - Cluster analysis and SIMPROF test (SIMPROF test was applied to each node of the dendrogram, and it highlights, in red, the significative branches) on macrozoobenthic assemblages of *P. oceanica* restored and natural meadow.
Cluster analysis e test SIMPROF (il test SIMPROF è stato applicato a ciascun nodo del dendrogramma ed evidenzia, in rosso, i rami significativi) sul popolamento macrozoobentonico della prateria di P. oceanica *ripristinata e della prateria naturale.*

Encrusting algae were the dominant group of *P. oceanica* leaf epiphytes with the highest percentage cover values among other groups both in the restored meadow (32.5% ± 19.9) and the natural meadow (29.9% ± 17.1), followed by the group of bryozoans (restored meadow: 3.7% ± 5.1; natural meadow: 5.1% ± 7.3) and erected algae (restored meadow: 1.5% ± 3.8; natural meadow: 0.1% ± 0.6). The other groups of hydroids followed by spirorbids, ascidians and foraminifers showed mean values of less than 1% in both restored and natural meadow.

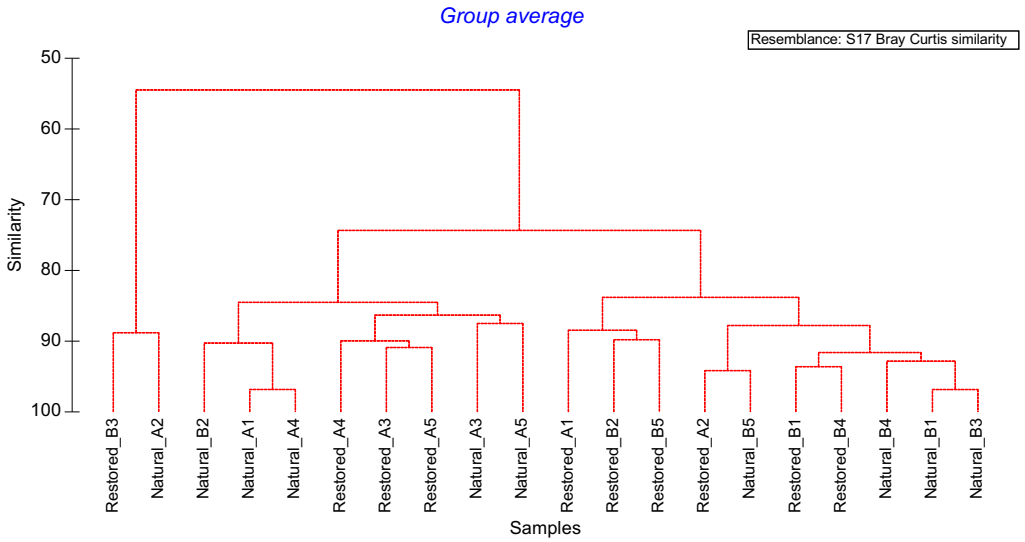

Fig. 3 - Cluster analysis and SIMPROF test (SIMPROF test was applied to each node of the dendrogram, and it highlights, in red, the significative branches) on *P. oceanica* leaf epiphytes of restored and natural meadow.
Cluster analysis e SIMPROF test (il test SIMPROF è stato applicato a ciascun nodo del dendrogramma ed evidenzia, in rosso, i rami significativi) sulla *comunità epifita fogliare della prateria di* P. oceanica *ripristinata e della prateria naturale.*

Cluster analysis on *P. oceanica* leaf epiphytes showed statistically significative cluster structure, in all nodes of the dendrogram, which doesn't reflect differences between restored meadow and natural meadow. All the samples collected in both restored and natural meadow are dispersed in the dendrogram (Fig. 3).

Conclusions - In the long term, after approximately 10 years from the transplantation, the macrozoobenthic assemblages associated to restored meadow showed similarities to the assemblages of neighboring natural meadow. At the same time, the slight differences observed by the indices analysis indicated that the structure of the macrozoobenthic assemblages inhabiting the transplanted meadow was characterized by a lower biodiversity. This pattern could be related to the recovery phase of the restored meadow in the long term as observed in a comparison study on *P. oceanica* structural descriptors collected in both the wider study area of transplanted and natural meadow in Ischia Porto (Bacci *et al*., 2024). Concerning the analysis of leaf epiphytes, the assemblages structure of transplanted meadow was strongly similar to that of natural meadow. Further research on these issues will allow to examine in depth the assemblages patterns occurring in a restored meadow.

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