



## Comparison of different anchoring techniques for seagrass (*Posidonia oceanica*) restoration

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### ABSTRACT

Seagrass ecosystems face threats from multiple human-derived stressors that are causing the loss of these important habitats worldwide. Active restoration using innovative, cost-effective techniques, particularly with uprooted fragments that avoid damage to donor meadows, offers potential for recovery. However, non-ecological factors such as cost, logistics or supply are rarely considered but are essential for decision-making and implementation. This two-year study evaluates the feasibility and effectiveness of four anchoring techniques for securing uprooted *Posidonia oceanica* fragments to the substrate, integrating ecological success with feasibility to identify optimal methodologies. Experiments were conducted in two bays in Mallorca characterized by historical *P. oceanica* presence. Big staples, small staples, bamboo sticks and calcite pegs, were tested to fix fragments with plagiotropic rhizomes. Across sites, small staples and bamboo sticks supported high establishment success (94 %) while having a low carbon footprint, and ease of use. These methods effectively utilized smaller fragments, which are more commonly available, making them practical for large-scale projects. Although larger fragments may develop more new shoots, their limited availability restricts widespread application. As expected, shoot survival declined, a common pattern observed early in *P. oceanica* restoration process, indicating that a two-year period is insufficient to fully assess long-term restoration success. Consistent performance across sites indicates broad applicability of these techniques in Mediterranean shallow, sheltered areas. Non-ecological indicators also highlight the socio-environmental benefits of bamboo sticks and small staples, making them well-suited for sustainable restoration. These findings underscore the need for a balanced, integrated approach that aligns ecological outcomes with efficiency metrics.

### 1. Introduction

The world's coastal ecosystems are under increasing threat from anthropogenic activities, climate change, and habitat destruction (Nichols et al., 2019). Among the critical components of these ecosystems, seagrasses play a pivotal role in providing ecosystem services to humans, including the provisioning of essential habitat for marine life (including commercially important species), nutrient cycling, coastal protection or mitigation of climate change through their role as blue carbon systems (Nordlund et al., 2016). Despite their vital importance, seagrass systems are experiencing significant declines due to local and global disturbances such as rising temperatures, extreme events like storms and heatwaves, urban development, or pollution (Dunic et al., 2021). Consequently, the loss of these seagrass habitats can have strong ecological and socio-economic repercussions (Duarte et al., 2013;

Unsworth et al., 2019), and thus, facilitating the recovery of these habitats is essential.

In response to this environmental crisis, different management actions have taken place in the past few decades to improve conservation of seagrass beds. A key first step to recovery is to eliminate or control the disturbances that caused degradation (van Katwijk et al., 2009), such as improving water quality and reducing nutrient input, or controlling anchoring that causes physical damage and uproots plants (Pergent-Martini et al., 2022). Indeed, these approaches have allowed recovery of seagrass beds in some areas (Santos et al., 2019). Moreover, the rate of natural recovery following the removal of the disturbance has often been significantly faster than previously expected in many seagrass habitats, including *Zostera noltii*, *Z. marina*, and *P. oceanica* (Boudouresque et al., 2021). In addition, there has been increasing interest in enhancing recovery through active restoration (i.e., planting) strategies, especially

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considering that such “passive restoration” or recovery through natural succession is a slow process in marine angiosperms, and can require years for colonization and the establishment of a permanent meadow (Vaudrey et al., 2010; Greening et al., 2011). Furthermore, in many cases, passive restoration has limited success due to various factors (e.g., herbivory, low number of propagules, new disturbances; Balestri and Cinelli, 2003; Infantes et al., 2016). This is why, in most cases, active restoration is undertaken, with the aim of facilitating and promoting natural succession processes. For example, it has been suggested that transplanting the fast-growing seagrass *Cymodocea nodosa* could accelerate natural recolonization through secondary succession, supporting the recovery of the slower-growing *P. oceanica* (Montefalcone, 2024).

Traditionally, restoration has involved transplantation of adult shoots from donor meadows sometimes anchored to different structures such as staples or pegs (reviewed in Van Katwijk et al., 2021). Sexual propagules are also used in restoration, as they increase genetic diversity (Reynolds et al., 2013), but seed-based seagrass active restoration is often limited by seed supply, germination success, variable outcomes among species, and it generally requires more time to reach the structural complexity of a meadow (Van Katwijk et al., 2021; Unsworth et al., 2023).

The success of active restoration projects (understood as the simple survival of plants) is low in marine angiosperms (less than 40 %) when compared to other terrestrial plants or other coastal systems (e.g., corals, mangroves, marshes) and costs remain high compared to terrestrial systems (US \$106k/ha seagrass restoration vs \$2328/ha global average for forest restoration; Bayraktarov et al., 2016; Oakes et al., 2022). Thus, ongoing research endeavors must prioritize the discovery of innovative, low cost, and efficient restoration techniques in order to preserve and restore these valuable ecosystems. Moreover, due to the difficulty, complexity, and high cost of active restoration practices (e.g., SCUBA, use of boats, personnel, etc.), it is also important to consider an assessment of different factors such as logistics, feasibility, replicability, financial cost, or environmental footprint, when considering the most adequate technique to apply.

*P. oceanica* is an endemic Mediterranean seagrass and is highly vulnerable to human activities such as eutrophication, anchoring and fishing (Guidetti and Sala, 2007). It is among the longest-lived seagrass species (Marbà et al., 1996), and also the slowest growing of all, with known values between 1 and 10 cm·year<sup>-1</sup> (Marbà and Duarte, 1998). Nonetheless, *P. oceanica* constitutes one of the most important and productive ecosystems in the Mediterranean coastal zone (Ruíz et al., 2009). *P. oceanica* grows creating a reef from its senescent plant tissue and sediment; these extensive deposits are called ‘matte’, and they act as significant sinks for carbon (Mateo et al., 1997; Mateo and Romero, 1999; Serrano et al., 2012; Monnier et al., 2020; Pergent-Martini et al., 2021; Apostolaki et al., 2022; Monnier et al., 2022). In fact, *P. oceanica* meadows are among the top blue carbon sinks compared to other seagrass species (Lavery et al., 2013; Thorhaug et al., 2017; Röhr et al., 2018; Serrano et al., 2018). As a result of their ecological and socio-economic importance, *P. oceanica* beds are protected under the European Union Habitats Directive (92/43/EEC), and this species has been included among the strictly protected flora species of the Bern Convention on the conservation of European wildlife and natural habitats (1979). Furthermore, it is also included in the List of the Endangered or Threatened species of the Protocol on especially protected areas and biological diversity in the Mediterranean in the context of the Barcelona Convention (1976), and in national and regional regulations in several European countries.

Despite these different levels of protection, *P. oceanica* has undergone human-induced major losses as a result of coastal development, pollution, mechanical damage, introduced species and climate change (Boudouresque et al., 2009; Marbà and Duarte, 2010). Most *P. oceanica* meadow expansion is performed through clonal growth (Migliaccio et al., 2005) while sexual reproduction is generally variable in space and time. Flowering events occur irregularly, more frequently than once

believed, but not necessarily every year and likely linked to warmer temperatures (Montefalcone et al., 2013; Balestri et al., 2017; André et al., 2023; Boudouresque et al., 2024; Tomas et al., 2024) although the successful rate of seedling establishment remains limited (Balestri and Cinelli, 2003 but see Díaz-Almela et al., 2006; Balestri et al., 2017). Given the abovementioned low growth rates, enhancing recovery of *P. oceanica* meadows via active restoration has become of increasing interest to managers. The first active restoration approaches in *P. oceanica* were started in the 1970s–80s (Boudouresque et al., 2021), and consisted of transplants of adult shoots. More recently, in addition to adult plants and seedlings, uprooted fragments (e.g., detached as a result of a storm or by anchoring) are also being used for active restoration of this species (Balestri et al., 2011; Castejón-Silvo et al., 2020; Ward et al., 2020) and the promising results of previous studies make them a good choice for large-scale restoration (Mancini et al., 2021, 2022). Importantly, the use of uprooted adult plant fragments, besides capitalizing on the already existing root and shoot systems, avoids the damage to donor meadows. Using a non-destructive method for obtaining seagrass planting material is not only of great advantage for seagrasses in general, but it is particularly key for those with low growth rates, such as *P. oceanica* (Marbà and Duarte, 1998). Furthermore, if these fragments come from different meadows, it can also enhance genetic diversity, which increases resilience (Reusch et al., 2005; Jahnke et al., 2015).

The main techniques used to anchor *P. oceanica* fragments in situ are the use of metallic staples (Castejón-Silvo and Terrados, 2021), bamboo sticks (Ward et al., 2020), mats made of biodegradable materials (e.g., Piazzini et al., 2021), and “bare” planting (without any anchoring material; see review by Pansini et al. (2022)). These techniques confer certain advantages (e.g., depending on the substrate where fragments are planted, the size of the fragment needed, etc.), and the use of mats, in particular, seems to be a promising technique for large-scale restoration (De Luca et al., 2025). Yet, and importantly, most of them have not been compared under the same conditions, being our understanding of the efficacy of different restoration treatments still incomplete for *P. oceanica*. Moreover, most of the restoration studies in seagrasses do not consider crucial non-ecological factors that can influence the large-scale application of restoration methods such as ease of use of anchoring materials, monetary cost or supply availability. By failing to account for these real-world constraints, existing studies often overlook the practical barriers to implementation at larger spatial and temporal scales. Therefore, in this study we address this gap by experimentally comparing different active restoration techniques for securing *Posidonia oceanica* fragments to the substrate (referred hereafter as anchoring) not only in terms of ecological success but also through a novel assessment of other aspects associated with the restoration approach (e.g., cost, logistics). By integrating ecological outcomes with practical implementation considerations, our approach identifies optimal approaches and offers a more comprehensive framework to guide decision-making in large-scale *P. oceanica* restoration efforts.

## 2. Methodology

### 2.1. Study sites

The study was carried out in two shallow sheltered embayments in Mallorca, Balearic Islands; Portocolom and Formentor (Fig. 1). Both sites are within protected areas of the Natura 2000 Network. The area of *Posidonia oceanica* meadow has regressed in both sites, mainly as a result of anchoring activities, a pressure that has been eliminated by establishing a delimited area within which anchoring is now forbidden. *P. oceanica* meadows alternate with sand gaps and patches of dead *P. oceanica* matte.

### 2.2. Experimental design

Following guidelines by van Katwijk et al. (2009, 2016) and

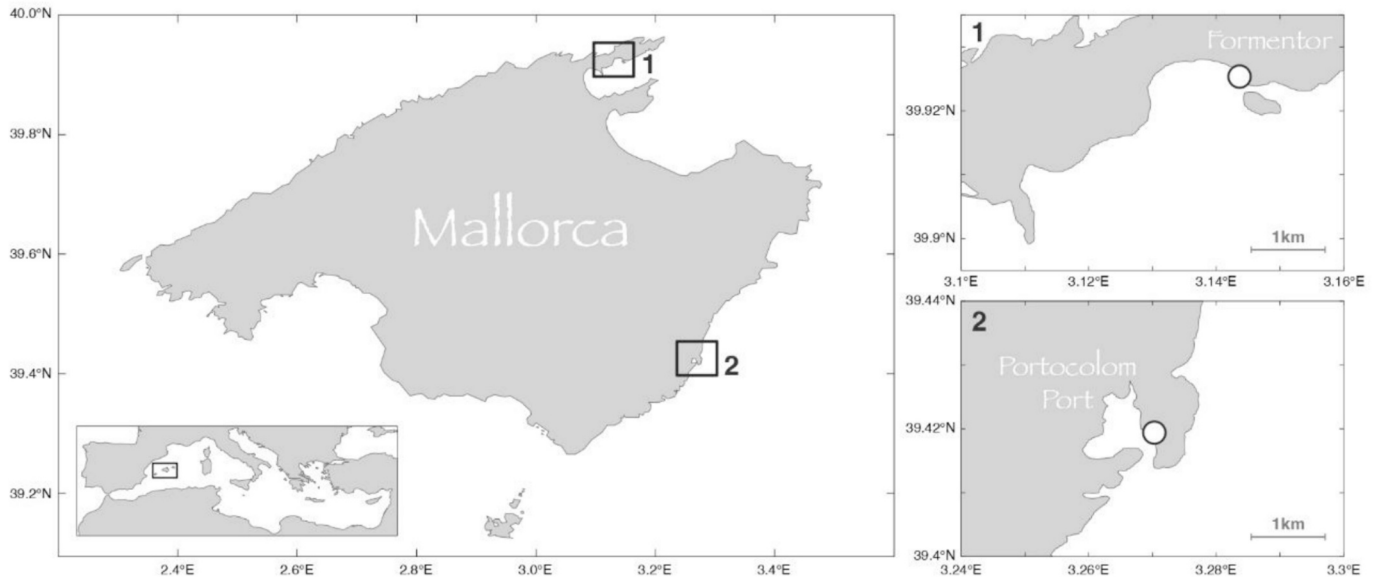


Fig. 1. Study sites and transplant areas in 1) Formentor and 2) Portocolom.

Boudouresque et al. (2021), planting experiments were conducted in shallow zones (between 3 and 5 m depth) that were very sheltered from currents and the predominant waves, where there was evidence of prior existence of *P. oceanica* (indicated by the presence of dead matte), where the removal of the anthropogenic stressor (here, anchoring) had occurred, and where natural recolonization was observed. Experimental plots (1 m × 1 m) were haphazardly placed in areas of approximately 45m<sup>2</sup> of dead matte with *Cymodocea nodosa*.

Previous studies have shown that active restoration with vegetative fragments is more successful with horizontal (i.e., plagiotropic) rhizomes, as these rhizomes are the ones contributing to meadow

expansion, particularly those units containing the apical and at least two lateral rhizomes (Molenaar et al., 1993; Piazzini et al., 1999; Pergent-Martini et al., 2024).

Dead matte areas were chosen for restoration as they are one of the best options for shallow depths, especially when using plagiotropic rhizomes (Pansini et al., 2022; Pergent-Martini et al., 2024).

Detached fragments of *P. oceanica* with plagiotropic rhizomes were collected in situ via snorkeling or using SCUBA in late fall-winter, and planted the same day or the day after collection. Fragments planted the following day were maintained in seawater and exposed to natural light. Fragments were collected from a depth similar or shallower than

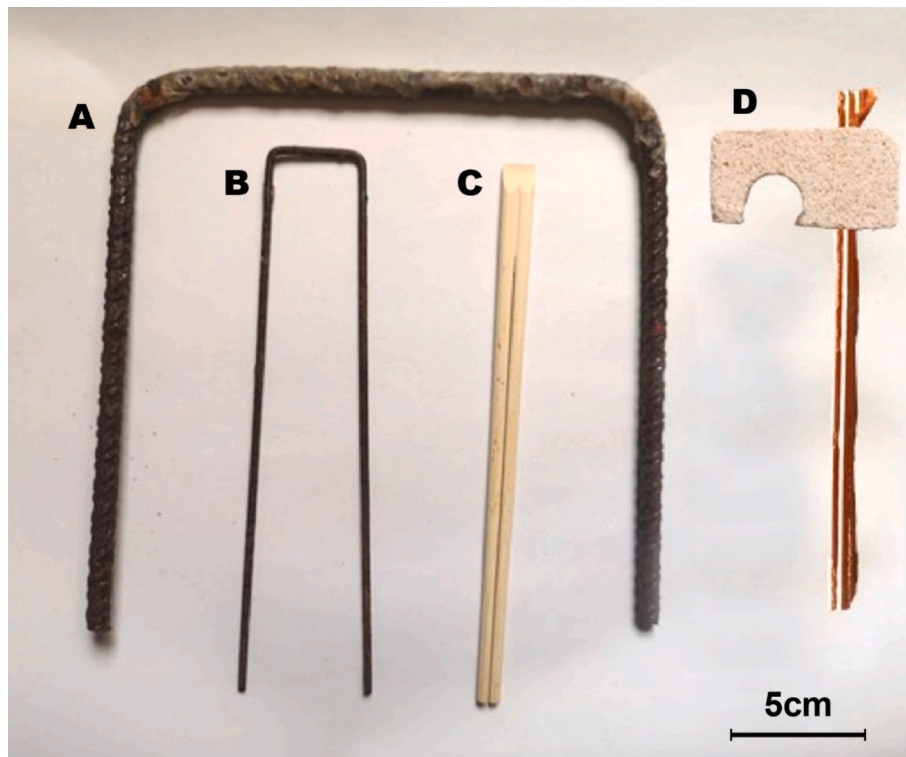


Fig. 2. Photo of anchoring devices tested: (A) big steel staples, (B) small steel staples, (C) bamboo sticks, (D) handmade calcite structure (metal pin inserted for anchoring into the sediment).

planting depth (Molenaar and Meinesz, 1992; Genot et al., 1994). All the fragments selected presented these four characteristics: they were all plagiotropic with an active growing meristem, they exhibited a good and lively general presence (fresh green leaves), presented intact and lively roots (white, pale brown, green) and more than 95 % had at least two additional orthotropic shoots (Pergent-Martini et al., 2024).

We tested four anchoring techniques for active restoration. One technique, previously used in another restoration project in Mallorca, involved securing fragments with a thin rope to large steel staples (20 cm × 20 cm × 0.8 cm) coated in beeswax (Castejón-Silvo and Terrados, 2021) (hereafter referred to as the “big staple” treatment). Another technique used is small steel garden staples (Gardenix; 20 cm × 3 cm × 0.3 cm) (hereafter “small staple”). A third method involved bamboo sticks (Gokoco®; 20 cm × 0.4 cm), while the fourth one consisted of handmade calcite structures (“calcite peg”; 20 cm × 5 cm × 1 cm; plus a floor tile cut as a peg (20 cm × 1 cm × 0.8 cm) Fig. 2). At Formentor, only the first three anchoring techniques were implemented (excluding the calcite peg), with five replicate plots (1 m × 1 m) established per treatment. In Portocolom, all four methods were tested, with four replicate plots (1 m × 1 m) for each method. Each plot contained 16 planted fragments. At the beginning of planting, we recorded the date and location of fragment collection, the date and site of planting (end of October 2022–December 2022 in Portocolom and November 2022 in Formentor), planting depth, and the number of shoots per fragment. Monitoring of presence or absence of fragments and number of live shoots was conducted at both sites in June 2023, October 2023, January–February 2024, May 2024, July 2024, and October 2024.

### 2.3. Non-ecological indicators of restoration

Feasibility metrics were assessed for each anchoring technique. We obtained a suite of quantitative indicators (i.e. weight, buoyancy, number of steps needed to modify the material prior to planting, anchoring devices lost after 2 years, supply time, carbon footprint and direct monetary cost) and qualitative indicators (i.e. supply availability and contribution to local economy). These indicators were weighted in a semi-quantitative scale based on defined criteria from 0 to 2, being 0 the least feasible for active restoration and 2 the most feasible. For example, in terms of diving logistics heavy devices were given a score of 0 and light devices 2, and devices with positive buoyancy were given a score of 0 and those with negative buoyancy 2. Regarding supply logistics, materials available for purchase on-line were given a score of 2 and those handmade by local artisans a 0, and for supply time, materials received in less than one week were given a score of 2 and those received in more than one month were given a 0. In relation to socio-environmental indicators, we calculated carbon footprint as CO<sub>2</sub>e which is the amount of carbon dioxide (CO<sub>2</sub>) emission that would have an equivalent effect on a specified key measure of climate change, over a specified time horizon, as an emitted amount of another greenhouse gas (GHG) or a mixture of other GHGs. For a mix of GHGs, CO<sub>2</sub>e is obtained by summing the CO<sub>2</sub>-equivalent emissions of each gas (IPCC Glossary). Carbon footprint for the production of each anchoring technique and its shipping to the restoration site (Mallorca-Spain) was calculated with 3 different online calculators “2030calculator” (Omnicall Solutions AB, 2024), “footprintcalc” (Segers and Van den Herik, 2024) and “ecocostsvalue” (Stichting Sustainability Impact Metrics, 2021). We used information from the supplier and made some assumptions to the best of our knowledge (e.g., the type of vessel used to ship steel from China to Europe or the European harbor where steel was discharged, the type of vehicle used to transport clay from the Netherlands to Spain). We then scored each anchoring technique according to the carbon footprint (kg CO<sub>2</sub>e). In relation to contribution to local economy, we gave a score of 2 when the cost of the operation (purchase and manipulation) benefits a local supplier (<200 km) and 0 when the cost of the operation does not benefit a local supplier (>200 km). More information of the detailed categorization of indicators is described in Tables S1 and S2. A total

feasibility score was estimated as the sum of the scores for all the non-ecological indicators, with lower values indicating lower total feasibility.

### 2.4. Statistical analyses

Establishment success was measured for each site as presence/absence of fragments at each monitoring time for each anchoring method. Similarly, fragment development at the end of the experiment was categorized into four groups based on shoot count: “net positive” for fragments exhibiting more shoots than at planting time, “net negative” for those with fewer shoots than at the beginning, “net zero inactive” for those that did not change their number of shoots throughout the two years, and “net zero active” for those with the same number of shoots as at planting after two years but had undergone loss of shoots and recruitment of shoots throughout the study period. Establishment success was treated as a binomial variable, and the effects of anchoring method and monitoring time on this variable was analyzed using a logistic regression model with a binomial family. Plot was considered a random factor to account for the inherent variability among plots, and anchoring method and monitoring time were fixed factors. Model fitting was conducted using the maximum likelihood estimation method and evaluated based on the Akaike Information Criterion (AIC; Table S3). Differences in fragment development were treated as a multinomial variable with four levels (net negative, net positive, net zero active, net zero inactive) and the effects of method were evaluated using multinomial logistic regression models. The significance of the coefficients was assessed based on the z-values and associated p-values.

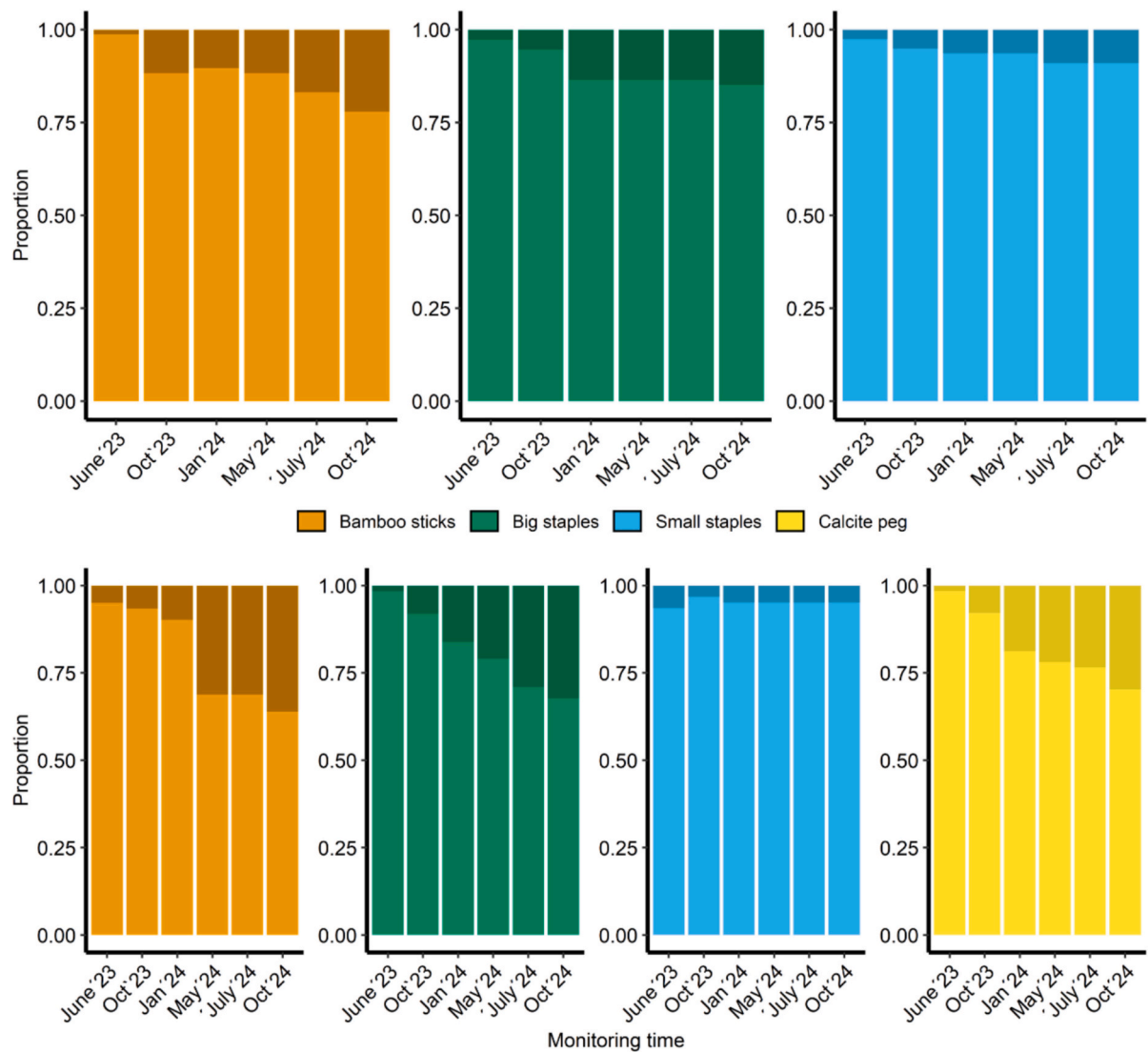
Fragment dynamics were measured as net change in number of shoots per fragment calculated as the difference in number of shoots in each fragment between time of planting and each monitoring time. We also assessed mean number of shoots per fragment at each monitoring time. Effects of anchoring method and monitoring time on fragment dynamics and on the mean number of shoots per fragment were analyzed using Generalized Linear Mixed-Effects Models considering plot as a random factor and anchoring method and monitoring time as fixed factors. The models were fitted using the Restricted Maximum Likelihood method, and model fit was evaluated based on the AIC (Table S3). Box Cox transformations were applied for the mean number of shoots per fragment since residuals of the models were not normal. The significance of fixed effects was assessed using t-tests, with corresponding p-values indicating the strength of the effects.

## 3. Results

### 3.1. Ecological indicators

There were differences in establishment success among monitoring times and anchoring methods at both sites. In Formentor, establishment success significantly decreased across time from 98 % (±1 %) 7 months after planting (June 2023) to 85 % (±7 %) ca. two years after planting (October 2024). In Portocolom there was also a significant reduction of establishment success with time from 93 % (±2 %), one year after planting, (October 2023) to 74 % (±14 %) in October 2024. At both sites, small staples provided significantly higher establishment success (94 ± 1 %) two years after planting compared to the other methods used (84 ± 5 % and 86 ± 5 % for bamboo sticks and big staples, respectively; and 82.8 ± 10.5 % for calcite pegs in Portocolom; Fig. 3, Table 1).

Approximately two years after planting (October 2024) at both sites there was a statistically higher proportion of fragments losing shoots (52 ± 4.5 % and 51 ± 8.2 % for Formentor and Portocolom respectively), than gaining them (34.5 ± 4 % and 34.2 ± 12 %). Both sites also had the same proportion of fragments that had the same number of shoots as when initially planted but undergoing both recruitment and mortality of shoots (“net zero active”, 10.9 ± 1.2 % in Formentor and 10.8 ± 3.3 % in Portocolom) and similarly low proportion of fragments that had the



**Fig. 3.** Establishment success (proportion) of planted *P. oceanica* fragments across time in Formentor (top) and Portocolom (bottom) using the different anchoring methods. Light colors represent established fragments, darker colors represent lost or dead fragments.

same number of shoots throughout the whole study time (“net zero inactive”,  $2.6 \pm 1.4$  % in Formentor and  $4 \pm 2.1$  % in Portocolom). No significant difference in fragment development was found between anchoring methods, except in Portocolom, where the big staple method increased the likelihood of shoot gain (Fig. 4, Table 2).

Fragment dynamics were different among anchoring methods between sites. In Formentor, the overall change in number of shoots per fragment anchored with small staples was significant and negative, showing a tendency to a decline in shoots per fragment after planting. In contrast, the big staple method and bamboo sticks exhibited fluctuating changes in mean number of shoots per fragment but were positive overall, even though bamboo sticks exhibited negative values at later monitoring times. However, in Portocolom, bamboo sticks showed a steady decline over time while the other methods exhibited an overall increase in the number of shoots per fragment (Fig. 5, Table 1).

Both at the beginning and throughout the study period, the mean number of shoots per fragment was statistically different among anchoring methods (Fig. 6, Table 1). The overall mean number of shoots per fragment in the initial population of uprooted fragments that were planted was  $5.4 (\pm 2.9$  SD), ranging from 2 to 22, with 72.5 % of the fragments having 4 or more shoots, and 37.1 % having 6 or more. For all anchoring methods used, planted fragments at the beginning had a median of 4 shoots per fragment, with mean values ranging between ca.

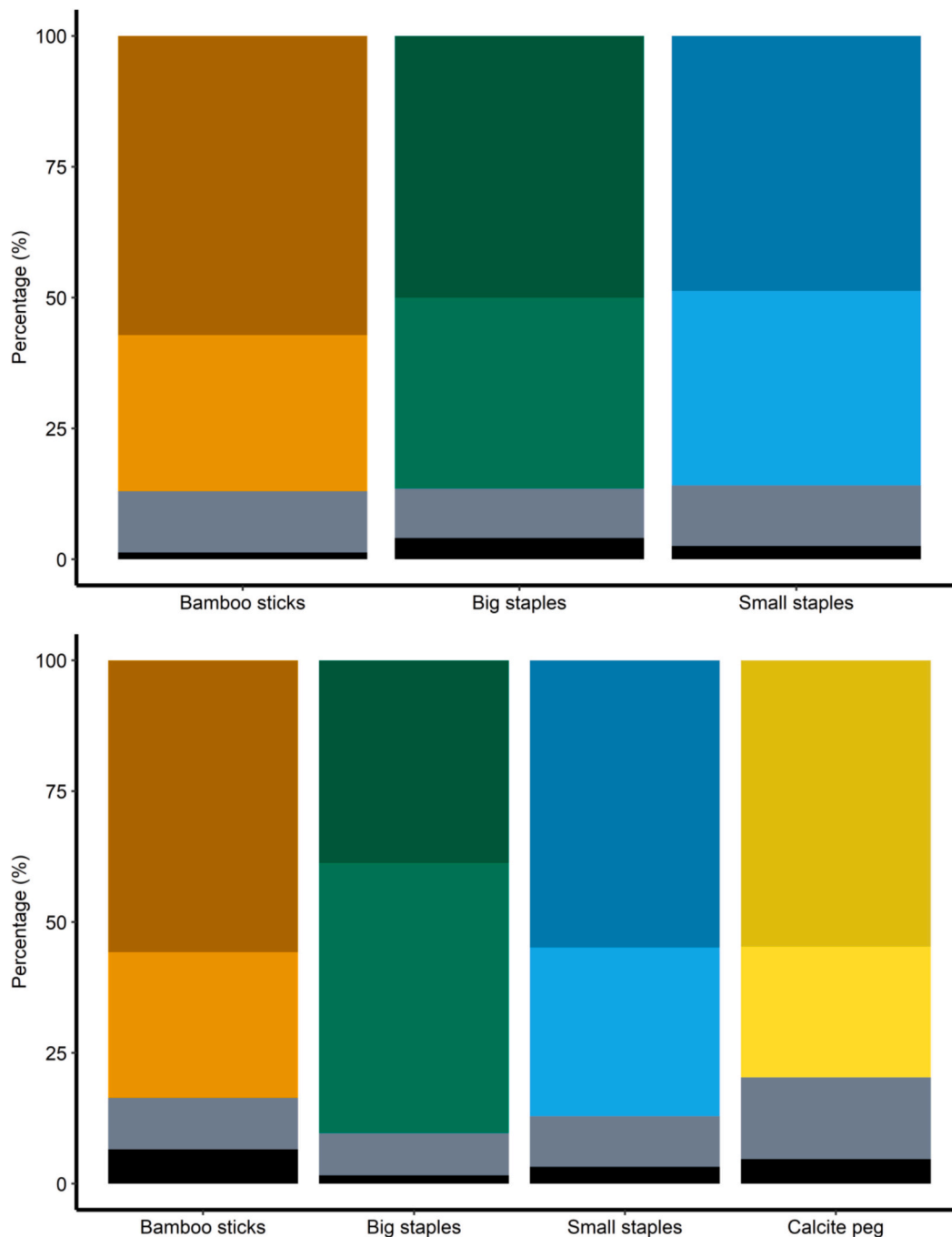
4.5 and 6 among methods ( $5 \pm 2.8$ ,  $4.5 \pm 1.5$  and  $5.5 \pm 3.4$ ,  $6.3 \pm 2.7$ , for bamboo sticks, calcite pegs, small staples and big staples, respectively).

At both sites, fragments anchored with big staples had a statistically higher overall number of shoots ( $7 \pm 4.1$  in Formentor,  $6.3 \pm 3.3$  in Portocolom) exhibiting a consistent high number of shoots throughout the study period, with an increasing trend that changed in July in Formentor to a slight decrease (0.5 shoots per fragment). Bamboo sticks ( $4.6 \pm 2.7$  in Formentor,  $5.2 \pm 3.1$  in Portocolom) and small staples ( $4.8 \pm 2.6$  in Formentor,  $6 \pm 4.9$  in Portocolom) had a statistically lower number of shoots per fragment throughout the study period. In accordance with the results of the change in number of shoots, the mean number of shoots per fragment anchored with small staples show minimal variation over time but started to decline towards later monitoring times (after the first year). Likewise, fragments anchored with bamboo sticks exhibited a more dynamic trend in mean number of shoots declining towards the end or over time. In Portocolom, fragments planted with the calcite peg method had an overall lower number of shoots ( $4.3 \pm 2.3$ ) compared to the other methods. In contrast to Formentor, in Portocolom the numbers of shoots per fragments tend to decrease over time mainly bamboo sticks and calcite pegs (Fig. 6, Table 1).

**Table 1**

Modeling results of General Linear and Mixed Models on different response variables and different predictive variables (monitoring times and anchoring methods). Site indicates the sampling site, levels correspond to the levels of the predictive variables, SE indicates standard error, SD Random indicates the standard deviation of the random factor. Statistically significant results are highlighted in bold. Models selected based on their AIC (Table S3).

Response variable	Site	Predictive variable	Levels	Estimate	SE	z/t-Value	p-Value	Random variable	SD Random	Marginal R2 (%)	Conditional R2 (%)	Family
Establishment success	Formentor	Method + monitoring time	Intercept	3.713	0.569	6.529	<b>&lt;0.0001</b>	PLOT	0.721	13.58	25.35	Binomial
			Big staples	0.161	0.215	0.749	0.4538					
			Small staples	0.758	0.239	3.16	<b>0.0016</b>					
			Monitoring 2	-1.298	0.518	-2.506	<b>0.0122</b>					
			Monitoring 3	1.638	0.503	-3.256	<b>0.0011</b>					
			Monitoring 4	-1.687	0.501	-3.365	<b>0.0008</b>					
			Monitoring 5	-1.95	0.493	-3.953	<b>&lt;0.0001</b>					
	Monitoring 6	-2.138	0.489	-4.374	<b>&lt;0.0001</b>							
	Portocolom	Method + monitoring time	Intercept	2.977	0.368	8.093	<b>&lt;0.0001</b>	0.169	25.02	25.67	Binomial	
			Big staples	0.099	0.194	0.51	0.61					
			Calcite pegs	0.193	0.195	0.992	0.321					
			Small staples	1.656	0.279	5.928	<b>&lt;0.0001</b>					
			Monitoring 2	-0.74	0.421	-1.761	0.078					
			Monitoring 3	-1.355	0.392	-3.455	<b>0.0006</b>					
Monitoring 4			-1.919	0.378	-5.081	<b>&lt;0.0001</b>						
Monitoring 5	-2.072	0.375	-5.525	<b>&lt;0.0001</b>								
Monitoring 6	-2.282	0.372	6.133	<b>&lt;0.0001</b>								
Net change in number of shoots per fragment	Formentor	Method	Intercept	0.383	0.496	0.772	0.4405	PLOT	0.5147	1.03	11.7	Gaussian
			Big staples	0.218	0.226	0.967	0.3339					
			Small staples	-0.533	0.219	-2.44	<b>0.0148</b>					
	Portocolom	Method	Intercept	-0.672	0.175	-3.851	<b>0.0001</b>	0.0001	1.95	1.95	Gaussian	
			Big staples	1.2	0.245	4.902	<b>&lt;0.0001</b>					
			Calcite pegs	0.556	0.242	2.298	<b>0.0218</b>					
Mean number of shoots per fragment	Formentor	Method	Intercept	2.043	0.086	23.751	<b>&lt;0.0001</b>	PLOT	0.144	9	10.2	Gaussian
			Big staples	0.891	0.081	11.041	<b>&lt;0.0001</b>					
			Small staples	0.1	0.079	1.272	0.204					
	Portocolom	Method + monitoring time	Intercept	2.573	0.111	23.213	<b>&lt;0.0001</b>	0.066	5.22	5.41	Gaussian	
			Big staples	0.446	0.111	4.035	<b>0.0001</b>					
			Calcite pegs	-0.409	0.109	-3.738	<b>0.0003</b>					
			Small staples	0.165	0.107	1.538	0.1243					
			Time	-0.083	0.019	-4.343	<b>&lt;0.0001</b>					



**Fig. 4.** Percentage of fragments of *P. oceanica* exhibiting different temporal trends in shoot numbers (i.e. fragment development) in Formentor (top) and Portocolom (bottom) for the different anchoring methods. Light colors represent fragments developing new shoots (net positive), darker colors represent fragments losing shoots (net negative), grey represents fragments losing or developing shoots through time but having a net zero change overall (i.e., same number of shoots at planting time and at the end of the experiment; net zero active) and black represents percentage of fragments remaining with the same number of shoots through time (net zero inactive).

### 3.2. Non-ecological indicators

Regarding non-ecological indicators, bamboo sticks and small staples appeared to be the most feasible anchoring techniques, showing a total feasibility score of 12 and 13, respectively, whereas big staples had the lowest feasibility score (5). The non-ecological indicators that contribute to making bamboo sticks a highly feasible technique are its reduced weight (0.0032 kg), low monetary cost (0.48€/unit), fast (less than one week) and easy supply (available online), low C footprint ( $0.007 \pm 0.0032$  kg CO<sub>2</sub>e) and the inexistence of steps needed prior to

planting. Its feasibility score is reduced by its positive buoyancy and low contribution to the local community. The indicators that contribute to the high feasibility score of small staples are its negative buoyancy, low monetary cost (1.36€/unit), fast (less than one week) and easy supply (available online) and the low percentage of material loss after 2 years (2.82 %). The low contribution to the local community is the only indicator that would make this method less suitable for a restoration project. Regarding calcite pegs, negative buoyancy and community contribution are the indicators that most contribute to its feasibility score, being the high percentage of material loss (16 %), the restricted

**Table 2**

Modeling results of multinomial logistic regression models on fragment development with anchoring methods as predictive variable. Site indicates the sampling site, SE indicates Standard Error. Statistically significant results are highlighted in bold.

	Site	Fragment development	Intercept	Method: big staples	Method: small staples	Method: calcite peg
Estimate	Formentor	Net positive	-0.649	0.334	0.378	
		Net zero active	-1.587	-0.078	0.147	
		Net zero inactive	-3.784	1.272	0.840	
	Portocolom	Net positive	-0.693	0.981	0.162	-0.090
		Net zero active	-1.735	0.166	0.000	0.482
		Net zero inactive	-2.140	-1.038	-0.693	-0.317
SE	Formentor	Net positive	0.257	0.361	0.356	
		Net zero active	0.366	0.551	0.521	
		Net zero inactive	1.011	1.176	1.245	
	Portocolom	Net positive	0.297	0.401	0.409	0.423
		Net zero active	0.443	0.662	0.626	0.570
		Net zero inactive	0.529	1.149	0.899	0.801
Zscore	Formentor	Net positive	-2.521	0.924	1.062	
		Net zero active	-4.338	-0.141	0.282	
		Net zero inactive	-3.742	1.081	0.675	
	Portocolom	Net positive	-2.333	2.443	0.397	-0.212
		Net zero active	-3.917	0.251	0.000	0.846
		Net zero inactive	-4.049	-0.903	-0.771	-0.396
p-Value	Formentor	Net positive	0.012	0.355	0.288	
		Net zero active	0.000	0.887	0.778	
		Net zero inactive	0.000	0.280	0.500	
	Portocolom	Net positive	0.020	0.015	0.691	0.832
		Net zero active	0.000	0.802	1.000	0.398
		Net zero inactive	0.000	0.366	0.441	0.692

availability (local artisan) and slow supply (one month) and high monetary cost (3.13€/unit) the indicators contributing negatively to its low feasibility score. Finally, for big staples, its negative buoyancy is the most favorable non-ecological indicator while weight (0.9 kg), preparation needed prior to planting, high percentage of material loss (16.4 %), monetary cost (3.4€/unit) and C footprint ( $1.71 \pm 0.74$  kg CO<sub>2</sub>e) are the indicators that more negatively impact their feasibility score (Tables 3, S2).

#### 4. Discussion

The results of this study highlight the differences in establishment success and shoot retention across anchoring methods and monitoring times. Overall, establishment success was high and decreased over time, though small staples consistently provided the highest survival rates compared to other methods. The mean number of shoots per fragment at planting was relatively high when using big staples, which required longer fragments in order to be adequately attached. However, only one third of the initial pool of uprooted fragments had the necessary length. This method also exhibited more promising fragment dynamics gaining shoots over the 2 years albeit some declines in later monitoring times. Fragment development exhibited an overall loss in shoots in most fragments regardless of the anchoring method, except for big staples in one of the two study sites. Non-ecological indicators also revealed key feasibility differences among anchoring methods, with bamboo sticks and small staples emerging as the most practical and cost-effective techniques, whereas big staples, despite their effectiveness in promoting shoot growth, had the lowest feasibility due to their weight, cost, and environmental footprint.

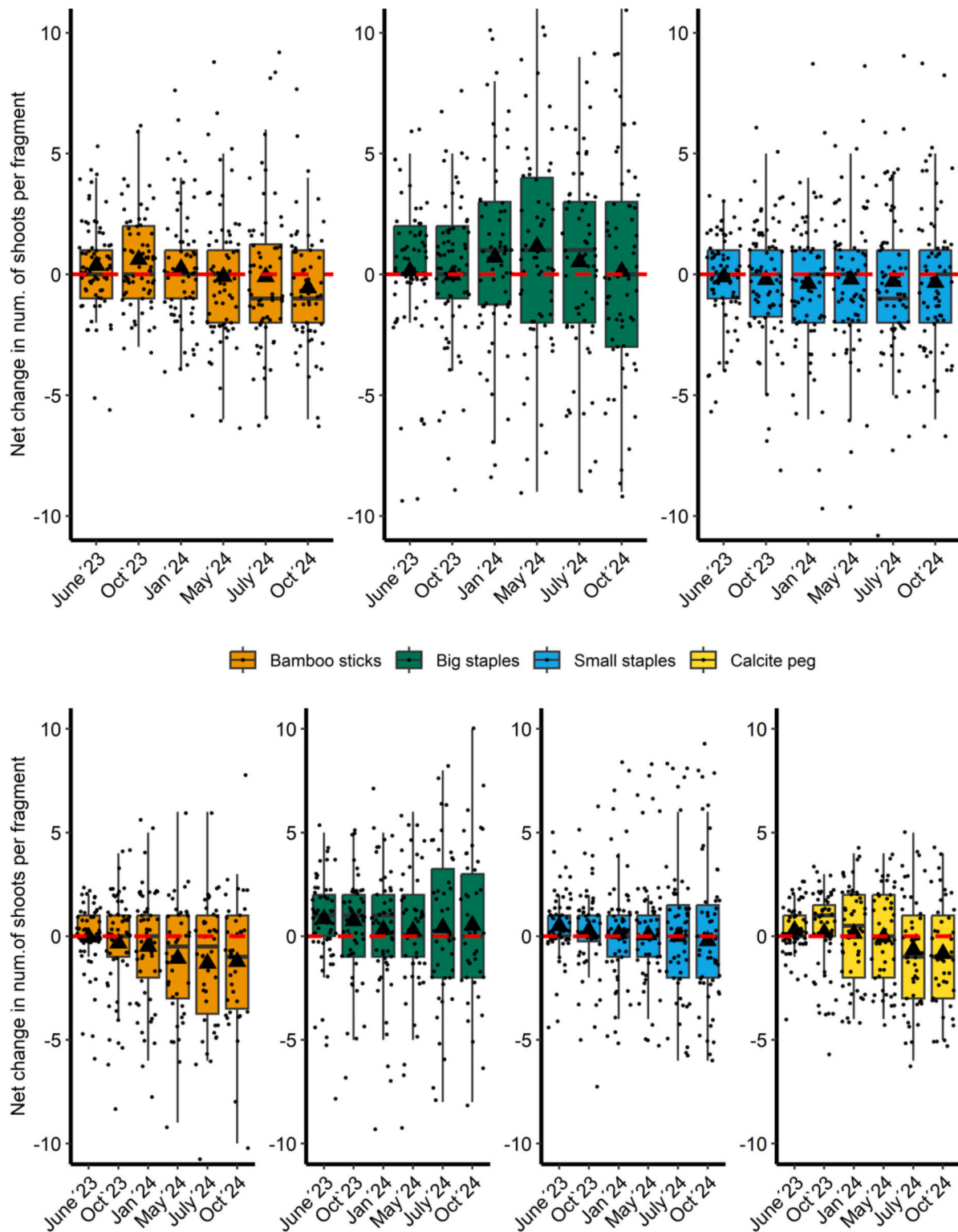
The initial population of uprooted fragments available for planting displayed considerable variability in the number of shoots they harbored, but almost 60 % of them had less than 6 shoots. Importantly, the number of shoots per fragment differed significantly among anchoring methods, with the choice of anchoring technique influencing the number of shoots per fragment needed to efficiently attach the fragments to the anchoring devices. For instance, the big staple method required fragments with higher number of shoots (mean  $6.3 \pm 2.7$ ), whereas the other methods allowed to use fragments that had lower number of shoots, which were the most common readily fragments available. Thus, the latter methods may offer advantages in terms of

constraints, cost-effectiveness and replicability when considering the pool of uprooted fragments available.

Overall, the establishment success of fragments varied through time, being significantly lower at later monitoring times, indicating potential challenges in maintaining planted fragments over time. Nevertheless, overall success after two years of planting was reasonably high (more than 70 % at both sites, mean of ca. 80 %), as it has also been observed in other *P. oceanica* transplanting experiments conducted on dead matte (76 % after 3 years; Piazzini et al., 1998). Although, it is during the first two years of planting when the biggest drop in survival tends to occur, with mortality slowing down after that (Mancini et al., 2022; Pergent-Martini et al., 2024), we may expect further decreases (albeit less pronounced) in establishment success of our transplants in the forthcoming years.

Importantly, the anchoring methods used did differ in establishment success, with small staples providing the highest (ca 95 %) establishment success at both sites, being higher than other methods used here and elsewhere for *P. oceanica*. For example, the use of big staples on uprooted fragments yield only between 0 and 30 % of establishment success after four years of planting (Castejón-Silvo and Terrados, 2021), but these assays were conducted in deeper areas (15-25 m) and on sand and burlap bags with gravel rather than on matte, which may have further decrease their success. In fact, a recent review found that restoration efforts in *P. oceanica* result in higher success (70 % of studies) in shallower depths (less than 10 m), when conducted on dead matte (80 %) as well as when using individual anchoring techniques (78 % rather than modular devices or degradable carpets (Pansini et al., 2022). However, the use of small staples on matte has not been as successful as in our study (59 % after four years; Pergent-Martini et al., 2024), further highlighting the need to monitor success at longer time scales. In fact, even after a decade some traits of restored *P. oceanica* meadows (e.g., phenological and lepidochronological) still fail to converge with those of the natural meadows (Bacci et al., 2025).

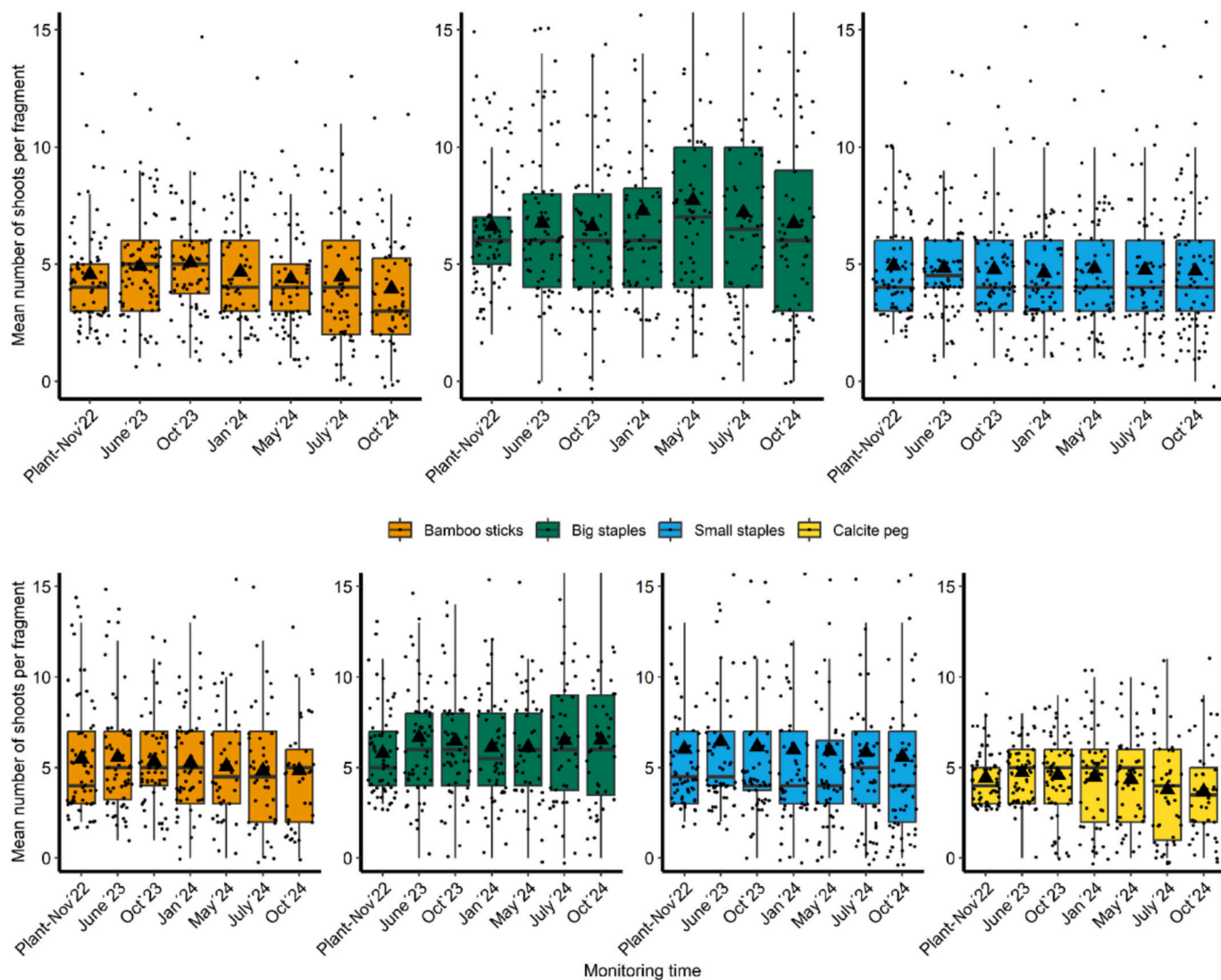
In addition to establishment success, however, it is also important to consider how the established fragments develop, as this is key for the long-term expansion and recovery of the meadow. Overall, and regardless of method used or site, a very small percentage of fragments (ca.3 %) were inactive, neither losing nor developing new shoots. In contrast, half of the planted fragments had lost shoots after 2 years of planting while one third of the fragments were developing new shoots.



**Fig. 5.** Net change in number of shoots per fragment in Formentor (top) and Portocolom (bottom) sites for the different anchoring methods. Small black dots show individual values, triangles represent mean value, horizontal line represents median value and vertical lines show the smallest and largest values within 1.5 times the interquartile range ( $1.5 \times IQR$ ) from Q1 and Q3. Dashed red lines mark 0 change. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

The only exception regarding effects of anchoring methods was detected in Portocolom when using big staples, with significantly higher percentage (52 %) of fragments having more shoots after 2 years than having less shoots (39 %). Research on the restoration of *Posidonia australis* using naturally detached fragments reported that

approximately one third (36.3 %) of the surviving fragments produced new shoots 5–6 months after planting (Ferretto et al., 2021). This suggests a recovery period of ca. 6 months from collection and transplantation stress during the year following planting for this species. In contrast, in our study, the dynamics observed on shoot gain and losses



**Fig. 6.** Mean number of shoots per fragment in Formentor (top) and Portocolom (bottom) using the different anchoring methodologies. Small black dots illustrate individual values, triangles represent mean values, horizontal lines represent median values, and vertical lines show the smallest and largest values within 1.5 times the interquartile range ( $1.5 \times IQR$ ) from Q1 and Q3.

**Table 3**

Non-ecological parameters of feasibility (being 2 the most feasible). Total feasibility score was calculated as the sum of individual scores. Explanation of feasibility parameters can be found in Table S1.

	Weight	Buoyancy	Steps prior to plant	Material loss	Supply availability	Supply time	Total cost	C footprint	Community	Total feasibility score
Big staple	0	2	0	0	1	1	0	0	1	5
Small staple	1	2	1	2	2	2	1	1	0	12
Bamboo sticks	2	0	2	1	2	2	2	2	0	13
Calcite peg	1	2	2	0	0	1	0	1	2	9

were slower. For instance, after 8 months, most fragments maintained the same number of shoots except for those anchored with the big staples method in Portocolom which were close to gaining one shoot ( $0.8 \pm 3.2$ ). Furthermore, it is only after 19 months that we detected a mean net shoot gain ( $1.5 \pm 4.5$  shoots per fragment) which was restricted to one anchoring method (big staples) at one site (Formentor). However, the overall net change of number of shoots per fragment remained close to zero across all methods, even after two years of planting, reflecting the slower growth rate of *P. oceanica* (Marbà and Duarte, 1998) compared to *P. australis* (Marbà and Walker, 1999). Thus, while shoot survival may

still decline over time, new shoots may not fully compensate for mortality, indicating that a two-year study period may be insufficient to assess long term restoration success in *P. oceanica*. Importantly, *P. australis* fragments with more shoots exhibited higher survival after one year (Ferretto et al., 2023) and also global analysis of seagrass restoration show that larger-scale plantings, involving a higher number of shoots or seeds, tend to have higher survival rates and positive population growth (van Katwijk et al., 2016). This suggests that planting larger fragments, when available, can enhance restoration success.

Overall, the results of our study followed similar trends between the

two study sites, Formentor and Portocolom. This consistency suggests that the outcomes of the anchoring methods used are not strongly site-specific and may be generalizable to similar shallow, sheltered areas with a history of *Posidonia oceanica* presence. These findings further support the broader applicability of the tested restoration methods and reinforce their potential effectiveness in comparable Mediterranean environments where anthropogenic pressures have been mitigated.

While establishment success and fragment development are critical ecological indicators for identifying best restoration methods from the perspective of plant health and meadow recovery, this study introduces a novel dimension by integrating non-ecological indicators which are crucial in helping decision-making. These complementary indicators, including cost, logistics or carbon footprint, have been largely overlooked in seagrass restoration but they play a key role in determining the overall feasibility of restoration actions. Indeed, seagrass restoration can be financially demanding, with a median cost of over 100k USD/ha, which is significantly higher than those associated with terrestrial ecosystem restoration (Bayraktarov et al., 2016). Thus, considering integrated approaches that take into account ecological and non-ecological factors will be particularly important for large-scale projects, where the success of the initiative depends not only on ecological outcomes but also on operational efficiency and alignment with broader socio-environmental goals.

In this context, we found that small staples may be an optimal option to use, as not only does it exhibit the highest establishment success, but also is highly feasible, particularly in terms of low monetary costs, low material loss and weight. Similarly, bamboo sticks exhibit a good feasibility score having the lowest weight, monetary cost, C footprint and easier supply. In contrast, large staples provide less versatility in terms of the size of fragments used (they are restricted to larger fragments, which are not the most commonly found), and are also more expensive, need a more complex setup with more difficult logistics (high weight per unit) and have a higher C footprint. However, since larger fragments, such as those used in big staples, tend to have a more successful development, this method may be particularly useful when there is an abundance of uprooted large fragments or in combination with other methods that target a broader range of fragment sizes.

## 5. Conclusion

In this study, we have identified key factors that should be considered to ensure the sustainability and effectiveness of restoration methods in *Posidonia oceanica*, introducing a novel integrative approach that balances both ecological outcomes and logistical feasibility to optimize the selection of anchoring methods. The consistent results across our sites suggest that the methodologies tested can be broadly applied to similar Mediterranean environments, particularly in shallow, sheltered areas with dead matte where *P. oceanica* previously existed. However, future studies should aim to test restoration methods across a wider range of environmental conditions. In fact, the techniques tested in this study are cost-effective in shallow waters, while other techniques that reduce underwater working time, such as mats, might be more convenient in deeper sites (De Luca et al., 2025).

We have identified two methodologies, small garden staples and bamboo sticks, which offer significant advantages in terms of cost-effectiveness, low carbon footprint, and logistical ease, involving also smaller fragments with fewer shoots which are more commonly found in the wild, and which have high establishment success and acceptable fragment development. While larger fragments tend to enhance fragment development they are also less commonly found, and the use of big staples involves lower feasibility due to more complicated logistics, higher costs and C footprint. Also, our study emphasizes that long-term monitoring remains critical to understanding fragment establishment and shoot development dynamics, particularly as shoot loss was observed over time and development of new shoots did not compensate for mortality. Ultimately, our findings emphasize the need for a

balanced approach that integrates ecological success with practical feasibility. This integrated approach is particularly important for large-scale projects, where the success of the initiative depends not only on ecological outcomes but also on operational efficiency and alignment with broader socio-environmental goals. Incorporating these considerations into a decision-making matrix ensures that restoration methodologies are both effective and sustainable in the long-term paving the way for long-term restoration and resilience of *Posidonia oceanica* meadows in the Mediterranean region.

## CRedit authorship contribution statement

**Gema Hernán:** Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Formal analysis, Data curation. **Laura Royo:** Writing – review & editing, Project administration, Methodology, Investigation, Data curation, Conceptualization. **Tatí Benjumea:** Methodology, Investigation, Conceptualization. **José Escaña:** Project administration, Conceptualization. **Fiona Tomas:** Writing – review & editing, Project administration, Methodology, Investigation, Conceptualization.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.marpolbul.2025.118746>.

## Data availability

Data is privately available at figshare and will be made public upon acceptance and publication at DOI 10.6084/m9.figshare.28574180

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