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RESTORING *POSIDONIA OCEANICA* (L.) DELILE: PRELIMINARY OUTCOMES OF TWO TRANSPLANTING TECHNIQUES

RESTAURO DI *POSIDONIA OCEANICA* (L.) DELILE: RISULTATI PRELIMINARI DI DUE TECNICHE DI TRAPIANTO

Abstract – *Posidonia oceanica* meadows are key ecosystems in the Mediterranean Sea but are increasingly threatened by human activities. Active restoration has become an important tool to enhance recovery, although the effectiveness of different transplanting techniques is still debated. This study provides preliminary results from two transplanting methods tested on the Ligurian coast: R.E.C.S.®-Cocco biomats and metallic stakes. The short-term monitoring highlighted distinct responses between techniques and sites, underlining the crucial role of the first months after transplantation. Stakes ensured higher initial stability, while biomats, after an early decline, showed signs of recovery over time. The results point out the importance of considering local environmental conditions in restoration design. The two techniques can be regarded as two complementary tools: stakes for restoring small, degraded areas, and biomats for larger-scale restoration where conditions are more favourable. Long-term monitoring is essential to fully evaluate their effectiveness.

Keywords: *Posidonia oceanica*; restoration; techniques; NW Mediterranean; Ligurian sea

Introduction - *Posidonia oceanica* meadows represent pivotal ecosystems in maintaining biodiversity and seabed stability in the Mediterranean Sea, constituting a vital resource that provides ecosystem services and functions (Vassallo *et al.*, 2013). Despite being subject to various forms of protection, particularly in recent decades, increasing coastal development and escalating anthropogenic pressures have resulted in substantial direct and indirect damage to seagrass meadows (Boudouresque *et al.*, 2009; Montefalcone *et al.*, 2010).

Posidonia oceanica is a slow-growing plant and, even in areas where the causes of decline have ceased, its natural recovery is a protracted process (Marbà & Duarte, 1998). Despite indications of partial recovery in select regions of the Mediterranean (de los Santos *et al.*, 2019), particularly within areas subject to protection and management regimes, these measures frequently prove inadequate. In recent decades, there has been a notable increase in the focus on enhancing the recovery of this species through the implementation of active conservation strategies, such as the restoration of portions of damaged seagrass meadows. Despite numerous transplantation methods have been tested since the 1990s, the selection of the most effective technique remains a subject of ongoing discussion (Pansini *et al.*, 2022), partially attributable to the absence of standardised protocols. The existing literature increasingly agrees on some important factors for seagrass restoration, such as transplant depth, substrate type and initial transplant density. However, issues related to the type of anchoring, which can strongly

influence the stability and survival of the transplant, remain debated (Pergent-Martini *et al.*, 2024).

The present study offers preliminary findings from an indirect comparison between two transplanting methods: the R.E.C.S.® - Cocco technique and the anchoring of cuttings with metal stakes. The effectiveness of the techniques was evaluated in two sites along the Ligurian coast (Sanremo, Italy). The objective was to evaluate early re-implantations success during the initial months, and to assess the differences between the two techniques.

Materials and methods - The study was conducted in Sanremo (Liguria, NW Mediterranean, Italy), at two sites located approximately 2 km apart: Porto Vecchio and Pian di Poma. Both sites are situated in clearings within *Posidonia oceanica* meadow, at depths between 11 m and 13 m, on a mixed substrate of dead mat and sand. The exposure is south-east, and the area is subject to storms generated by both Libeccio and Scirocco winds. Anthropogenic pressure is high due to coastal infrastructures and the proximity of a marina.

Two transplanting techniques were tested: (i) 5 m × 2 m biodegradable coconut fibre mats combined with a wide-mesh metallic net (R.E.C.S.® - Cocco) (Fig. 1a), and (ii) metallic mulch stakes used to anchor individual cuttings directly to the substrate (Fig. 1b). At both sites, cuttings were collected from material naturally detached by storms or anchoring, retrieved from deposition areas within the Sanremo meadow. Before transplantation, cuttings were cleaned, and necrotic roots were removed. Patch was the experimental unit (approximately 1 m²). Using the biomat technique, each patch consisted of about 20 orthotropic or plagiotropic cuttings, each bearing between one and three shoots. With the stake technique, each patch included 10 plagiotropic cuttings, selected with at least three shoots.

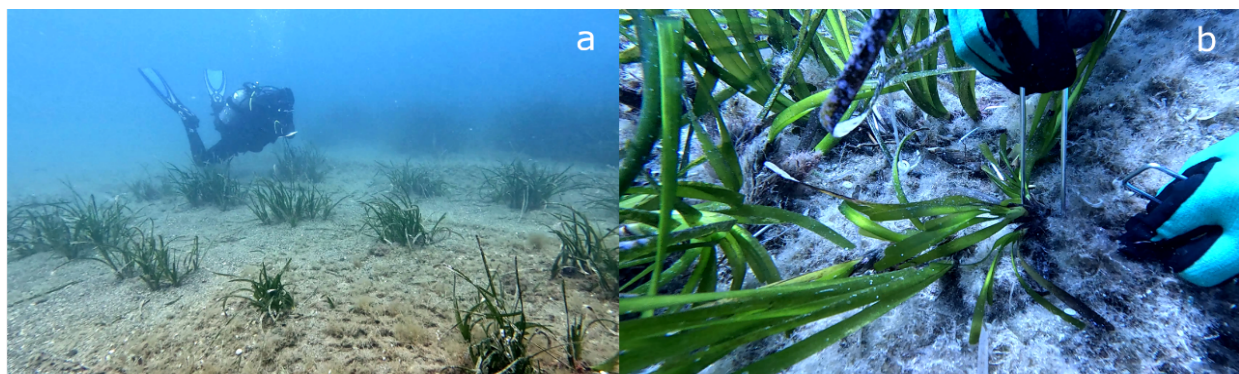


Fig. 1 - (a) R.E.C.S.® - Cocco biomats; (b) metal stakes.
(a) *Biostuoie R.E.C.S.® - Cocco*; (b) *picchetti metallici*.

Transplantation was carried out in July 2024 (T0). Further monitoring surveys were performed in November 2024 (T1), March 2025 (T2), and July 2025 (T3). The experimental design included 3 fixed and orthogonal factors: Site (2 levels: Pian di Poma vs. Porto Vecchio), Technique (2 levels: mats vs. stakes) and Time (4 levels: T0, T1, T2, T3). At each monitoring time, shoot density within each patch was recorded. Patch survival, stability of anchoring structures (biomats and stakes), and qualitative observations on burial were also assessed. Shoot density data were analysed using non-parametric tests due to the non-normal distribution of values. A permutational multivariate analysis of variance (PERMANOVA) was applied to test the effects of Site,

Technique and Time, as well as their interactions. Homogeneity of multivariate dispersion among groups was assessed with PERMDISP. All statistical analyses were performed in R v. 4.5.0.

Results - The trend in shoot density showed clear differences between the two techniques and between the transplant sites (Fig. 2). Despite the initial difference in the number of cuttings transplanted for the two techniques, density at time zero was similar for both techniques and for both sites.

For the biomats technique, the density of shoots after a significant decrease at T1 showed a stable trend at successive monitoring times. In Porto Vecchio, density halved at T1 compared to the initial measurement. At Pian di Poma, despite the similar downward trend, the biomats maintained relatively higher densities than in Porto Vecchio.

For the stakes technique, a stable trend was demonstrated over time since the first monitoring: at Pian di Poma, densities remained higher and, in some cases, increased at T3. In contrast, at Porto Vecchio, although the density at T1 showed a slight decrease than in Pian di Poma, it remained stable in the last two monitoring periods.

The PERMANOVA statistical analysis showed significant effects of the factors Technique ($p < 0.001$, $R^2 = 0.22$), Time ($p = 0.002$, $R^2 = 0.05$) and Site ($p = 0.006$, $R^2 = 0.02$) on shoot density. However, none of the interactions between factors were found to be statistically significant (Tab. 1).

The PERMDISP test verified the homogeneity of the variances for Technique and Site, while a slight heterogeneity emerged for Time ($p = 0.055$).

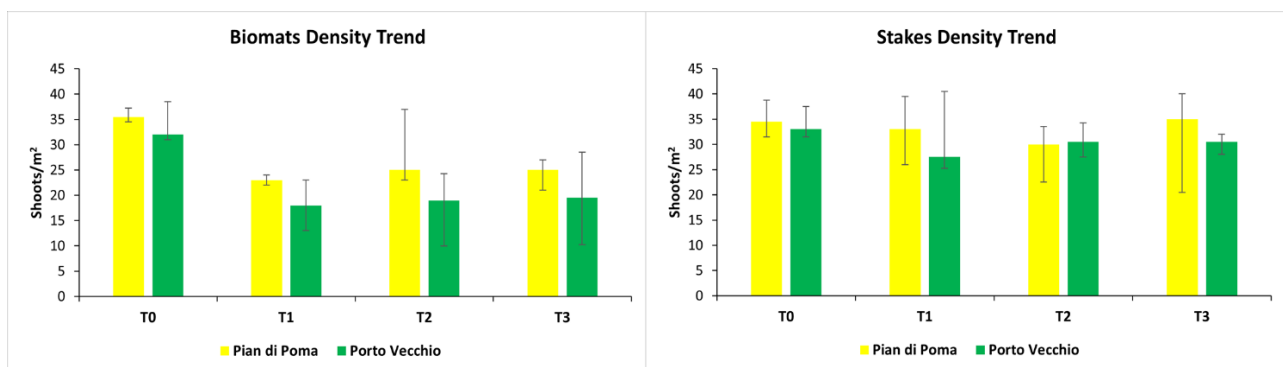


Fig. 2 - Trend of mean shoot density for the two techniques (i.e., biomats and stakes). Error bars represent the Interquartile range.

Andamento della densità fogliare per biostuoie e picchetti. Le barre di errore rappresentano gli intervalli interquartili.

Tab. 1 - Results of PERMANOVA on shoot density according to Technique (biomats vs. stakes), Time (T0, T1, T2, T3) and Site (Pian di Poma vs. Porto Vecchio), and their interactions.

Risultati della PERMANOVA sulla densità dei fasci in relazione alla Tecnica (biostuoie vs. picchetti), al Tempo (T0, T1, T2, T3) e al Sito (Pian di Poma vs. Porto Vecchio) e alle loro interazioni.

Source	SS	Df	R ²	F	P
Technique	7055.20502	1	0.22313	71.30906	0.001 ***
Time	1554.31116	3	0.04915	5.23662	0.002 **
Site	763.83223	1	0.02415	7.72028	0.006 **
Technique:Time	456.46483	3	0.01443	1.53787	0.235
Technique:Site	262.35496	1	0.00829	2.65169	0.107
Time:Site	88.37025	3	0.00279	0.29772	0.825

Source	SS	Df	R ²	F	P
Technique:Time:Site	165.54835	3	0.00523	0.55774	0.635
Residual	21271.75737	215	0.67277		
Total	31617.84416	230	1.00000		

Conclusions - Preliminary results showed that Technique was the factor that best explained the differences observed in shoot density. Stakes showed greater initial stability, while biomats suffered a significant initial decline. However, densities appeared stable for both sites and techniques after T1. The initial selection of both orthotropic and plagiotropic cuttings for biomats and only plagiotropic ones for stakes could play a role in the observed differences since plagiotropic cuttings are faster and more efficient in the rooting process. Monitoring over time demonstrated that the initial months are of paramount importance, particularly in the context of biomats, and that density was not constant over time. The Site factor appeared to influence the success of restoration: in Porto Vecchio a marked decline was observed, probably due to environmental factors (e.g., currents, substrate), and it deserves further investigation. PERMANOVA confirmed that no significant interactions between factors occurred, indicating that they act largely independently. Considering the PERMDISP results, it is essential to recognize the possibility that the Time factor may also reflect seasonal variations and irregular time intervals between monitoring campaigns. From a management perspective, the two transplanting methods can be considered complementary. Stakes provide greater stability and are more suitable for restoring small, degraded areas, whereas biomats, although initially suffer a greater mortality, are more effective for large-scale interventions, under favourable environmental conditions. Continued long-term monitoring is essential to confirm these trends and to fully evaluate restoration success in relation to local hydrodynamic conditions.

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