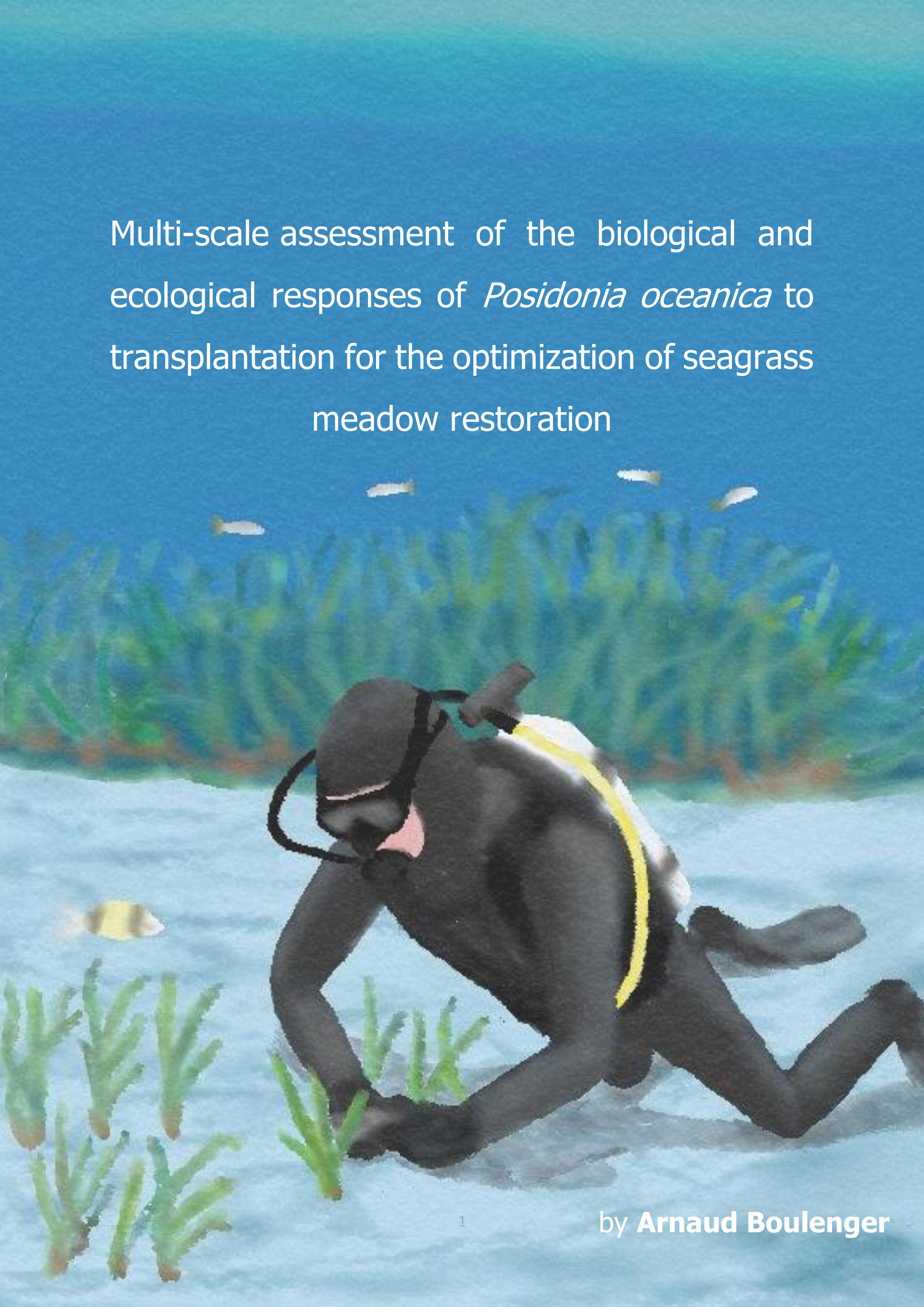
Multi-scale assessment of the biological and ecological responses of *Posidonia oceanica* to transplantation for the optimization of seagrass meadow restoration



by **Arnaud Boulenger**

Multi-scale assessment of the biological and ecological responses of *Posidonia oceanica* to transplantation for the optimization of seagrass meadow restoration

by **Arnaud Boulenger**

A thesis submitted in conformity with the requirements for the degree of *Philosophiæ doctor*
(Faculty of Sciences, Doctoral College in Oceanography)

Academic year 2025 – 2026

University of Liege
Faculty of Sciences
Department of Biology, Ecology and Evolution - FOCUS
Laboratory of Biological Oceanography

Supervisors:
Prof. Sylvie Gobert, supervisor (Université de Liège, Liege, Belgium)
Prof. Eric Goberville, co-supervisor (Sorbonne Université, Paris, France)

Thesis jury:
Prof. Aida Alvera Azcarate, president (Université de Liège, Liege, Belgium)
Prof. Stéphane Roberty, secretary (Université de Liège, Liege, Belgium)
Prof. Monica Montefalcone (Università degli Studi di Genova, Genoa, Italy)
Dr. Michel Marengo (STARESO, Calvi, France)
Dr. Julie Deter (Université de Montpellier, Montpellier, France)
Dr. Magali Cannac-Padovani (Office de l'Environnement de la Corse, Corte, France)
Prof. Sylvie Gobert, supervisor (Université de Liège, Liege, Belgium)
Prof. Eric Goberville, co-supervisor (Sorbonne Université, Paris, France)

AVANT-PROPOS

Cette thèse a été réalisée d'octobre 2021 à octobre 2025 au laboratoire d'Océanographie Biologique (FOCUS, ULiège). Les travaux de recherche ont été supportés financièrement par l'Université de Liège et le Fonds National de la Recherche Scientifique – FNRS. La Station de Recherche Océanographiques et sous-marines (STARESO) a soutenu financièrement et logistiquement ces travaux de thèse grâce au projet STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) financé par la Collectivité de Corse et l'Agence de l'eau Rhône-Méditerranée-Corse.

Les travaux présentés dans cette thèse ont été encadrés par Pr. Sylvie Gobert (promotrice ; ULiège) et Pr. Eric Goberville (co-promoteur ; Sorbonne Université) et a bénéficié des conseils avisés des membres du comité de thèse Dr. Michel Marengo, Dr. Stéphane Roberty, Dr. Julie Deter et Pr. Gérard Pergent.

REMERCIEMENTS

La réalisation d'une thèse de doctorat est connue pour être un travail solitaire et polyvalent, entre gestion de projet, recherche de financement, analyse de données, rédaction et vulgarisation. Pourtant, la réalisation d'un tel travail ne serait pas possible sans l'ensemble des personnes qui ont apporté leur pierre à cet édifice scientifique. J'espère que ces remerciements arriveront à valoriser du mieux possible l'incroyable travail que vous avez chacun.e réalisé et qui a permis l'aboutissement de cette thèse.

Le premier remerciement vient tout naturellement à ma promotrice, Sylvie Gobert, sans qui cette thèse n'aurait jamais eu lieu. Merci d'avoir répondu positivement à ma demande de thèse et d'avoir fait confiance au parfait inconnu que j'étais. Merci de m'avoir transmis tes connaissances et ta passion pour le poumon de la Méditerranée. Merci pour ta disponibilité, ta réactivité et ta manière d'aborder les problèmes qui fait qu'il n'y a que des solutions. Merci pour la liberté, l'autonomie et la créativité que tu m'as laissées pendant cette thèse, et qui ont contribué à faire de ces quatre années une expérience inoubliable. Merci pour toutes les opportunités que tu m'as offertes.

Merci à Eric Goberville d'avoir accepté d'être co-promoteur de cette thèse. Je suis sincèrement désolé de ne pas avoir pu faire aboutir aussi loin que je le souhaitais les travaux de modélisation que nous avons entamés ensemble pendant cette thèse, et j'espère que tu ne regrettes pas le temps passé à rendre fonctionnel mes scripts R. Tu m'avais laissé un souvenir très positif pendant les cours de modélisation de mon master, et je dois dire que je n'ai pas été déçu de choisir de réaliser cette thèse avec toi. Je n'ai jamais eu une grande affinité pour l'informatique, le codage, ou les statistiques, mais tu as réussi à me transmettre ta passion pour la modélisation. Merci de m'avoir accueilli au sein de ton laboratoire à Paris pour qu'on puisse travailler ensemble en présentiel sur la modélisation. J'ai adoré ce séjour. Merci pour ta disponibilité et ta flexibilité. J'espère que j'arriverai à valoriser le mieux possible les travaux que nous avons réalisé ensemble au cours de cette thèse, et que nous aurons encore l'occasion de travailler ensemble par la suite.

Merci aux membres de mon comité de thèse Sylvie Gobert, Eric Goberville, Stéphane Roberty, Michel Marengo, Gérard Pergent et Julie Deter pour nos réunions annuelles et nos discussions constructives pour améliorer ces travaux de thèse. Merci pour votre bienveillance, vos encouragements et vos félicitations qui m'ont aidé à atténuer mon syndrome de l'imposteur.

Merci aux membres de mon jury de thèse Sylvie Gobert, Eric Goberville, Aida Alvera Azcarate, Stéphane Roberty, Michel Marengo, Julie Deter, Magali Cannac-Padovani et Monica Montefalcone d'avoir accepté d'évaluer le résultat final de ces quatre années de thèse.

Merci au Fonds National de la Recherche Scientifique – FNRS, FOCUS, l'Université de Liège, l'agence de l'eau Rhône-Méditerranée-Corse et la Collectivité de Corse pour le soutien financier qui a permis la réalisation de ces quatre années de thèse. Merci à la Fondation de la Mer pour le financement du projet REPAIR 2.0, qui ouvre de belles perspectives venant compléter ces travaux de thèse.

Merci aux membres du laboratoire d'Océanographie biologique Sylvie Gobert, Cédric Delforge, Krishna Das, Laurence Lefebvre et Marianna Pinzone pour votre accueil au sein du labo dés mon arrivée en octobre 2021. Merci également à Françoise Bruls et Mariella Lunetta, pour votre travail si précieux qui nous facilite tellement la vie dans notre travail de tout les jours.

Merci à Cédric Delforge pour ta compagnie et ton aide (développement de méthodo, plongées, grattage, analyses ICP-MS, et j'en passe…) tout au long de ces quatre années. Tu es sans aucun doute le binôme avec qui j'ai passé le plus de temps à compter les faisceaux de posidonies. A défaut de pouvoir te décerner le titre d'employé du mois, je devrais te donner le titre de binôme de plongée pour les quatre années passées. Sincèrement, merci pour tout : les midis au labo, les apéros en Corse, les plongées,…

Merci à Stéphane Roberty pour m'avoir initié aux « joies » de la photosynthèse. J'ai eu du mal au début, mais avec le temps j'ai fini par ne plus diaboliser le PAM. Merci de m'avoir accueilli dans ton labo pour les dosages de pigments chlorophylliens, et pour tes retours très détaillés sur nos articles en commun.

Merci à Gilles Lepoint pour avoir pris le temps de discuter ensemble de mon design expérimental en début de thèse, et pour l'ensemble des dosages C-N-S. L'étuve était toujours pleine à rabord, mais tu t'es toujours arrangé pour que je puisse avoir rapidement mes résultats.

Merci à Annick Wilmotte pour m'avoir accueilli au sein de ton labo pour les manips sur les bactéries fixatrices d'azote au sein des racines de la posidonie. Merci également à Marcelo Gomes Marçal Vieira Vaz pour tout le temps que tu as accordé à Morgan et moi, et tes nombreux conseils pour l'optimisation de l'extraction d'ADN.

Merci à Gérard Bonifacio et Pierre Lejeune pour m'avoir permis de réaliser ces travaux de thèse à STARESO et votre confiance. Merci pour cette confiance que vous continuez de m'accorder au-delà de mes travaux de thèse pour l'année à venir. Merci Pierre pour tes conseils dans la conception de la thèse pendant les premiers mois, les diners chez toi à contempler la magnifique baie de Calvi, et les histoires inspirantes sur la biodiversité méditerranéenne. J'espère un jour être capable de raconter des histoires aussi bien que toi.

Merci à Michel Marengo pour ton accueil à bras ouverts, ta confiance, ton ouverture d'esprit et ton enthousiasme envers mes (trop) nombreuses idées de nouveaux projets à développer à la station. Ton ouverture à la créativité aura grandement contribué à mon épanouissement au sein de la station. De mon point de vue, au-delà de ton rôle de directeur scientifique, tu es la personne qui incarne le mieux les valeurs de STARESO qui me tiennent le plus à cœur : innovation, ambition, excellence scientifique, humanité et convivialité. Merci pour la grande liberté que tu m'as laissée au cours de ces quatre années, et pour la valorisation constante de mon travail et de mes compétences.

Merci à Quentin Fontaine et Lovina Fullgrabe d'avoir été là au cours de ces quatre années. Vous êtes, chacun à votre manière, deux icones emblématiques de la station qui font d'elle ce qu'elle est aujourd'hui. Merci pour votre rigueur et excellence scientifique, votre passion et votre dévouement envers notre station. Merci de m'avoir transmis votre amour de la Corse. Merci pour les belles plongées en baie d'Ajaccio et à la recherche des nids de picarels, j'ai hâte que vous ayez vos recycleurs pour qu'on puisse pousser ces explorations un peu plus loin. Merci Lovina pour tes témoignages débordant d'affection, et pour ton amitié.

Merci à Cyril Steibel pour ton accueil et de toujours avoir fait en sorte que je puisse être logé dans les meilleures conditions possibles à chacun de mes séjours à la station, malgré une logistique à s'en tirer les cheveux et mes dates de départ assez instables. Merci pour les séances de crossfit, les superbes concerts en ville et le son si envoutant du ukulélé les week-ends calmes à la station.

Merci à Sylvain Plaza pour les (très) nombreuses sécu surface, ton aide pour les manips sous l'eau, et ton apprentissage en plongée scientifique. J'ai commencé ma thèse en étant très jeune plongeur, et tu m'as vraiment permis de développer mes compétences en tant que plongeur, de surcroit comme plongeur scientifique. Merci pour tous tes conseils.

Merci à Hervé Arranz, l'« oncle » de STARESO. « La science, vous êtes des débiles profonds » mais heureusement tu restes là pour veiller au bon fonctionnement de la station. Merci pour ta constance à nous traiter de touristes, ton humour (même si parfois un peu limite) et pour toutes les anecdotes sur tes nombreuses vies antérieures.

Merci à Annick Donnay pour le suivi des demandes d'AOT nécessaires pour l'initiation des travaux de transplantation, et ton aide pendant ceux-ci.

Merci à l'équipe de choc 2022 Sébastien Cnudde, Nine Doutreloux, Dimitri Theuerkauff, Justine Castrec et Nils Mellet pour toutes les soirées en ville et à la station, Calvi on the rock, les plongées, les randos en montagne,… Mes trois mois passés à transplanter des boutures de posidonie auraient été bien moins agréable sans vous.

Merci à Aubin Woehrel, arrivé presque en même temps que moi à la station. Merci d'avoir été mon coloc pendant ma première longue campagne de terrain à la station. Trois ans et demi de chemin parcouru ensemble, entre les plongées du bord et les nombreuses soirées partagées à la station. Merci pour ton enthousiasme, ta passion, et ton humour si unique. Rappelle-moi que je dois te passer un savon.

Merci à Noémie Chabrier pour les discussions profondes après les soirées un peu trop alcoolisées et tes longues tirades sur mes « nombreuses qualités ». Je n'ai jamais su recevoir des compliments, mais ça m'a tout de même touché à chaque fois. J'espère que tu auras l'occasion d'un jour plonger à 100m comme tu en rêvais, c'est vraiment magnifique là-dessous.

Merci à Charlotte Defortescu, ma collègue du stud, pour les nombreuses soirées et week-ends passés ensemble. De manière générale, merci pour ton amitié. A jeudi !

Merci à Lou Balsaux pour notre passion partagée des gorgones, et le super week-end dans la région de Porto Vecchio. Le stud n'a plus jamais été pareil sans Charlotte et toi. J'espère que j'aurai l'occasion de reprendre les recherches que tu avais initiées pendant ton stage, et qu'on aura l'occasion de se revoir à mon prochain retour en Corse.

Merci à notre babelutte de la station, Lucie Borrossi. Pikachu t'est définitivement associé depuis que je te connais.

Merci à Julie Séveno, ma fraté de verre. Merci pour ta bienveillance, ta générosité, ton altruisme et ton humour. On s'est rencontré alors que tu finissais ta thèse, et à présent c'est à toi de me voir finir la mienne. Je dois dire que je me suis un peu assagi au cours des années et que je n'ai pas été très présent comme fraté de verre au cours de mes séjours en 2025. Ca n'est que partie remise pour 2026, en apéro après-plongée en recycleur.

Merci à Mélodie Chapat, qui s'efforce de gérer au mieux tout le matériel de la station. Merci pour ton humour, ton rire communicatif, ton énergie de boute-en-train et ton amour des restaurants (et des bars). Merci d'avoir fait de Kiki la mascotte de la station, malgré son caractère de princesse.

Merci à Xavia Martelli, sauveuse de poulpes et dompteuse de murènes, pour ton humour et ton grand sourire contagieux que tu affiches chaque jour. J'attends toujours avec impatience de voir ton spectacle avec Michel.

Merci à Margaux Gouazé pour le printemps 2023 qu'on a passé ensemble. Mes retours à la station n'ont plus jamais été les mêmes après toi. Tous ces moments ensemble en font sans aucun doute la campagne de terrain dont je garde les meilleurs souvenirs. *Abbracciu*.

Merci à Titouan Seimandi pour ta bonne humeur, ton humour, tes chants corses, italiens et québécois. Merci de m'avoir fait découvrir *A ghitarra* et *L'italiano*. Chaque fois que je les écoute, je me souviens de toi en train de les chanter.

Merci à Jonathan Richir, mon collègue de la salle de réunion. Merci cher compatriote pour avoir apporté un peu de Belgique dans mon quotidien corse. Merci pour tes anecdotes de scaphandrier et pour les perspectives stimulantes de travaux futurs dans la zone mésophotique. J'espère que les projets que j'ai en tête verront le jour et que nous pourrons aller explorer les profondeurs des canyons corses.

Merci à Giulio Cavalieri pour ta polyvalence et la séance de grimpe en falaises dans la vallée de la Restonica. J'espère qu'on aura l'occasion d'en refaire et que je serai un peu meilleur d'ici là !

Merci à nos capitaines Espartaco Orticoni, dont la relève a ensuite été prise par David Taupin, pour veiller sur le STARESO. J'admire votre détermination et entêtement devant la résolution des multiples soucis techniques du STARESO.

Merci à Fanny Jonnon et Céline Leroch de veiller à la propreté de la station. Merci Fanny pour ta bienveillance, ton rire communicatif, et d'avoir toujours fait en sorte que ma chambre soit dans un état impeccable à chaque retour. La petite tortue que tu m'y as laissé continuera de veiller sur elle.

Merci aux cuisinières et cuisiniers Géraldine Faravel, Joelle Boccheciampe et Robin Goldbronn pour vos bons petits plats, vous m'avez régalé.

Merci à tous les stagiaires Aubin Woehrel, Marion Blanchard, Marina Dumas, Margaux Gouazé, Titouan Seimandi, Alice de Saint Jacob, Julie Garcia, Marine Bitaly , Sara Epaminondas, Lisa Laviolette et Marti Poch avec qui j'ai pu partager des moments de vie commune sur la station. Sans oublier la multitude d'animaux de compagnie en tout genre (chiens, chats, renards, goéland, cormoran,…) qui sont venus égayer mon quotidien au cours de ces 4 dernières années.

Un merci général à l'ensemble des personnes citées et celles que j'aurais éventuellement oubliés. Vous avez toutes et tous contribuer à faire de STARESO ma deuxième maison, me laissant un pincement au cœur à chaque retour sur le continent. *Vi ringraziu è à prestu* !

Merci à l'ensemble des stagiaires et mémorant.e.s qui ont contribué directement à ces travaux de thèse. Ce travail n'aurait pas pu voir le jour sans vous, et j'espère qu'il retranscrit au mieux tous les efforts que vous avez pu mettre lors de vos stages et mémoires.

Merci aux étudiant.e.s du master MER Argelia Bustillo, Uxue Moreda, Iñigo Uriarte, Héctor Ibáñez, Maria Margarita Lopez Velosa, Husna Sheik, Maria Gabriela Aponte et Ana Lucía Azul

Acuña d'avoir consacré quelques semaines de votre été à venir construire des pièges à sédiment, compter des bateaux ou faire des dosages de nutriments.

Merci à Chiara Ruello pour m'avoir fait découvrir le rôle d'encadrant de mémoire, alors que je venais de commencer ma thèse. J'espère que j'ai réussi à t'encadrer du mieux que j'ai pu et que tu gardes une bonne expérience de ton passage à Liège.

Merci à Maria Margarita Lopez Velosa pour avoir déchiffrée les mystères des données PAM. Je dois bien avouer que je procrastinais ce travail depuis un moment, et ton mémoire a permis de nettement me faciliter la vie.

Merci à Morgan Frémal d'avoir accepté le challenge d'étudier les bactéries fixatrices d'azote au sein des racines de la posidonie, même si ça n'a pas abouti malgré les nombreuses semaines passées à essayer d'optimiser l'extraction d'ADN. Merci d'avoir apporté un peu de Bruxelles à Liège, et pour ton aide précieuse à STARESO pendant les campagnes d'échantillonnage.

Merci à Juliana Chapeyroux pour t'être attaquée à l'analyse des orthophotos et d'avoir mis au point le protocole méthodologique mettant en lumière les différentes dynamiques de recolonisation naturelle. La réalisation du Chapitre 2.1 a été possible en grande partie grâce à toi.

Merci à Ana Lucía Azul Acuña pour les nombreux dosages qui m'ont permis de sauver un temps précieux, et qui constituent une part essentielle du Chapitre 3.2. J'espère que ton FRIA sera accepté et que tu pourras reprendre les portes que j'ai ouverte sur le rôle du microbiome dans la réussite de la transplantation de la posidonie.

Merci à Lilly Bucher pour ta détermination à réaliser un mémoire de qualité. Malgré les nombreuses difficultés rencontrées, tu n'as jamais baissé les bras et tu es arrivée aux objectifs que nous avions fixé ensemble. Bien que tu n'en sois pas convaincue, ton travail constitue une partie essentielle des futures perspectives de cette thèse. Les herbiers de posidonies corses te remercient !

Je remercie en particulier mes nombreux binômes de plongée de STARESO et ULiège déjà cités ci-dessus pour m'avoir assisté dans les nombreuses heures passées sous l'eau (166 plongées !). Ces travaux de doctorat n'auraient pas été possibles sans vous toutes et tous. Merci à vous de n'avoir jamais rechigné à venir compter des faisceaux de posidonies avec moi, dans de l'eau pas toujours très chaude…

Merci à Aschwin Engelen et Gerard Muyzer de m'avoir fait confiance et d'avoir accepté de collaborer avec moi pour percer les mystères du microbiome racinaire de la posidonie. Les résultats qui ressortent de ce travail commun est probablement la partie la plus innovante de ma thèse, et je vous remercie pour ça. Merci Aschwin pour ton accueil au sein de ton laboratoire à Faro, ta gentillesse, ta disponibilité et ta sympathie. Tu as réussi à me transmettre ta passion pour le microbiome, et je te remercie de m'avoir permis de découvrir ce monde microscopique mais si important. J'ai adoré mon séjour à Faro et j'espère qu'on aura encore de nombreuses occasions de travailler ensemble.

Merci à Tania Aires pour m'avoir encadré et aidé pour l'ensemble de mes manipulations à Faro, et les analyses bio-informatiques à distance. Merci pour ton aide précieuse, ta patience et ta gentillesse.

Merci à Julia van Duijnhoven et Joana De Lemos Filipe pour les moments passés ensemble à CCMAR. Mon séjour n'aurait pas été aussi agréable sans votre accueil chaleureux et votre sympathie.

Merci à Arnaud Abadie pour tes conseils sur le choix des appareils photo pour la photogrammétrie, et sur la photogrammétrie sous-marine en elle-même.

Merci à Sophie Klenkenberg pour les précieux conseils en biostatistique pendant mes premières années de thèse, alors que je me tirais les cheveux pour analyser correctement mes données.

Merci à Patrick Panneels et Sophie Fally pour m'avoir transmis leur amour de la plongée et du milieu marin, alors que je n'y connaissais encore rien. Je ne serais probablement pas là où je suis si je n'avais pas atterri au GBRS au cours de ma troisième année de bachelier.

Merci à la team des plongeurs de l'extrême Martin Dogniez, Killian Bouillon, Lucas Terrana et Gregory Dumont pour toutes ces plongées. Nos carrières belges sont peut-être froides et sombres, y plonger tous les dimanches m'a vraiment permis de recharger mes batteries.

Merci à Ronan Strebler, Thomas Pieters, Guillaume Van Oye, Simon Lefevere Alifierakis, Martin Veinstein, Olivier Fontaine, Victor Janssens, Milan Lobo, Pierre Herinckx, Guillaume Pé, Margaux Vanhussel, Eliza Sack, Reine Cernero, Eline Decaestecker, Philippine Moret et Lea Katz pour votre amitié sans faille et votre soutien tout au long de ces années. Merci pour tous ces bons moments passés ensemble et d'avoir toujours été là, malgré mes absences prolongées.

Les meilleurs pour la fin, je remercie du fond du cœur ma famille : Papa, Maman, Maxime,… Merci pour tout votre amour, votre soutien inconditionnel, et vos encouragements sans faille envers tous mes projets. Ces travaux de thèse n'auraient jamais vu le jour sans vous.

Enfin, un remerciement plein d'amour pour toi, Margaux, mon amoureuse et ma meilleure amie. Merci de m'avoir soutenu dans les moments les plus difficiles, entre stress et perte de sens. Merci d'avoir toujours pu compter sur toi, malgré mon caractère pas toujours facile…

Arnaud

RÉSUMÉ

La restauration des écosystèmes côtiers est désormais considérée comme une priorité afin de soutenir le rétablissement de leurs services écosystémiques. En Méditerranée, de nombreux projets ont été menés au cours des dernières décennies pour tenter de restaurer les herbiers endémiques de *Posidonia oceanica* (L.) Delile. Cependant, d'importantes lacunes scientifiques persistent concernant la transplantation de cette espèce, notamment concernant les performances de différentes méthodes de fixation des transplants et de différentes origines de boutures. Cette thèse avait pour objectif d'évaluer l'efficacité relative de trois méthodes de transplantation utilisant des matériaux biodégradables et de déterminer la performance de deux origines de boutures : des boutures naturellement décrochées de l'herbier dérivantes sur le fond (i.e., boutures-épaves) et des boutures prélevées au sein d'herbiers donneurs. Pour répondre à ces objectifs, un suivi sur trois ans a été mené en baie de Calvi (Corse, France), où 693 boutures ont été transplantées sur de la matte morte en testant trois méthodes de transplantation : individuelle (agrafe métallique), tridimensionnelle souple (géotextile en fibres de noix de coco) et tridimensionnelle rigide (élément BESE). La performance de ces méthodes de transplantation et origines de boutures a été évaluée sur base d'un suivi du taux de survie, du rapport coût-bénéfice, mais également de la dynamique morphologique, microbiologique, physiologique et biochimique des transplants et des herbiers de référence.

Les méthodes de fixation individuelles présentent un très bon rapport coût-bénéfice mais sont adaptées uniquement dans des zones à faible hydrodynamisme et sur de la matte morte peu altérée physiquement. Les structures tri-dimensionnelles rigides sont coûteuses mais permettent d'obtenir de bons taux de survie dans des zones à hydrodynamisme important. Les méthodes qui permettent un contact direct entre les transplants et la matte morte favorisent le développement du système racinaire et l'établissement d'une structure des communautés bactériennes similaire à celle des herbiers naturels. Les boutures-épaves présentent des performances similaires aux boutures issues de l'herbier en termes de taux de survie et de morphologie foliaire et racinaire. Cependant, les boutures prélevées dans l'herbier ont une structure du microbiome racinaire, ainsi que des traits physiologiques et biochimiques plus similaires aux herbiers naturels que les boutures-épaves deux ans après transplantation. Les différences physiologiques et biochimiques s'estompent deux ans après la transplantation. Ces résultats suggèrent une meilleure performance des boutures issues de l'herbier durant les deux premières années qui suivent la transplantation. Ces travaux de thèse apportent de nombreux éléments de réponse à des axes majeurs de recherche et développement concernant la transplantation des herbiers de *P. oceanica*.

ABSTRACT

The restoration of coastal ecosystems is now considered a priority to support the recovery of their ecosystem services. In the Mediterranean Sea, numerous projects have been carried out over the past decades to restore the endemic *Posidonia oceanica* (L.) Delile meadows. However, major knowledge gaps remain regarding the transplantation of this species, particularly with respect to the performance of different anchoring techniques and donor sources. This thesis aimed to assess the relative efficiency of three transplantation methods using biodegradable materials and to evaluate the performance of two donor sources: naturally detached fragments drifting on the seafloor (i.e., storm-fragments) and cuttings harvested from donor meadows. To address these objectives, a three-year monitoring program was conducted in Calvi Bay (Corsica, France), where 693 cuttings were transplanted onto dead matte using three transplantation techniques: individual fixation (iron staples), soft three-dimensional structures (coconut fiber mats), and rigid three-dimensional structures (BESE elements). The performance of these transplantation methods and donor sources was evaluated based on survival rate, cost–benefit ratio, and the morphological, microbiological, physiological, and biochemical dynamics of transplants compared with reference meadows.

Individual fixation methods showed an excellent cost–benefit ratio but were suitable only in low-hydrodynamic areas with minimally degraded dead matte. Rigid three-dimensional structures were more expensive but achieved high survival rates in high-hydrodynamic environments. Methods allowing direct contact between transplants and the matte promoted root system development and the establishment of bacterial communities resembling those of natural meadows. Storm-fragments performed similarly to donor cuttings in terms of survival, leaf and root morphology. However, donor cuttings displayed root microbiome structure as well as physiological and biochemical traits more similar to natural meadows than storm-fragments two years after transplantation. Physiological and biochemical differences between the two donor sources diminished after two years, suggesting that donor cuttings outperform storm-fragments during the first two years following transplantation. This thesis provides key insights into major research and development issues related to the transplantation of *P. oceanica* meadows.

Articles scientifiques publiés et soumis dans des revues

Publications reprises dans la thèse

Boulenger, A., Roberty, S., Lopez Velosa, M. M., Marengo, M., & Gobert, S. (2024). The Use of Photo-Biological Parameters to Assess the Establishment Success of *Posidonia oceanica* Cuttings after Transplantation. *Water, 16(12)*, 1702. https://doi.org/10.3390/w16121702

Boulenger, A., Chapeyroux, J., Fullgrabe, L., Marengo, M., & Gobert, S. (2025). Assessing *Posidonia oceanica* recolonisation dynamics for effective restoration designs in degraded anchoring sites. *Marine Pollution Bulletin*, 216, 117960. https://doi.org/10.1016/j.marpolbul.2025.117960

Boulenger, A., Aires, T., Engelen, A.H., Muyzer, G., Marengo, M., & Gobert, S. (2025). Microbiome matters: how transplantation methods and donor origins shape the successful restoration of the seagrass *Posidonia oceanica*. *Environmental Microbiome*, 20(99). https://doi.org/10.1186/s40793-025-00764-9

Boulenger, A., Marengo, M., Boissery, P., & Gobert, S. (2025). Comparative assessment of transplantation methods and donor sources for the restoration of *Posidonia oceanica* meadows. *Science of the Total Environment*, 1000, 180488. https://doi.org/10.1016/j.scitotenv.2025.180488

Boulenger, A., Acuna, A.L.A., Roberty, S., Lepoint, G., Marengo, M., & Gobert, S. (2026). Physiological and biochemical performance indicators of the seagrass *Posidonia oceanica* restoration success. *Marine Pollution Bulletin*, 223, 119036. https://doi.org/10.1016/j.marpolbul.2025.119036

Autres publications rédigées pendant la thèse

Boulenger, A.*, Lanza-Arroyo, P.*, Langedock, K., Semeraro, A. & Van Hoey, G. (2024). Nature-based solutions for coastal protection in sheltered and exposed coastal waters: integrated monitoring program for baseline ecological structure and functioning assessment. *Environmental Monitoring and Assessment, 196,* 316. https://doi.org/10.1007/s10661-024-12480-x
*Ces auteurs ont contribué de manière égale.

Descamp, P., Personnic, S., Gobert, S., **Boulenger, A.**, Leduc, M., Delaruelle, G., Barroil, A., Marre, G., Holon, F., & Deter, J. (2025). Seagrass sod transplantation: A relevant tool for preventing the destruction of meadows in coastal construction projects. *Environmental Challenges*, 101087. https://doi.org/10.1016/j.envc.2025.101087

Astruch, P., Bensoussan, N., André, S., Boudouresque, C. F., Tomas, F., Teixidó, N., … , **Boulenger, A.**, … , Montefalcone, M. Mass flowering of the seagrass *Posidonia oceanica* after 2022 record-breaking marine heatwaves: a Pan-Mediterranean study. *Communications Earth & Environment*. Submitted.

Rapports de recherches

Fullgrabe, L., Fontaine, Q., Marengo, M., Donnay, A., Sirjacobs, D., Iborra, L., Cnudde, S., Mellet, N., Seveno, J., **Boulenger, A.**, Lefebvre, L., Binard, M., Katz, L., Abadie, A., Boissery, P., Gobert, S., & Lejeune, P. (2022). STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) – Année 2021. Rapport de recherches, STARESO, 123 p.

Fullgrabe, L., Fontaine, Q., Richir, J., Martelli, X., **Boulenger, A.**, Borrossi, L., Woerhel, A., Balsaux, L., Iborra, L., Seimandi, T., Cnudde, S., Leduc, M., Dumas, M., Seveno, J., Chapat, M., Donnay, A., Boissery, P., Gobert, S., Lejeune, P. & Marengo, M. (2024). STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) – Année 2022-2023. Rapport de recherches, STARESO, 133 p.

Martelli, X., Fullgrabe , L., Fontaine, Q., Richir, J., Woehrel, A., Bitaly, M., Borrossi, L., **Boulenger, A.**, Seveno, J., Chapat, M., Siedel, L., Garcoa, J., Gobert, S., Lejeune, P., & Marengo, M. (2025). STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) – Année 2024. Rapport de recherches, STARESO, 134 p.

Posters et communications orales dans des congrès

Boulenger, A., Marengo, M., Fontaine, Q., Lejeune, P. & Gobert, S. (2022). Projet REPAIR: Restauration des patchs anthropiques dans l'herbier de posidonie. Communication orale à la Présentation des principaux travaux en faveur des herbiers de posidonie. Agence de l'Eau Rhone-Méditerranée-Corse, en ligne, 6 janvier 2022.

Boulenger, A., Marengo, M., Fontaine, Q., Lejeune, P. & Gobert, S. (2022). Restoration of anthropogenic patches in *Posidonia oceanica* meadows. Communication orale au Global Seagrass Nursery Network kick-off seminar, en ligne, 31 mars 2022.

Fullgrabe, L., Marengo, M., Fontaine, Q., Donnay, A., Cnudde, S., **Boulenger, A.**, Gobert, S., Boissery, P. & Lejeune, P. (2022) STARECAPMED : STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts.. Communication orale au 5e Journées Internationales de Limnologie et d'Océanographie, Corte (France), 10 octobre 2022.

Boulenger, A., Marengo, M., Fontaine, Q., Fullgrabe, L., Lejeune, P., Boissery, P. & Gobert, S. (2022). Restauration des patchs anthropiques dans l'herbier de posidonie. Communication orale au 5e Journées Internationales de Limnologie et d'Océanographie, Corte (France), 12 octobre 2022.

Boulenger, A., Marengo, M., Fontaine, Q., Lejeune, P. & Gobert, S. (2022). Restauration des patchs anthropiques dans l'herbier de posidonie. Communication orale à l'Atelier DRIVER - Evaluer, améliorer et restaurer la qualité des fonds marins de Méditerranée, Montpellier (France), 27 octobre 2022.

Boulenger, A., Marengo, M., Fontaine, Q., Fullgrabe, L., Lejeune, P., Boissery, P. & Gobert, S. (2022). Restoration of anthropogenic patches in *Posidonia oceanica* meadows. Communication orale au FOCUS Early Career Scientists Day, Liège (Belgique), 13 décembre 2022.

Didderen, K., **Boulenger, A.**, van der Heide, T., Teunis, M., Gobert, S. & Lengkeek W. (2023). Restoration of seagrass meadows after mechanical damage - incorporating biomimicry and recovery of feedbacks in restoration techniques . Présentation poster à la 7th Conference on Wind energy and Wildlife impacts, Šibenik (Croatie), 18-22 septembre 2023.

Boulenger, A., Frémal, M., Vaz, M.G.M.V., Wilmotte, A., Lepoint, G. & Gobert, S. (2023). Diversity of endophytic nitrogen-fixing bacteria in the roots of *Posidonia oceanica*: Implications for restoration. Présentation poster au 11ème congrès de l'Association Francophone d'Ecologie Microbienne, Carqueiranne (France), 17-20 octobre 2023.

Astruch, P.,…**Boulenger, A.**,…., Montefalcone, M. (2024). Uneven flowering occurrences of *Posidonia oceanica* along the Mediterranean Sea: insights from the 2022 event. Présentation orale au 2024 World Seagrass Conference, Naples (Italie), 19 juin 2024.

Boulenger, A., Marengo, M., Lepoint, G., Didderen, K., Lengkeek, W., van der Heide, T. & Gobert, S. (2024). Assessment of different transplantation methods for *Posidonia oceanica* meadows restoration by means of morphological and physiological traits. Présentation orale au 2024 World Seagrass Conference, Naples (Italie), 19 juin 2024.

Boulenger, A., Marengo, M., Lepoint, G., Didderen, K., Lengkeek, W., & Gobert, S. (2024). Assessment of different transplantation methods and donor populations for *Posidonia oceanica* meadows restoration. Présentation orale au FOCUS Young Scientists Day, Liège (Belgique), 10 décembre 2024.

Boulenger, A., Aires, T., Engelen, A.H., Muyzer, G., Marengo, M., & Gobert, S. (2025). Micorbiome matters : How transplantation methods and donor origins shape the successful restoration of the seagrass *Posidonia oceanica*. Présentation poster au deuxième European Seagrass Restoration Workshop, Arcachon (France), 8-10 avril 2025.

Recherche de financements

Biodiversa Joint Research Call 2021 – Supporting the protection of biodiversity and ecosystems across land and sea [Non obtenu]
Titre: C4EC – Citizens for Effective Conservation: citizen science, eDNA, and AI to prioritize marine species and areas conservation.
Contribution: Participation à l'écriture du projet.

Bourse Barbault-Weber « Ecologie impliquée », 2022 [Non obtenu].
Titre : REPAIr - REstoration of anthropogenic PAtches in Posidonia oceanIca meadows
Contribution: Chef de projet, design et écriture du projet.

Crédit de recherche, F.R.S.-FNRS, 2022 [Obtenu].
Titre : UNSEAL - UNravelling SEAgrass transplantation Limits.
Budget : 53 000 €
Contribution : Participation au design et à l'écriture du projet, expertise technique.

Crédit facultaire de recherche UR FOCUS, ULiège, 2022 [Obtenu].
Titre : READ ME - REcord to Assess Diversified Marine Ecoystems: exploring fish acoustic similarities. Collaboration avec le doctorant Xavier Raick du Laboratoire de Morphologie Fonctionnelle et Evolutive (ULiège).
Budget : 8 000 €
Contribution : Participation au design et à l'écriture du projet.

Crédit pédagogique, ULiège, 2022 [Obtenu].
Acquisition d'un BlueROV 2 dans le cadre des cours du master en océanographie.
Budget : 10 000 €
Contribution : Participation à l'écriture du projet.

Projet de thèse, FRIA - F.R.S.-FNRS, 2023, [Non financé, thèse de Morgan Frémal].
Titre: PACcTe : Potentiel d'Acclimatation de *Cladocora caespitosa* face à l'augmentation de la Température des eaux de surface.
Contribution: Participation à l'écriture du projet.

Bourse Barbault-Weber « Ecologie impliquée », 2023 [Non obtenu].
Titre : UNSEAL - UNravelling SEAgrass transplantation Limits.
Contribution: Chef de projet, design et écriture du projet.

Projet de recherche, Fondation de la Mer, 2024 [Obtenu]
Titre : REPAIR 2.0 - Restauration des herbiers de posidonies endommagés par l'ancrage.
Budget : 38 000 €
Contribution : Chef de projet, design et écriture du projet.

Crédit facultaire de recherche UR FOCUS, ULiège, 2025 [Obtenu].
Titre : UNSEAL - UNravelling SEAgrass transplantation Limits.
Budget : 2 000 €
Contribution : Chef de projet, design et écriture du projet.

Projet de recherche, Fondation de la Mer, 2025 [Obtenu].
Titre : POSICARE - POsidonies : Sauvegarde, Innovation et Conservation par des Actions de Restauration Écologique.
Budget : 40 000 €
Contribution : Chef de projet, design et écriture du projet.

Projet de recherche, Fondation de la Mer, 2025 [En attente de financement].
Titre : CORSICORAL - Distribution, biodiversité et enjeux de conservation des forêts mésophotiques de coraux noirs en Corse.
Contribution : Chef de projet, design et écriture du projet.

Projet de thèse, Aspirant - F.R.S.-FNRS, 2025, [Financé, thèse de Baptiste Pinochet].
Titre: La capacité de séquestration du carbone des forêts sous-marines *de Posidonia oceanica* (L.) Delile du nord-ouest de la Méditerranée, dans le contexte du changement climatique et des espèces invasives.
Contribution: Participation au design et à l'écriture du projet, expertise technique.

Projet de recherche, Banque populaire de Méditerranée, 2025 [En attente de financement].
Titre : POSIBIO – POSIdonie et BIOingénierie pour la restauration des forêts marines de Méditerranée
Contribution : Participation au design et à l'écriture du projet, expertise technique

Projet de thèse, FRIA - F.R.S.-FNRS, 2025, [En attente de financement, thèse de Ana Lucía Azul Acuña].
Titre: SURF - Unveiling Microbiome Secrets to Enhance Seagrass Forests Restoration.
Contribution: Participation au design et à l'écriture du projet, expertise technique.

Missions de recherche

STARESO : Calvi (Corse) – Mai 2022 à Mai 2025

- T0 : Mai à Juillet 2022 (68 jours)
- T1 : Septembre/Octobre 2022 (44 jours)
- T2 : Avril à Juin 2023 (63 jours)
- T3 : Septembre/Octobre 2023 (21 jours)
- T4 : Avril/Mai 2024 (42 jours)
- REPAIR 2.0 : Juin 2024 (6 jours)
- T5 : Octobre 2024 (17 jours)
- T6 : Avril/Mai 2025 (35 jours)

Sorbonne Université : Paris (France) – Novembre/Décembre 2023 (21 jours)

Centro de Ciêncas do Mar : Faro (Portugal) – Juillet 2024 (21 jours)

Activités d'encadrement

Janvier – Juin 2022: Co-encadrement du mémoire de fin d'étude de Chiara Ruello (Università degli studi di Genova– Master en Biologie et Ecologie marine): « Long-term recolonization of *Posidonia oceanica* meadows – The case study of STARESO's harbour 54 years after its construction ».

Juillet 2022 : Encadrement de 5 stagiaires du master MER2030 lors de leur stage professionnalisant (2 semaines) à STARESO.

Janvier – Aout 2023 : Co-encadrement des mémoires de fin d'étude de :

- Morgan Frémal (Université de Liège – Master en Océanographie) : « Suivi du contenu azoté des boutures de *Posidonia oceanica* après une opération de restauration et mise au point d'une méthode d'identification de la présence des bactéries fixatrices d'azotes dans ses tissus racinaires ».
- Maria Margarita Lopez Velosa (MER2030 - Erasmus Mundus Joint Master Degree in Marine Environment) : " Temporal dynamics of photo-physiological and morphological parameters in *Posidonia oceanica* seagrass transplants in the Bay of Calvi, Corsica".

Juillet/Aout 2023 : Encadrement d'une stagiaire du master MER2030 dans le cadre d'un stage professionnalisant (3 semaines).

Janvier – Juin 2024 : Co-encadrement du mémoire de fin d'étude de Juliana Chapeyroux (Sorbonne Université – Master en Sciences de la Mer) : « Restauration des sillons d'ancrage dans l'herbier de *Posidonia oceanica* : suivi de la recolonisation naturelle et assistée en Corse ».

Aout 2024 : Encadrement de deux stagiaires du master MER2030 dans le cadre d'un stage professionnalisant (2 semaines).

Janvier – Aout 2025 : Co-encadrement des mémoires de fin d'étude de :

- Ana Lucía Azul Acuña (MER2030 - Erasmus Mundus Joint Master Degree in Marine Environment) : "Monitoring of the physiological performance of *Posidonia oceanica* (L.) Delile transplants from two donor populations in Alga Bay (Calvi, Corsica)".
- Lilly Mercedes Bucher (MER2030 - Erasmus Mundus Joint Master Degree in Marine Environment): "Integrating climate change and anthropogenic pressures in regional-scale site selection for *Posidonia oceanica* restoration in Corsica".

Formations suivies

Formations thématiques :

- **Télédétection appliquée à la gestion des milieux naturels** : Liège (Belgique) – Octobre/Décembre 2021
- **Modelling species distributions under climate change** : Copenhague (Danemark) – Aout/Septembre 2022 (5 jours)
- **Metabarcoding** : Roscoff (France) – Décembre 2022 (5 jours)
- **Threats to Marine Animal Forests and actions for conservation/restoration** : Banyuls-sur-Mer (France) – Septembre 2024 (5 jours)

Plongée scientifique, technique et moyens à la mer :

- **PA40 + CAH IB** : Ile Rousse (France) – Mai 2022 (10 jours)
- **N3 + Nitrox confirmé** : Marseille (France) – Octobre/Novembre 2022 (5 jours)
- **Recycleur diluant air (rEvo)** : La Ciotat (France) – Janvier/Février 2023 (5 jours)
- **Permis bateau côtier** : Anglet (France) – Aout 2023
- **Belgian Scientific Diver** : Calvi (France) – Septembre 2023 (12 jours)
- **CAH IIB** : Carnon (France) - Novembre 2023 (5 jours)
- **Recycleur trimix normoxique (rEvo)** : Marseille (France) – Avril 2024 (4 jours)
- **Recycleur trimix hypoxique (rEvo)** : La Ciotat (France) – Mai 2025 (5 jours)
- **CAH IIIB** : Concarneau (France) – Septembre 2025 (5 jours)

Communication et vulgarisation scientifique :

- Université de Liège. (2022). Le projet REPAIR : Des solutions fondées sur la nature pour réhabiliter les herbiers de posidonie. Retrieved from https://www.corsica.uliege.be/cms/c_15547781/fr/le-projet-repair-des-solutions-fondees-sur-la-nature-pour-rehabiliter-les-herbiers-de-posidonie
- Sea(e)scape (20 mars 2022). REPAIR: Des solutions fondées sur la nature pour réhabiliter les herbiers de posidonie. Retrieved from https://seaescape.fr/blog/2022/03/20/repair-restauration-posidonie/
- Ma Thèse en 180 secondes (MT180) - Comment sauver le poumon de la Méditerranée ? : Liège (Belgique) – Février/Mars 2023
  Lien de la présentation : https://www.youtube.com/watch?v=Ro4F6byyJfY
- Stassart, C. (2024, October 25). Mission de sauvetage pour les prairies sous-marines de Méditerranée. Daily Science. Retrieved from https://dailyscience.be/25/10/2024/mission-de-sauvetage-pour-les-prairies-sous-marines-de-mediterranee/

TABLE DES MATIERES

Chapitre I – Introduction générale 1

Chapitre II – Résilience et restauration de l'herbier de *Posidonia oceanica* après dégradation par l'ancrage : comparaison entre recolonisation naturelle et restauration active par transplantation 43

Article 1 : Assessing *Posidonia oceanica* recolonisation dynamics for effective restoration designs in degraded anchoring sites

Article 2 : Comparative assessment of transplantation methods and donor sources for the restoration of *Posidonia oceanica* meadows

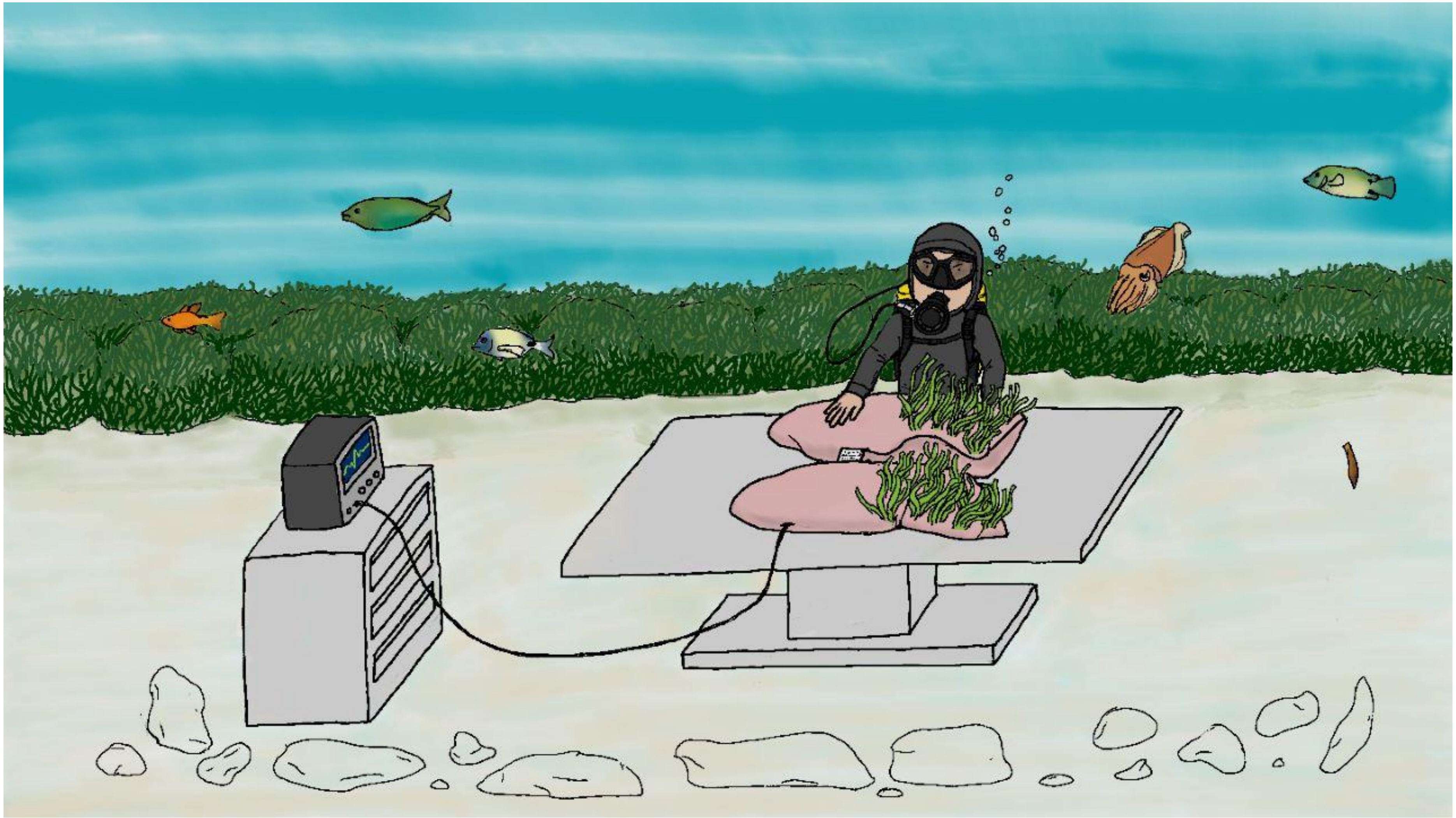
Article 3 : Microbiome matters: How transplantation methods and donor origins shape the successful restoration of the seagrass *Posidonia oceanica*

Article 4 : Physiological and biochemical performance indicators of *Posidonia oceanica* transplantation success

Liste des abréviations

1-λ′	Simpson index
ɑ	Photosynthetic efficiency
AHP	Analytical Hierarchy Process
AIC	Akaike Information Criterion
ANOVA	Analysis of variance
ASV	Amplicon Sequence Variant
B	Bore
C	Carbone
Ca	Calcium
$CaCO_3$	Carbonate de calcium
CAP	Canonical Analysis of Principal coordinates
CCMAR	Centro de Ciências do Mar
CIMAR	Centro de Investigação Marinha e Ambiental
Cl	Chlore
CO_2	Dioxyde de carbone
C_{org}	Carbone organique
Cu	Cuivre
DCE	Directive-Cadre sur l’Eau
DCSMM	Directive Cadre Stratégie pour le Milieu Marin
DNA	Deoxyribonucleic acid
df	Degree of freedom
EBQI	Ecosystem Based Quality Index
EMM	Estimated marginal mean
Fe	Fer
FNRS	Fonds National de la Recherche Scientifique
FOCUS	Freshwater and Oceanic sCience Unit of reSearch
Fv/Fm	Maximum photochemical quantum yield
GES	Gaz à effet de serre
GIEC	Groupe intergouvernemental d’experts sur l'évolution du climat
GIS	Geographical Information System
GLMM	Generalized Linear Mixed Model
H′	Shannon index
H_2S	Sulfure d’hydrogène
ICP-MS	Inductively Coupled Plasma Mass Spectrometry
K	Potassium
LDA	Linear Discriminant Analysis
LD	Limit of Detection
LEfSe	Linear Discriminant Analysis Effect Size
LETIS	Laboratory of Trophic and Isotope Ecology
LNHC	Low Nutrients High Chlorophyll
LOI	Loss on ignition
LQ	Limit of Quantification
MC	Monte Carlo
MCDA	Multi-criteria Decision Analysis
MHW	Marine Heat-Wave
Mg	Magnesium
Mn	Manganèse
Mo	Molybdène
MS	Mean Square
N	Azote

NA	Not Applicable
NDSM	Non-Destructive Shoot sampling Method
nMDS	Non-metric multidimensional Scaling
NW	North-West
P	Phosphore
PAM	Pulse-Amplitude-Modulated
PCR	Polymerase Chain Reaction
Perm	Permutations
PERMANOVA	Permutational multivariate analysis of variance
PIB	Produit Intérieur Brut
PPFD	Photosynthetic photon flux density
PREI	*Posidonia* Rapid Easy Index
PSII	Photosystem II
R&D	Recherche et Développement
rETR	Relative electron transport rate
rETRmax	Maximum relative electron transport rate
RLC	Rapid light curve
RNA	Ribonucleic acid
RSP	Réseau de Surveillance Posidonie
rRNA	Ribosomal ribonucleic acid
S	Soufre
SDM	Species Distribution Model
SIMPER	Similarity percentage
SS	Sum of squares
SSP	Shared Socio-economic Pathways
STARECAPMED	STAtion of Reference and rEsearch on Change of local and Global Anthropogenic Pressures on Mediterranean Ecosystems Drifts
STARESO	Station de recherches océanographiques et sous-marines
TCR	Total Carbohydrate Reserves
TEMPO	Réseau de suivi des herbiers de *P. oceanica* par télémétrie acoustique
ULiège	Université de Liège
UR	Unité de recherche
Y(II)	Effective photochemical quantum yield of the photosytem II
ZMEL	Zones de Mouillage et d'Equipement Légers
Zn	Zinc



Représentation graphique de ma thèse, illustré par Maxime Boulenger, lors de la finale régionale de "Ma thèse en 180 secondes" (Liège, 22 mars 2023).

Un jour, dit la légende, il y eut un immense incendie de forêt.

Tous les animaux terrifiés, atterrés, observaient impuissants le désastre qui s'étendait sous leurs yeux.

Seul le petit colibri s'activait, allant chercher quelques gouttes avec son bec pour les jeter sur le feu.

Après un moment, le tatou, agacé par cette agitation qui lui semblait dérisoire, lui dit :

« Colibri ! Tu n'es pas fou ? Ce n'est pas avec ces gouttes d'eau que tu vas éteindre le feu ! »

Et le colibri lui répondit : « Je le sais, mais je fais ma part. »

Légende amérindienne

Chapitre I

Introduction générale

"The next century will, I believe, be the era of restoration in ecology."

–E.O. Wilson (1992).

1. L'écologie de la restauration : une science en plein essor

La décennie 2021-2030 a été proclamée par les Nations Unies comme la *Décennie pour la restauration des écosystèmes*, appelant les pays du monde entier à mettre un terme à la dégradation des milieux naturels et à restaurer ceux déjà endommagés (UNEP, 2021). De plus, en février 2024, le Parlement européen a adopté la Loi sur la restauration de la nature. Dans le cadre de cette législation, les États membres de l'Union européenne sont tenus de restaurer au moins 30 % des habitats terrestres et marins en mauvais état d'ici 2030, 60 % d'ici 2040 et 90 % d'ici 2050. Depuis plusieurs décennies, l'écologie de la restauration connait un essor considérable, accompagnée d'une multiplication de projets sur le terrain. Cette discipline scientifique relativement récente trouve ses origines dans les années 1930 avec les travaux d'Aldo Leopold, professeur à l'Université du Wisconsin-Madison aux États-Unis. Considéré comme un pionnier du domaine, il initia les premiers projets de restauration écologique en reconstruisant les prairies indigènes du Wisconsin, caractéristiques des paysages nord-américains du XIXe siècle, avant l'arrivée des premiers colons (ALF, 2005). En 1987, la *Society for Ecological Restoration* fut fondée aux États-Unis afin de structurer et promouvoir les pratiques et politiques de restauration des écosystèmes à l'échelle internationale (Dutoit, 2011).

En France, l'émergence de cette discipline a été plus tardive. L'adoption de la loi relative à la protection de la nature (*Loi n° 76-629 du 10 juillet 1976*) a marqué une première avancée en instaurant l'obligation d'éviter, de réduire ou de compenser les impacts environnementaux des projets d'aménagement du territoire (Gallet et al., 2017). Cependant, cette législation n'a pas conduit au développement d'actions concrètes de restauration écologique (Barnaud, 1995). Il fallut attendre 1989 pour qu'un premier symposium dédié à la restauration des écosystèmes soit organisé en France, à l'occasion d'un projet de réhabilitation du marais Vernier en Haute-Normandie (Pfeiffer, 2007). Depuis cet événement fondateur, l'intérêt des pouvoirs publics pour l'écologie de la restauration n'a cessé de croitre, entrainant une multiplication des initiatives.

L'adoption des *Grenelles de l'Environnement* en 2008 a marqué un tournant décisif. L'article 23 des Grenelles I et II a imposé la mise en place de mesures de compensation écologique dans le cadre de la *Trame verte et bleue*, un réseau de continuités écologiques terrestres et aquatiques à l'échelle du territoire national français, à l'exception du milieu marin (Gauthier-Clerc et al., 2014 ; Bergès et al., 2010). Un an plus tard, en février 2009, le *Grenelle de la Mer* est venu compléter les engagements des *Grenelles de l'Environnement* en instaurant une *Trame bleue marine*. Cette dernière élargit ainsi le concept de connectivité écologique aux espaces littoraux et marins. Cette initiative vise à préserver, gérer et restaurer les corridors biologiques marins et côtiers, renforçant ainsi la cohérence écologique des milieux aquatiques en France (Le Livre Bleu des engagements du Grenelle de la Mer, 2009).

a. *Définitions et concepts clés*

L'écologie de la restauration est aujourd'hui couramment définie comme le processus visant à assister, de manière intentionnelle, le rétablissement d'un écosystème considéré comme dégradé ou détruit. Ce rétablissement est généralement envisagé comme un retour à l'état originel en termes de composition des espèces, de structure des communautés et de fonctionnement écologique (SER, 2004). Toutefois, cette définition idéaliste de la restauration est rarement applicable dans la réalité, car les dommages écologiques subis par les écosystèmes sont souvent irréversibles. En pratique, il s'agit fréquemment de « réhabilitation », c'est-à-dire du rétablissement de certaines fonctions écologiques essentielles, sans pour autant restaurer intégralement un écosystème à son état initial (Choi, 2007). Cette distinction est d'autant plus importante que la sélection d'un état de référence pour guider la restauration est souvent arbitraire et subjective (Choi, 2007). En effet, la perception de la normalité écologique varie d'une génération à l'autre, un phénomène connu sous le nom d'amnésie générationnelle ou syndrome du changement de base ('*shifting baseline syndrome*' en anglais) (Leather & Quicke, 2009 ; Papworth et al., 2009 ; Soga & Gaston, 2018). Ce concept, introduit par Pauly (1995) dans le domaine de la gestion des pêches, met en évidence la tendance des scientifiques et gestionnaires à considérer comme normaux les niveaux actuels d'abondance et de diversité, sans tenir compte des déclins historiques. Ainsi, chaque génération intègre progressivement la raréfaction des espèces et l'appauvrissement de la biodiversité comme un état de référence, effaçant progressivement la mémoire des conditions écologiques passées. Pour ces raisons, et afin de simplifier la terminologie dans ce manuscrit, les termes « restauration » et « réhabilitation » seront employés de manière interchangeable.

La restauration implique ainsi diverses actions visant à rétablir partiellement, ou totalement un état proche de l'état de référence d'un écosystème. Les interventions varient, allant de la non-intervention à la réintroduction d'espèces. Trois grandes approches sont généralement distinguées. La première, la restauration ou régénération naturelle est la plus simple à mettre en place et est appliquée à des milieux peu dégradés où des populations saines environnantes permettent, sur une échelle de temps raisonnable de recoloniser le milieu dégradé après cessation des activités néfastes (ex : amélioration de la qualité de l'eau, réglementation sur l'ancrage) (Figure 1.1). La deuxième, la restauration ou régénération assistée est utilisée lorsque les conditions environnementales ne sont plus adéquates afin de favoriser ensuite le rétablissement naturel des composantes biotiques (ex : création d'habitats artificiels) (Figure 1.1). Ces deux approches sont souvent regroupées sous le terme de « restauration passive ». La troisième, la reconstruction ou restauration dite « active » demande des moyens plus importants et est employée pour des écosystèmes fortement dégradés. Elle nécessite de réduire, voire d'éliminer toutes les causes de dégradation, de restaurer les composantes biotiques et abiotiques, et de réintroduire certaines espèces (Figure 1.1) (Atkinson & Bonser, 2020).

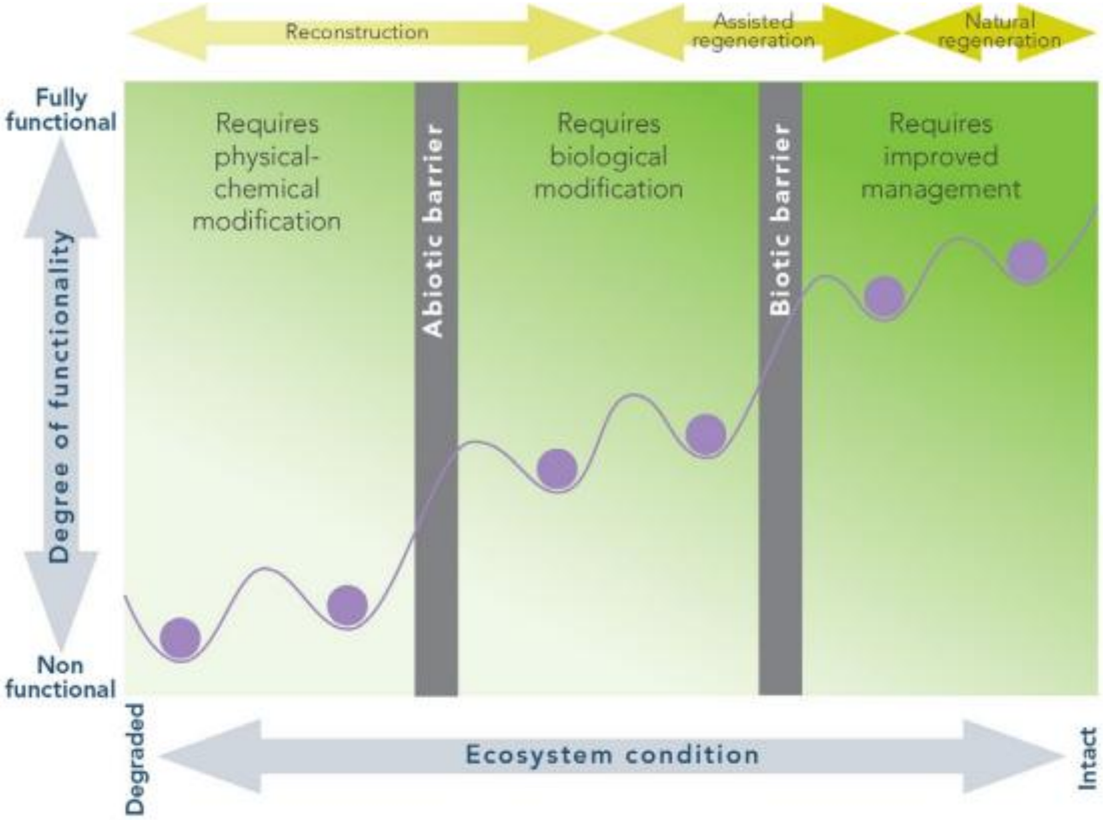


Figure 1.1. Modèle conceptuel de la dégradation des écosystèmes et des solutions apportées par différents niveaux de restauration. Les creux dans le diagramme représentent des points de stabilité dans lesquels un écosystème peut rester dans un état stable avant d'être déplacé (par un événement ou un processus de restauration ou de dégradation) par-dessus une barrière (représentée par les barres grises dans le diagramme) vers un degré de fonctionnalité supérieur ou inférieur (McDonald et al., 2016).

Une composante essentielle de l'écologie de la restauration concerne l'évaluation de réussite des opérations de restauration écologique. La sélection d'indicateurs de réussite est primordiale pour garantir les trajectoires initiées lors des projets de restauration et les valider rapidement, tout en minimisant les suivis couteux sur le long terme (Gauthier-Clerc et al., 2014). Un projet de restauration correctement planifié vise à atteindre des objectifs clairement définis dès le départ, reflétant les attributs essentiels de l'écosystème de référence (SER, 2004). Malgré la multiplication des projets de restauration et l'importance du suivi et de l'évaluation de leur succès, les retours d'expérience détaillés et critiques restent rares (Suding, 2011). En effet, il est fréquent que les objectifs initiaux soient mal définis, que le suivi du projet soit insuffisant ou que les données quantitatives fassent défaut. Ces lacunes entravent la compréhension et l'amélioration des pratiques de restauration écologique (Hobbs, 2007 ; Wortley et al., 2013). Concernant les paramètres fréquemment évalués, la majorité des études se focalisent sur des indicateurs écologiques, tels que la diversité biologique ou les fonctions écosystémiques (Ruiz-Jaen & Aide, 2005). En revanche, les paramètres socio-économiques sont rarement intégrés dans les objectifs de départ et les plans de suivi (De Groot et al., 2013 ; Hobbs, 2007; Martin, 2017). Pour conclure, il est essentiel de définir des objectifs adaptés à chaque projet, en fonction de son ampleur et des motivations sous-jacentes à l'effort de restauration (Ehrenfeld, 2000). Une approche intégrative, combinant des dimensions écologiques et sociales, notamment l'évaluation économique des services écosystémiques post-restauration, apparait indispensable lors de la mise en place d'opérations de restauration écologique (Hobbs, 2007).

b. La restauration des écosystèmes marins : prise de conscience

Les marais salants, les herbiers marins, les forêts de macroalgues, les récifs de bivalves, les récifs coralliens et les mangroves sont des écosystèmes côtiers extrêmement productifs qui abritent une biodiversité remarquable (James et al., 2024 ; Lau, 2013). Malheureusement, ces habitats uniques ont considérablement régressé au cours de ces dernières décennies et disparaissent à un rythme alarmant. Il a été estimé que leur taux de perte est plus rapide que celui des forêts tropicales à l'échelle mondiale (Duarte et al., 2008). En 2008, plus de 40 % de l'espace marin global était affecté par diverses pressions anthropiques, et en 2014, 66 % subissaient des impacts cumulatifs de plus en plus conséquents (IPBES, 2019). Toutefois, il convient de nuancer ces observations : la perte de biodiversité dans les océans est moins marquée qu'en milieu terrestre, et de nombreuses populations d'espèces marines peuvent se rétablir une fois les pressions réduites ou supprimées (Duarte et al., 2020; McCauley et al., 2015). Pourtant, comparée à la restauration des écosystèmes terrestres, celle des écosystèmes marins demeure un domaine relativement récent, avec un nombre de projets en cours encore limité (Blignaut et al., 2013 ; Elliott et al., 2007 ; Saunders et al., 2020 ; Stewart-Sinclair et al., 2020). Plusieurs facteurs expliquent ce retard : la mise en œuvre de tels projets en milieu marin est plus complexe, les coûts financiers sont élevés, et l'incertitude quant aux résultats limite la confiance des investisseurs et des décideurs (Saunders et al., 2020). Si certains processus, comme les interactions entre sédiments et hydrographie ou la qualité des sédiments après une pollution, sont relativement bien étudiés, d'autres restent encore mal compris. Par exemple, les conséquences écologiques d'une réintroduction d'espèces ou la taille minimale requise d'une population pour assurer sa viabilité sont des paramètres encore incertains. Ce manque de connaissances a constitué un frein majeur au développement et au financement des opérations de restauration écologique en milieu marin (Elliott et al., 2007).

Les premiers projets de restauration en milieu marin ont émergé dans les années 1960 et connaissent actuellement une expansion, en particulier pour certaines espèces côtières formatrices d'habitat comme les récifs d'huitres (Figure 1.2) (Duarte et al., 2020). En revanche, pour d'autres habitats tels que les forêts de kelp, la restauration en est encore à ses débuts, avec moins de 100 projets recensés à l'échelle mondiale en 2020 (Figure 1.2) (Duarte et al., 2020). De plus, la majorité des projets de restauration en milieu marin sont concentrés en Amérique du Nord, en Europe et en Australie, où des réglementations imposent des mesures de compensation/restauration et où les ressources financières permettent d'assumer les couts financiers importants de ces initiatives (Elliott et al., 2007).

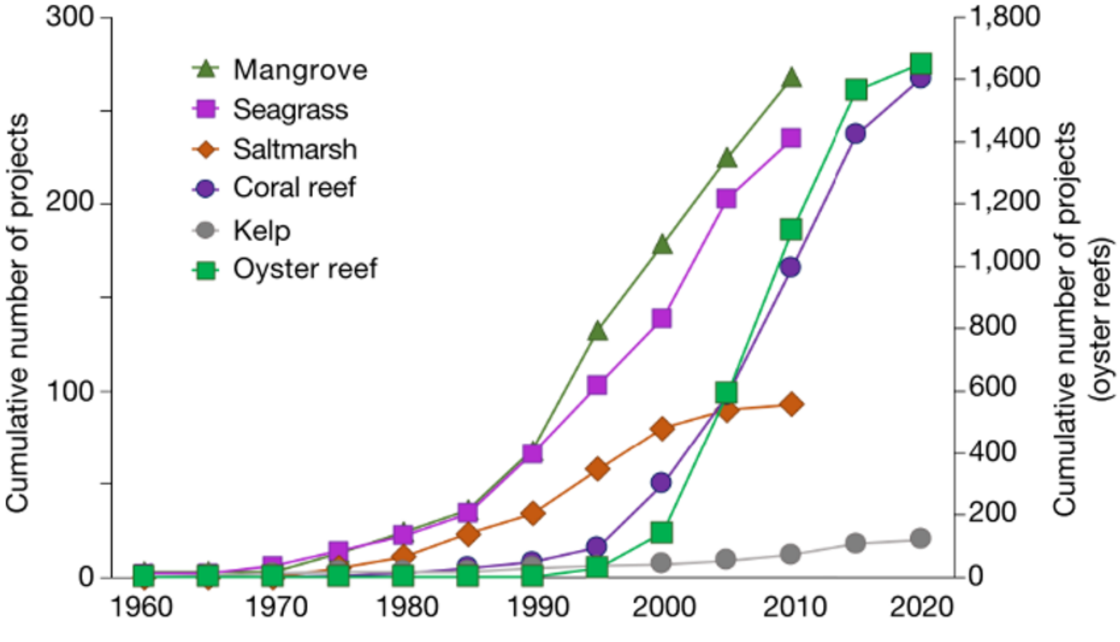


Figure 1.2. *Augmentation du nombre de projets de restauration en milieu marin de 1960 à 2020 (Duarte et al., 2020).*

L’évolution temporelle de la restauration des écosystèmes marins côtiers révèle que cette discipline s’est développée sur une période bien plus courte que la restauration des milieux terrestres, ce qui explique en partie son efficacité encore limitée. Toutefois, compte tenu de son développement récent et des connaissances encore relativement lacunaires, il est probable que la restauration des milieux marins progresse rapidement au cours des prochaines décennies et devienne une stratégie d'intervention efficace pour la réhabilitation des écosystèmes côtiers. Malgré cette relative immaturité, plusieurs projets de restauration d’écosystèmes côtiers ont déjà rencontré un succès notable. Des initiatives de grande envergure ont permis la restauration de plus de 1000 hectares de mangroves, de marais salants et d’herbiers marins, avec des résultats démontrant leur persistance sur plusieurs décennies (Saunders et al., 2020). Par exemple, la réserve de biosphère de la forêt de mangrove de Can Gio, au Vietnam, avait été fortement dégradée par l’épandage d’agent orange durant la guerre du Vietnam. Entre 1978 et 1998, 31 000 hectares ont été restaurés. Comparée à une foret naturellement régénérée, la forêt de Can Gio présente une structure de végétation et une capacité de stockage de carbone similaires, tout en abritant une plus grande diversité végétale (Nam et al., 2016). En Floride, aux États-Unis, une étude portant sur plusieurs sites d’herbiers marins a révélé que la plupart des herbiers restaurés jusqu’à 32 ans auparavant étaient toujours en place, avec une couverture et une composition d'espèces similaires à celles des herbiers de référence (Rezek et al., 2019). Toutefois, de nombreuses questions restent à explorer pour favoriser le développement de cette discipline dans les années à venir. Il sera notamment nécessaire d’établir un inventaire complet des zones restaurées à ce jour et de cartographier à grande échelle les sites potentiellement appropriés pour de futures opérations de restauration (Duarte et al., 2020 ; Waltham et al., 2020). Il est également essentiel de déterminer la vulnérabilité des écosystèmes marins restaurés face aux effets du changement climatique, ainsi que la variabilité spatio-temporelle des services écosystémiques qu’ils fournissent (Saunders et al., 2020 ; Waltham et al., 202).

c. L’ingénierie écologique et les solutions fondées sur la nature

Dans la continuité du développement de l’écologie de la restauration, le concept d’ingénierie écologique a émergé afin de faire le lien entre l’écologie et l’ingénierie. Cette discipline a été introduite par l’ingénieur Howard Odum dans les années 1960 et définie comme l’optimisation de la mise en œuvre de projets d’ingénierie s’appuyant sur les processus des écosystèmes naturels (Odum & Odum, 2003). Depuis, cette définition a été affinée. Les principaux objectifs de l’ingénierie écologique sont désormais axés sur la restauration d’écosystèmes fortement dégradés par l’activité humaine et sur la conception de systèmes écologiques pouvant constituer une alternative aux infrastructures artificielles pour répondre à divers besoins humains (Bergen et al., 2001 ; Mitsch, 2012 ; Mitsch & Jørgensen, 2003). Ce second objectif rejoint la définition des solutions fondées sur la nature (‘*nature-based solutions’* en anglais) proposée par l’UICN : « *les actions visant à protéger, gérer de manière durable et restaurer des écosystèmes naturels ou modifiés pour relever directement les défis de société de manière efficace et adaptative, tout en assurant le bien-être humain et en produisant des bénéfices pour la biodiversité.* » (Cohen-Shacham et al., 2016). Grace aux nombreux services écosystémiques qu’ils fournissent, les écosystèmes naturels sont de plus en plus intégrés comme alternatives ou compléments aux infrastructures artificielles (Seddon et al., 2020). Un exemple bien documenté d’application en milieu marin concerne la protection côtière (Borsje et al., 2011). En effet, certains écosystèmes côtiers, comme les herbiers marins et les récifs de bivalves, jouent un rôle clé dans la protection du littoral contre l’érosion et les inondations (Figure 1.3). Ces espèces dites « ingénieures » contribuent à atténuer ces phénomènes grâce à plusieurs processus écologiques, notamment l’amortissement des vagues, l’accumulation de sédiments et la stabilisation des fonds marins (Borsje et al., 2011 ; Gracia et al., 2018 ; Morris et al., 2018).

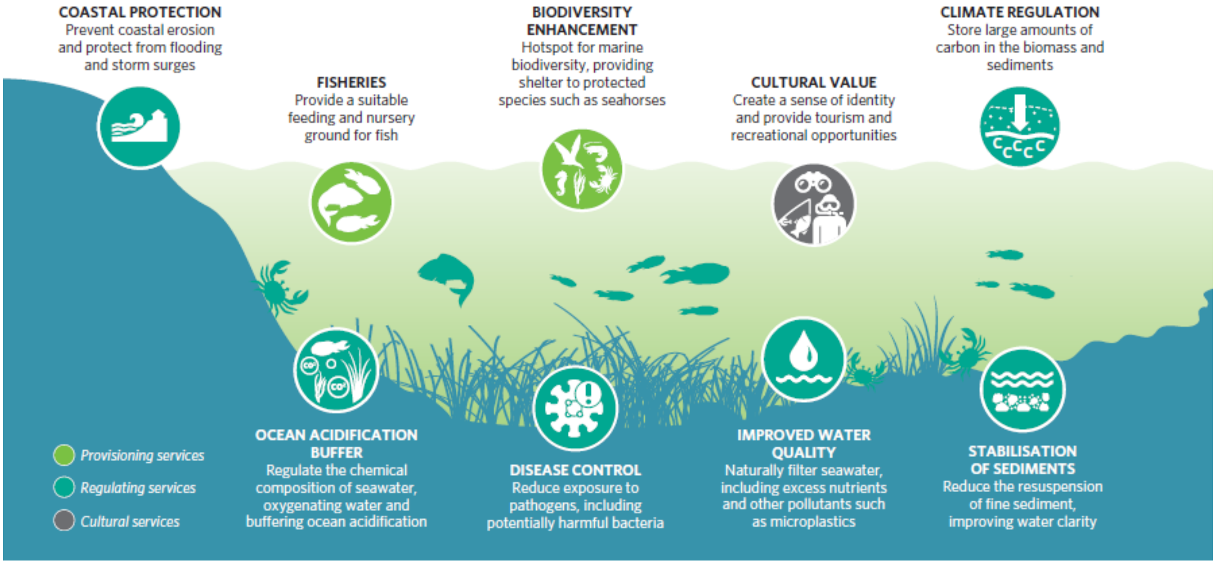


Figure 1.3. *Services écosystémiques apportés par les herbiers marins (Gamble et al., 2021).*

En outre, l'utilisation de ces écosystèmes pour la protection côtière peut s’avérer plus rentable à long terme que les infrastructures traditionnelles telles que les digues ou les brise-lames. Par exemple, dans l'estuaire d'Humber, au Royaume-Uni, une étude a montré qu’après 25 ans, la restauration des marais salants était économiquement plus avantageuse que le maintien des digues (Turner et al., 2007). En raison de leurs nombreux avantages tant écologiques qu'économiques, ces écosystèmes suscitent un intérêt croissant pour être restaurés ou recréés afin de compléter, voire remplacer, certaines infrastructures artificielles de protection côtière (Spalding et al., 2014 ; Temmerman et al., 2013).

2. La Méditerranée : une mer d’exception sous pressions

a. *Un océan miniature*

La *Mare medi terraneum*, en latin, désigne la Méditerranée comme une « mer au milieu de la terre », encadrée par l’Europe, l’Asie et l’Afrique. Elle est la plus vaste (2 969 000 km²) et la plus profonde (5 267 m) des mers semi-fermées sur Terre (Coll et al., 2010). Elle couvre 0,7 % de la surface totale des océans, 0,3 % du volume total, et sa profondeur moyenne est de 1500 m (Bethoux et al., 1999). La Méditerranée est souvent qualifiée d’ « océan miniature », où divers processus océanographiques, tels que la formation d’eau dense ou la circulation thermohaline, se produisent à une échelle réduite mais similaire à celle des océans (Bethoux et al., 1999). Elle est connectée à l’Atlantique par le détroit de Gibraltar (13 km de large) à l’ouest, et à la mer de Marmara et la mer Noire par le détroit des Dardanelles à l’est. Elle est également reliée à la mer Rouge par le canal de Suez depuis 1869. Le canal de Sicile divise la Méditerranée en un bassin oriental (1,65 million de km²) et un bassin occidental (0,85 million de km²) (Coll et al., 2010 ; Sebastián et al., 2021). La Méditerranée se distingue par plusieurs caractéristiques uniques. Les eaux méditerranéennes entre 300 et 500 m de profondeur sont homogènes en température (homothermie) jusqu’aux plaines abyssales, avec une température constante d’environ 13 °C (Bethoux et al., 1999 ; Coll et al., 2010). A profondeur équivalente, la température est d’environ 2 °C dans l’Atlantique (Bethoux et al., 1999). La Méditerranée est une mer fortement salée, avec une salinité comprise entre 37,5 et 39,5 psu, bien supérieure à celle des océans voisins (Coll et al., 2010).

La Méditerranée a connu une histoire géologique complexe, notamment une période d’isolement du reste des océans qui a conduit à son assèchement presque total lors de la crise messinienne, il y a environ 5,96 millions d’années. Cet évènement a entrainé des changements drastiques du climat, du niveau de la mer et de la salinité (Garcia-Castellanos et al., 2009). Aujourd’hui, la Méditerranée est caractérisée par un climat continental marqué avec des étés chauds et secs et des hivers doux et humides (Giorgi & Lionello, 2008). Ce climat entraine une évaporation importante, responsable d’un bilan hydrique négatif, car les faibles précipitations et les apports fluviaux sont insuffisants pour compenser les pertes par évaporation. Ce déficit hydrique est compensé par un apport d’eaux de surface en provenance de l’Atlantique, qui transitent par le détroit de Gibraltar (Bethoux et al., 1999 ; Coll et al., 2010). Par ailleurs, la température moyenne des eaux de surface de la Méditerranée varie fortement selon les saisons et présente un gradient marqué d’ouest en est ainsi que du nord au sud du bassin (Figure 1.4) (Hopkins, 1985 ; Pisano et al., 2020).

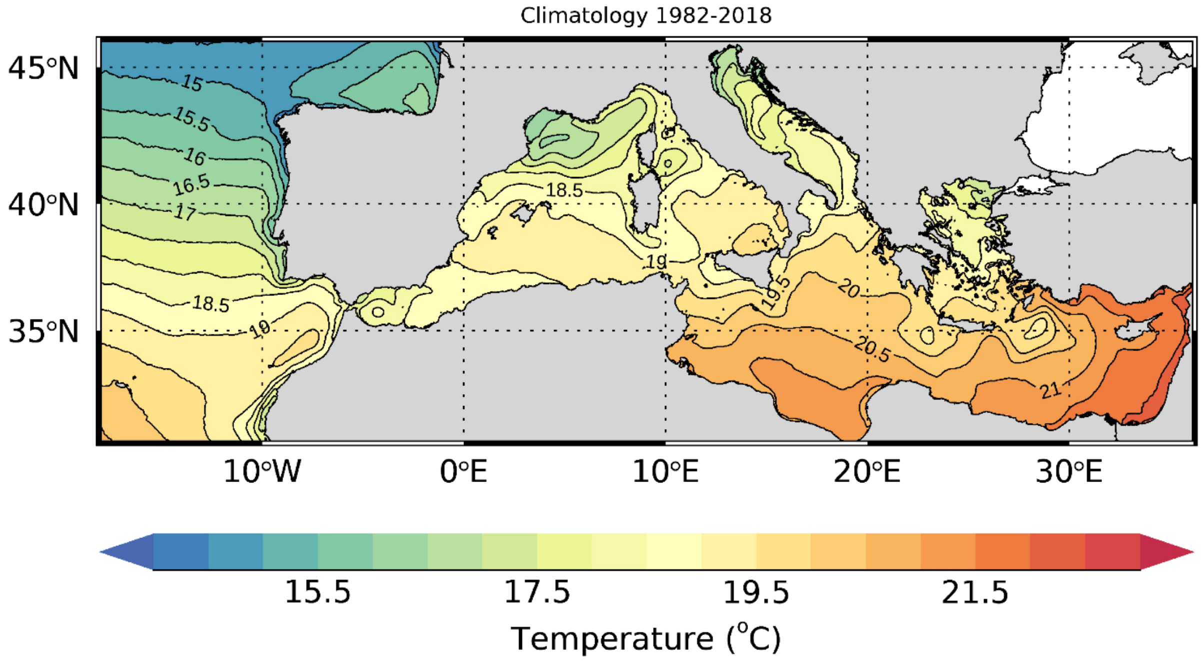


Figure 1.4. *Moyenne de la température (°C) des eaux de surface de la Méditerranée de 1982 à 2018 (Pisano et al., 2020).*

b. Oligotrophie : caractéristique intrinsèque de la Méditerranée

L'entrée des eaux de surface de l'Atlantique via le détroit de Gibraltar, combinée à la descente en profondeur des masses d'eau refroidies au niveau du golfe du Lion, du nord de l'Adriatique et du nord de la mer Égée génère une circulation thermohaline à l'échelle de la Méditerranée (Lejeusne et al., 2010). Ce flux d'eaux de surface venant de l'Atlantique est principalement dû à l'évaporation intense dans le bassin occidental, qui entraine une baisse localisée du niveau d'eau de la mer ainsi qu'une augmentation de la salinité d'ouest en est. Ce gradient crée un courant d'eau froide et relativement peu salée qui pénètre dans la Méditerranée via le détroit de Gibraltar (Coll et al., 2010). La faible profondeur et l'étroitesse du détroit limitent le mélange entre les différentes couches d'eau, ne permettant qu'un apport en eaux de surface (Sebastián et al., 2021). Ces eaux de surface n'étant pas enrichies par un phénomène d'upwelling, elles restent relativement pauvres en nutriments (Bethoux et al., 1999). Malgré cet apport limité, les nutriments fournis par les eaux de surface atlantiques, combinés au brassage hivernal des eaux de surface avec les eaux plus profondes, favorisent une productivité primaire légèrement plus élevée dans le bassin occidental. Outre cette contribution, les principales sources de phosphore, d'azote et de silice proviennent des apports fluviaux, des dépôts atmosphériques et des poussières sahariennes (Bethoux et al., 1999 ; Estrada, 1996). La productivité primaire de la Méditerranée ne représente qu'environ 1% de la productivité primaire globale (Uitz et al., 2010). Cependant, certains organismes photosynthétiques, tels que les herbiers de magnoliophytes marines, ont développé des adaptations remarquables à cette faible disponibilité en nutriments, leur permettant de maintenir une biomasse et une productivité importantes. On parle de système « LNHC » pour « *Low Nutrients – High Chlorophyll* » (Gobert et al., 2002).

c. Point chaud de la biodiversité : endémisme et espèces invasives

L'histoire géologique et les conditions océanographiques particulières de la Méditerranée ont favorisé l'expansion d'une grande richesse d'espèces et d'un fort taux d'endémisme faisant de la Méditerranée un point chaud mondial de biodiversité (Bianchi et al., 2012 ; Coll et al., 2010 ; Lejeusne et al., 2010). Bien qu'elle ne représente que 0,7 % de la surface totale des océans et 0,3 % de leur volume total, elle abrite entre 4 et 18 % des espèces marines mondiales (Bianchi & Morri, 2000 ; Bianchi et al., 2012). Parmi les 17 000 espèces recensées en Méditerranée, environ un quart sont endémiques (Bianchi et al., 2012 ; Coll et al., 2010). D'autre part, plus de 1 000 espèces invasives ont été recensées, allant des macrophytes aux poissons et invertébrés (Katsanevakis et al., 2014 ; Öztürk,2021). Elle présente le taux le plus élevé d'espèces exotiques parmi toutes les mers du globe (Boudouresque & Verlaque, 2005). Depuis la fin des années 1990, le taux moyen annuel d'invasions a considérablement augmenté (Galil & Zenetos, 2002 ; Katsanevakis et al., 2014 ; Öztürk,2021). Certains auteurs rapportent une hausse de près de 150 % (Raitsos et al., 2010). Cette accélération est principalement due au réchauffement des eaux méditerranéennes, permettant ainsi à des espèces thermophiles de s'installer et de prospérer (Galil & Zenetos, 2002 ; Lejeusne et al., 2010). Par conséquent, la barrière thermique et saline qui existait autrefois entre la Méditerranée et la mer Rouge s'est atténuée, favorisant la colonisation d'espèces invasives en provenance de la Mer Rouge via le canal de Suez (Raitsos et al., 2010). Avec les prévisions de hausse continue des températures de l'eau en Méditerranée dans les décennies à venir, il est probable que cette dynamique favorise encore davantage les espèces thermophiles, au détriment des espèces indigènes (Galil & Zenetos, 2002). L'aquaculture constitue une autre voie majeure d'introduction d'espèces invasives, soit par l'évasion d'espèces élevées, soit via l'introduction accidentelle d'espèces associées à ces activités. Le transport maritime et les échappées d'aquarium sont également des vecteurs d'introduction (Boudouresque & Verlaque, 2005 ; Raitsos et al., 2010).

d. Impacts du changement global

La Méditerranée est une zone de transition située à l'interface entre l'aridité de l'Afrique du Nord et les régions tempérées et pluvieuses d'Europe centrale. Cela implique que même de faibles changements de la circulation atmosphérique globale peuvent engendrer d'importantes modifications du climat méditerranéen. En conséquence, cette région est considérée comme l'une des plus vulnérables au changement global (Giorgi, 2006 ; Giorgi & Lionello, 2008). Cette vulnérabilité est déjà perceptible à travers l'augmentation des températures de l'eau, aussi bien en surface qu'en profondeur, depuis 1975 (Coma et al., 2009). Selon les projections, d'ici la fin du XXIe siècle, l'augmentation de la température et de la salinité de la colonne d'eau pourrait réduire la circulation thermohaline, diminuant de 40 % les eaux intermédiaires et de 80% les eaux profondes (Somot et al., 2006). Cette réduction du brassage pourrait limiter l'apport en nutriments par les mouvements d'upwelling, entrainant une diminution de la production primaire et perturbant l'écosystème méditerranéen dans son ensemble (Skliris, 2014). Par ailleurs, le changement climatique entraine une augmentation de la fréquence et de l'intensité des évènements extrêmes tels que les canicules marines, responsables de mortalités massives parmi les organismes marins (Garrabou et al., 2022 ; Templado, 2014). Ces canicules, combinées au réchauffement des eaux, constituent une menace majeure pour la biodiversité et les écosystèmes marins, déjà fragilisés par d'autres pressions (Smale et al., 2019 ; Templado, 2014).

En parallèle, l'augmentation des émissions de CO_2 , principale cause des changements climatiques, entraine une acidification progressive des océans. Depuis la révolution industrielle, une baisse globale du pH de 0,1 a été observée dans les océans, mais cette diminution atteint 0,14 dans la Méditerranée occidentale (Calvo et al., 2011). Le pH de la Méditerranée pourrait diminuer de 0,3 d'ici 2100 (Mcneil et al., 2007). La combinaison de températures élevées et de l'acidification affecte particulièrement les organismes marins calcifiants, dont les squelettes, les coquilles ou les plaques sont constitués de carbonate de calcium ($CaCO_3$). Ces organismes sont nombreux et d'origine phylogénétiques très variée : coraux scléractiniaires, bryozoaires, algues corallines, crustacés, ainsi que certains phylums de phytoplancton (comme les coccolithophoridés) et de zooplancton (Templado, 2014).

e. Artificialisation côtière et tourisme de masse

La Méditerranée est une région densément peuplée, bordée par 21 pays répartis sur trois continents. La population totale des pays méditerranéens est passée de 276 millions en 1970 à 466 millions en 2010, dont un tiers vit dans des agglomérations côtières (UNEP/MAP, 2012).

A cela s’ajoutent près de 400 millions de touristes annuels, soit environ un tiers du tourisme mondial (Plan Bleu, 2022). Ces visiteurs se concentrent principalement dans les zones côtières, faisant du tourisme côtier un moteur économique clé et un important générateur d’emplois pour les pays méditerranéens (Drius et al., 2019). Cette dynamique touristique, combinée à la demande croissante en logements, a favorisé le développement d'infrastructures massives et l’artificialisation des zones côtières (García-Nieto et al., 2018). Cette urbanisation croissante du littoral engendre des conséquences environnementales graves : destruction d’habitats, modification des caractéristiques environnementales locales, augmentation de l’érosion côtière (hydrodynamisme, sédimentation) et intensification de la pollution (Gianni et al., 2013 ; Pasqualini et al., 1999). La destruction des habitats marins figure parmi les principales causes responsables de la régression de la biodiversité en Méditerranée (Coll et al., 2010).

3. Les herbiers de *Posidonia oceanica* : joyaux de la Méditerranée

a. Les Magnoliophytes marines : adaptations au milieu marin et fonctions écologiques

Parmi les plantes supérieures ayant quitté le milieu marin il y a environ 400 millions d'années, les Magnoliophytes marines ('*seagrasses*' en anglais) se distinguent comme le seul groupe à avoir recolonisé l'environnement marin et retrouvé un mode de vie entièrement submergé. Les Magnoliophytes marines possèdent des caractéristiques communes qui leur ont permis de s'adapter à une vie immergée en milieu halin, à savoir pouvoir accomplir l'entièreté de leur cycle vital en milieu salin, pouvoir croitre et se développer en étant complètement submergées, posséder un système d'ancrage (rhizomes et racines) pour résister à la force du courant et une stratégie de pollinisation hydrophile (Den Hartog & Kuo, 2006). Ces plantes forment un groupement écologique polyphylétique comprenant 65 espèces (Den Hartog & Kuo 2006; Larkum et al., 2018; Les et al., 1997). L'ensemble de ces espèces fait partie de six familles de Monocotylédones au sein de la sous-classe des Alismatidae : Hydrocharitaceae, Zannichellaceae, Ruppiaceae, Zosteraceae, Cymodoceaceae et Posidoniaceae (Den Hartog & Kuo, 2006 ; Les et al., 1997). Ces espèces se retrouvent principalement dans les régions tempérées et tropicales (Figure 1.5).

La superficie totale couverte par les herbiers marins est estimée entre 160 387 km² et 266 562 km², soit près de 1 % des océans côtiers et 0,07 % de l'océan mondial (Figure 1.5). A titre de comparaison, les récifs coralliens couvrent environ 284 803 km² et les mangroves 152 361 km² (McKenzie et al., 2020). La faible diversité phylogénétique des Magnoliophytes marines s'explique par leur gamme restreinte de cycles de vie (Short et al., 2007). En effet, toutes ces espèces se reproduisent soit de manière asexuée par croissance horizontale des rhizomes, soit de manière sexuée par production de fruits, de graines ou de plantules vivipares (Kuo & Kirkman, 1987).

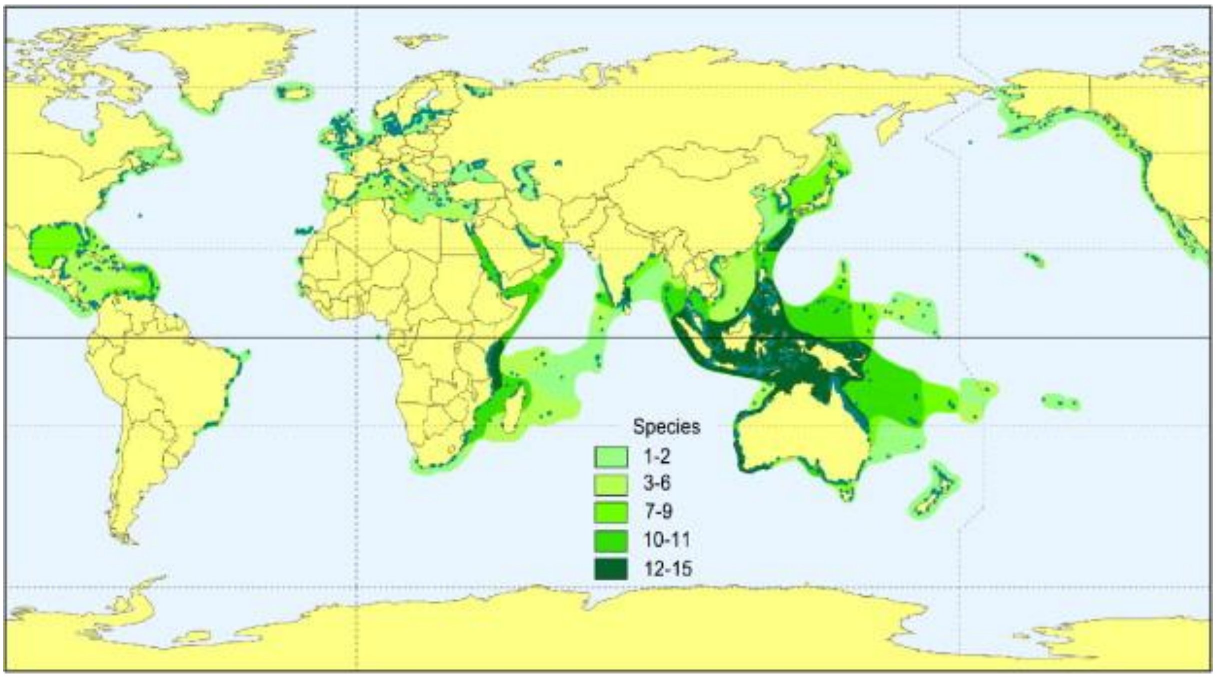


Figure 1.5. *Distribution globale de la diversité spécifique des Magnoliophytes marines (Short et al., 2007).*

Les herbiers marins constituent des écosystèmes côtiers fondamentaux en raison de leur importance pour les réseaux trophiques, la connectivité écologique et le cycle de vie de nombreuses espèces (Cullen-Unsworth & Unsworth, 2013). En effet, les assemblages de Magnoliophytes marines forment un habitat pour les stades de développement de diverses espèces, fournissant ainsi un rôle de nurserie, d'abri et de zone d'alimentation pour de nombreux poissons et invertébrés (Cullen-Unsworth & Unsworth, 2013 ; Unsworth et al., 2019). Ce rôle fonctionnel essentiel est assuré par la combinaison du complexe rhizome-racines et de la canopée foliaire, offrant une complexité structurelle semblable à celle des espèces calcifiantes formatrices de récifs. Pour ces raisons, les herbiers marins sont également qualifiés d'espèces ingénieures ou fondatrices, au même titre que les récifs coralliens (Cullen-Unsworth & Unsworth, 2013). Un des services écosystémiques essentiels dans le contexte de changement climatique actuel est le rôle des herbiers marins en tant que puits de carbone. A l'inverse des forêts où le carbone organique (C_{org}) est stocké dans la biomasse vivante, les herbiers marins séquestrent du carbone majoritairement dans les sédiments et ce jusqu'à 50 fois plus que les forêts tropicales, tempérées ou boréales (McLeod et al., 2011). Ensuite, la présence de Magnoliophytes marines permet également de limiter l'érosion côtière. La canopée des herbiers marins réduit l'intensité des vagues et du courant, ce qui favorise la sédimentation et en diminue la remise en suspension. Quant aux organes souterrains que constituent les rhizomes et les racines, ils stabilisent les fonds sédimentaires en limitant leur érodabilité (Christianen et al., 2013; Gracia et al., 2018).

Enfin, il convient de souligner que les 65 espèces de Magnoliophytes marines présentent des différences significatives en termes de taille, de structure et de productivité, ce qui influence directement leur capacité à fournir les services écosystémiques mentionnés précédemment. Par exemple, les espèces appartenant aux genres les plus grands (comme *Posidonia* et *Enhalus*) semblent offrir une plus large gamme de services que les espèces des genres plus petits (comme *Halophila* et *Lepilaena*) (Figure 1.6). Cette variabilité fonctionnelle souligne l'importance d'évaluer les services écosystémiques effectivement fournis par chaque espèce dans des contextes géographiques spécifiques. Il ne serait pas pertinent de généraliser les bénéfices écologiques estimés à l'ensemble des Magnoliophytes marines, sans tenir compte des différences locales ou des caractéristiques propres à chaque espèce (Mtwana Nordlund et al., 2016). Une approche régionale et spécifique est donc essentielle pour optimiser les efforts de conservation et de gestion de ces écosystèmes critiques.

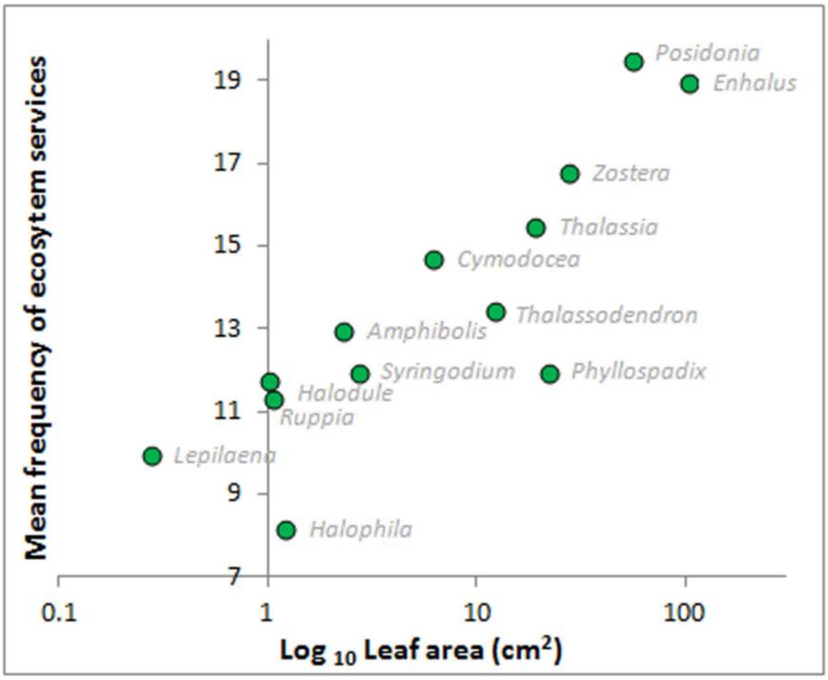


Figure 1.6. *Relation entre la fréquence moyenne perçue des services écosystémiques et la surface foliaire des différents genres de Magnoliophytes marines (Mtwana Nordlund et al., 2016).*

b. Les herbiers de Posidonia oceanica

Posidonia oceanica (L.) Delile (Figure 1.7) est une Magnoliophyte marine endémique de la mer Méditerranée, couvrant environ 1 à 2 % de sa surface totale (Pasqualini et al., 1998). Les herbiers marins qu'elle forme sont plus abondants dans la partie occidentale de la Méditerranée que dans la partie orientale, où ils ne semblent pas être présents le long des côtes du Liban, d'Israël et de la Syrie (Telesca et al., 2015). La superficie totale connue couverte par les herbiers de *P. oceanica* est estimée à 1 224 707 hectares, répartis entre 713 992 hectares dans le bassin occidental et 510 715 hectares dans le bassin oriental (Figure 1.8) (Telesca et al., 2015).

Figure 1.7. *Herbier de P. oceanica (©Fabrice Dudenhofer).*

Cette espèce, caractéristique de l'étage infralittoral, se développe entre 0,5 m et 40 m de profondeur dans des eaux de faible turbidité (Gobert et al., 2006). Espèce sténohaline, elle est majoritairement cantonnée à des eaux d'une salinité de 36,5 à 39,5 psu bien que certaines populations isolées aient été observées dans des eaux plus douces (21,5 psu dans la mer de Marmara) et plus salées (40 psu à l'est du bassin méditerranéen) (Gobert et al., 2006 ; Telesca et al., 2015). Ces variations suggèrent l'existence de génotypes locaux adaptés à des conditions environnementales spécifiques (Telesca et al., 2015). Les herbiers de *P. oceanica* sont absents aux abords de l'embouchure des fleuves (ex. Rhône, Pô, Nil) en raison de l'apport important d'eau douce et de fines particules en suspension, qui accroissent la turbidité (Gobert et al., 2006). En ce qui concerne la température, *P. oceanica* tolère une gamme comprise entre 9 et 29 °C (Boudouresque & Meinesz, 1982). Elle préfère les substrats sableux avec des sédiments grossiers, mais peut également coloniser les fonds rocheux (Gobert et al., 2006).

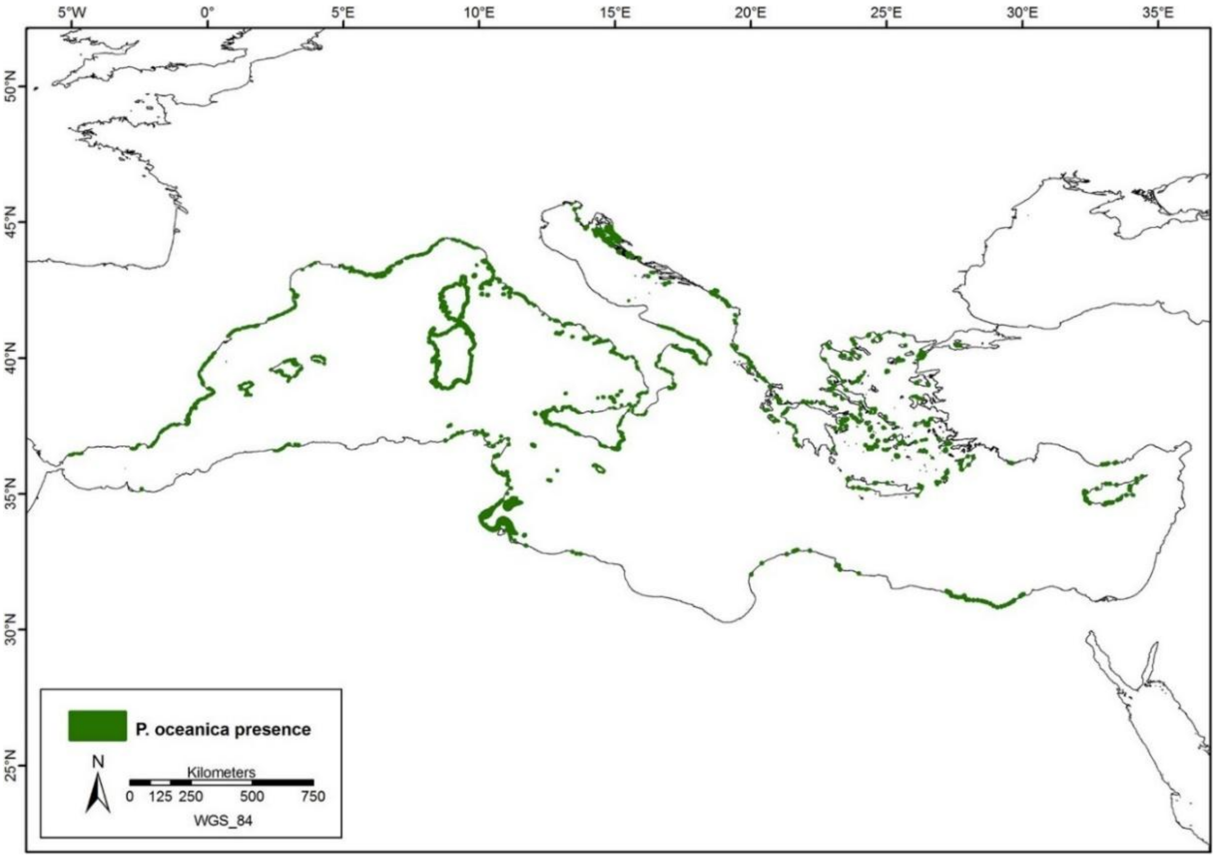


Figure 1.8. *Distribution actuelle des herbiers de P. oceanica (Telesca et al., 2015).*

P. oceanica est une espèce climacique, s'installant à la fin d'un long processus de succession écologique (Den Hartog, 1977). Avant son établissement, des espèces pionnières (comme *Caulerpa prolifera* ou *Cymodocea nodosa*), avec une croissance plus rapide et un cycle de vie plus court que *P. oceanica*, modifient progressivement les caractéristiques physico-chimiques des sédiments, notamment par un enrichissement en matière organique, créant ainsi un environnement favorable à la colonisation par *P. oceanica*. Au fil du temps, le développement et la croissance de l'herbier réduisent progressivement la luminosité ambiante, ce qui entraine la disparition des espèces pionnières et conduit à la formation d'un herbier monospécifique (Boudouresque & Meinesz, 1982).

c. Biologie de P. oceanica

Comme toutes les Magnoliophytes marines, *P. oceanica* présente une morphologie composée de trois organes : les faisceaux foliaires, le rhizome et les racines (Figure 1.9). Les faisceaux foliaires portés par les rhizomes sont composés de cinq à huit feuilles d'âges et de tailles différentes (juvéniles, intermédiaires et adultes) (Boudouresque & Meinesz, 1982 ; Giraud, 1979). Les feuilles adultes sont constituées d'un limbe (photosynthétique), d'une ligule et d'un pétiole, attaché au rhizome. La production foliaire est continue tout au long de l'année, avec une production maximale en fin d'été (Pergent & Pergent-Martini, 1991). À l'automne, le limbe tombe, tandis que le pétiole persiste et forme des écailles visibles à la base des faisceaux foliaires (Figure 1.9) (Giraud, 1979 ; Boudouresque et al. 2012). Le rhizome, qui soutient les faisceaux foliaires, peut présenter deux types de croissance. Le rhizome orthotrope correspond à une croissance verticale qui permet d'éviter l'enfouissement par l'accumulation sédimentaire tandis que le rhizome plagiotrope correspond à une croissance horizontale et favorise la colonisation de nouvelles zones (Figure 1.9) (Gobert et al., 2006). L'accumulation de sédiments et l'entrelacement des rhizomes, racines et matière organique donnent naissance à une structure tridimensionnelle appelée « matte », qui joue un rôle clé dans la stabilité physique des herbiers de *P. oceanica* (Gobert et al., 2006 ; Vacchi et al., 2017).

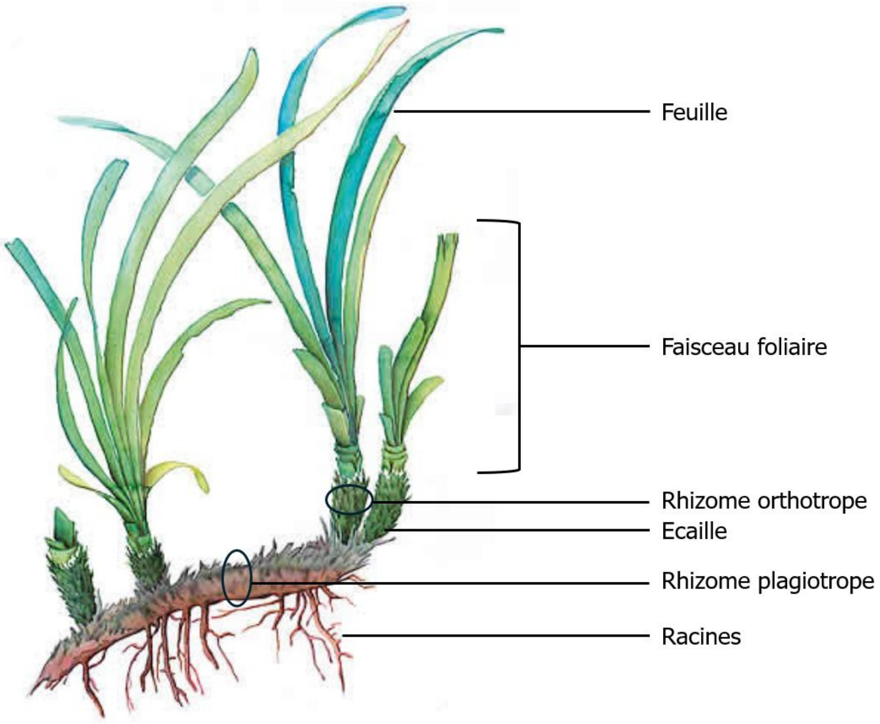


Figure 1.9. *Morphologie de P. oceanica (modifié d'après Abadie & Gobert, 2008).*

P. oceanica se reproduit de manière asexuée, par propagation végétative des rhizomes souterrains, et de manière sexuée, par germination des graines. La reproduction végétative joue un rôle fondamental dans la stabilité et la résilience des herbiers, tandis que la reproduction sexuée favorise la dispersion à longue distance et le brassage génétique (Buia et al., 1992). La floraison se produit à l'automne, généralement en septembre-octobre dans les zones peu profondes et en novembre dans les zones plus profondes (Balestri & Cinelli, 2003). *P. oceanica* est une espèce monoïque, portant des fleurs hermaphrodites regroupées en inflorescences terminales de trois à quatre fleurs (Figure 1.10). La maturation des fruits dure 6 à 9 mois. Le fruit, contenant une seule graine, ressemble à une olive vert foncé. Bien que le fruit ait une flottabilité positive, la graine a une flottabilité négative et est dépourvue de dormance, germant en quelques jours après la libération du fruit (Den Hartog, 1970; Boudouresque & Meinesz, 1982 ; Caye & Meinesz, 1984). Les plantes produisent en général deux fruits, rarement quatre, malgré la production de 4 à 10 fleurs par inflorescence. La flottaison des fruits facilite leur dispersion par les courants et réduit la compétition avec les plantes parentes. L'abondance des fleurs semble liée à la structure de l'herbier, avec une densité maximale de fleurs se trouvant généralement dans les peuplements les plus denses, favorisant ainsi la pollinisation entre les tiges florales appartenant probablement à la même plante parente (i.e. autopollinisation) (Buia & Mazzella, 1991). La floraison ne se produit pas tous les ans et semble être influencée par des facteurs environnementaux tels que la température de l'eau. Les changements de température au cours de l'année semblent jouer un rôle important dans la régulation du cycle de reproduction de *P. oceanica*. Les variations observées dans les périodes de floraison entre les herbiers à différentes profondeurs sont attribuées à des facteurs tels que la thermocline estivale, qui influence également la croissance des feuilles (Buia & Mazzella, 1991 ; Mazzella & Ott, 1984). Certaines années exceptionnelles ont vu des floraisons massives à l'échelle de la Méditerranée : 1971, 1982, 1993, 1997, 2003 et 2022 (Balestri, 2004 ; Diaz-Almela et al., 2007 ; Gobert et al., 2005). Toutefois, la faible fréquence de la floraison, la production limitée de graines, ainsi que l'autopollinisation, combinées à la dominance de la reproduction végétative, entrainent une diversité génétique réduite qui pourrait constituer un facteur de vulnérabilité pour l'espèce (Raniello & Procaccini, 2002).

Figure 1.10. *Inflorescence de P. oceanica.*

d. Phénologie de l’herbier de P. oceanica et cycle des nutriments

Les herbiers de *P. oceanica* présentent une diversité de paysages, allant de formations continues à des patchs isolés, voire à des rangées linéaires (Abadie et al., 2018 ; Borg et al., 2005 ; Molinier & Picard, 1952). Leur densité varie considérablement, allant de 150 à 300 faisceaux/m² dans les formations clairsemées, et pouvant dépasser 700 faisceaux/m² dans les herbiers denses (Giraud, 1977 ; Gobert et al., 2006). Le cycle saisonnier des herbiers est fortement influencé par des facteurs environnementaux, en particulier la lumière et la température de l'eau (Marbà et al., 1996). La disponibilité des nutriments constitue un facteur limitant local et saisonnier : la croissance est généralement limitée par les nutriments dans les zones où les eaux interstitielles et la colonne d'eau s’appauvrissent en nutriments durant l’été (Alcoverro et al., 1995). Les éléments nutritifs jouent un rôle essentiel dans la croissance et la survie de *P. oceanica*. Les macroéléments tels que l'azote (N), le phosphore (P), le soufre (S), le potassium (K), le calcium (Ca) et le magnésium (Mg) sont nécessaires en grande quantité et représentent une part significative du poids sec de la plante (Gobert, 2002). Les microéléments tels que le fer (Fe), le manganèse (Mn), le zinc (Zn), le cuivre (Cu), le bore (B), le chlore (Cl), le molybdène (Mo) jouent également un rôle crucial, bien que dans des quantités bien plus petites (Gobert, 2002). Ces éléments sont absorbés par les racines depuis les sédiments et par les feuilles depuis la colonne d’eau (Lepoint et al., 2002).

L'azote, en particulier, est essentiel pour la croissance de *P. oceanica*, mais sa disponibilité peut être limitante. En effet, les formes d'azote inorganique disponibles sont souvent insuffisantes pour soutenir pleinement la croissance des magnoliophytes marines (Touchette & Burkholder, 2000). Pour pallier cette limitation, *P. oceanica* a développé plusieurs mécanismes adaptatifs, tels que la fixation d'azote atmosphérique (N_2) par des bactéries diazotrophes associées à leurs racines (Garcias-Bonet et al., 2012, 2016 ; Mohr et al., 2021). L'exportation des feuilles vers d'autres écosystèmes entraine une perte d'éléments nutritifs (Gobert, 2002). Toutefois, la remobilisation des nutriments à partir des feuilles âgées avant leur abscission et leur translocation vers les tissus en croissance permet de compenser partiellement ces pertes (Patriquin, 1972 ; Lepoint et al., 2002). Alors que de nombreuses magnoliophytes marines prospèrent dans des environnements pauvres en nutriments, les stratégies de conservation des nutriments ne sont pas toujours très développées (Hemminga et al., 1999). Chez *P. oceanica*, la réutilisation interne des nutriments peut contribuer jusqu'à 40 % des besoins annuels en azote, une valeur relativement élevée par rapport à d'autres magnoliophytes marines (Lepoint et al., 2002). Cependant, cette réutilisation ne compense pas totalement la perte d'azote liée à la chute des feuilles en automne. Ainsi, le cycle de réutilisation interne des nutriments joue un rôle dans l'adaptation saisonnière de l'herbier et sa capacité à faire face aux fluctuations locales en nutriments. La disponibilité limitée des nutriments des eaux côtières contribue à façonner la phénologie et la dynamique de croissance des herbiers de *P. oceanica*.

e. Services écosystémiques et importance économique des herbiers de P. oceanica

Les herbiers de *P. oceanica* sont souvent comparés, à juste titre, aux forêts terrestres en raison de leur rôle essentiel dans le maintien des équilibres écologiques et leur importance pour les activités économiques associées (Boudouresque et al., 2006). L'une des caractéristiques majeures de *P. oceanica* est sa forte productivité primaire, qui contribue à l'oxygénation et à la structuration des écosystèmes côtiers. Chaque jour, un mètre carré d'herbier produit jusqu'à 14 litres d'oxygène (Bay, 1978). L'importante production de matière végétale permet de soutenir tout un ensemble de chaines trophiques et joue un rôle clé dans la biodiversité marine méditerranéenne et les ressources halieutiques (Campagne et al., 2014 ; Pergent-Martini et al., 1994). Les herbiers de *P. oceanica* offrent un habitat essentiel pour de nombreuses espèces marines. D'une part, ils servent de refuges et de nurseries pour les juvéniles de nombreuses espèces de poissons et d'invertébrés (Cheminée et al., 2021 ; Diaz-Gil et al., 2019). L'importance économique des herbiers se traduit notamment par leur contribution à la pêche méditerranéenne. Par exemple, en 2010, la criée de Sète a récolté plus de 4 000 tonnes de poissons sur les côtes françaises de la Méditerranée (hors Corse), pour une valeur totale de 14,7 millions d'euros. Les herbiers de *P. oceanica*, couvrant 27 220 hectares dans cette zone de pêche, abritent 16 espèces de poissons et d'invertébrés directement associées à cet écosystème. La contribution des herbiers à la production de ressources halieutiques a été estimée à environ 35 € par hectare et par an (Campagne et al., 2014). Bien que la proportion de la production primaire des herbiers soit abondante, seule une faible part est directement consommée par les herbivores. En effet, une grande partie est stockée au sein de la matte ou dégradée par les détritivores au sein de la couche de litière de l'herbier. De plus, une partie de cette production est également exportée vers d'autres écosystèmes sous forme de feuilles mortes (Pergent et al., 1994). Cette exportation de quantités considérables de feuilles mortes représente une opportunité pour les zones plus profondes bénéficiant d'une faible productivité primaire (Boudouresque et al., 2016 ; Wolff, 1976). De plus, les plages reçoivent également d'importantes quantités de feuilles mortes qui participent à la dynamique sédimentaire et contribuent à la stabilité des écosystèmes côtiers (Boudouresque et al., 2016 ; Mateo et al., 2003).

Parmi les 65 espèces de magnoliophytes marines, *P. oceanica* se distingue comme l'une des plus efficaces en matière de stockage du carbone, jouant ainsi un rôle significatif dans la lutte contre le changement climatique (Pergent et al., 1994 ; Mateo et al., 2006 ; Pergent et al., 2014 ; Jamaludin, 2015). Durant sa croissance, *P. oceanica* fixe une quantité importante de dioxyde de carbone (CO_2) atmosphérique à travers la production de ses différents organes, estimée à 1 302 tonnes de carbone par hectare par an en moyenne (Pergent-Martini et al., 2021). La fixation par les feuilles est estimée à 1 024 t C/ha/an, celle des écailles foliaires à 220 t C/ha/an et celle des rhizomes à 58 t C/ha/an. Le taux de séquestration des écailles foliaires et des rhizomes morts est estimé à 278 t C/ha/an. Bien que cette fixation annuelle de carbone ne représente en moyenne que 0,61 % des émissions de CO_2 des pays méditerranéens, son impact local est bien marqué. Elle atteint 3,1 % des émissions pour les grandes iles méditerranéennes et peut atteindre jusqu'à 14,4 % pour la Corse (Pergent-Martini et al., 2021). Au-delà de la fixation annuelle, la principale contribution des herbiers de *P. oceanica* au stockage du carbone réside dans leur matte. Celle-ci permet de piéger le carbone organique et inorganique sur des échelles de temps allant de plusieurs siècles à des millénaires (Romero et al., 1994 ; Tomasello et al., 2009; Boudouresque et al., 2016; Monnier et al., 2020). Le stock moyen de carbone trouvé dans la matte, résultant de l'accumulation de 21 % de la production primaire annuelle, varie entre 40 et 237 kg de carbone par mètre carré

(Serrano et al., 2014). Ces valeurs sont comparables, voire supérieures, à celles mesurées dans certains écosystèmes terrestres réputés pour leur capacité de stockage du carbone, tels que les tourbières (120 kg C/m²; Warner et al., 1993) ou les zones humides (13 à 73 kg C/m²; Laffoley & Grimsditch, 2009). Le stockage à long terme du carbone par les herbiers de *P. oceanica* revêt une importance cruciale dans le contexte du changement climatique mondial (Romero et al., 1994; Tomasello et al., 2009 ; Pergent-Martini et al., 2021).

Outre sa capacité exceptionnelle à stocker le carbone, *P. oceanica* joue un rôle crucial dans la protection des zones côtières contre l'érosion. Son système racinaire dense stabilise les sédiments et empêche leur remise en suspension (Gacia et al., 1999 ; Gacia & Duarte, 2001). Sa canopée sous-marine réduit l'impact de l'hydrodynamisme, limitant l'érosion des fonds marins. La chute des feuilles de *P. oceanica* est continue tout au long de l'année, mais particulièrement marquée en automne et en hiver (Mateo & Romero, 1996 ; Gallmetzer et al., 2005). Ces feuilles mortes suivent plusieurs trajectoires : (1) elles s'accumulent au sein de l'herbier et au niveau des intermattes, formant la litière (Pergent et al., 1994) ; (2) Elles sont exportées vers les écosystèmes adjacents (plages, milieux profonds, zones humides) (Pergent et al., 1994). En automne, la chute importante des feuilles conjuguée à l'action du vent, au renforcement de l'hydrodynamisme et aux tempêtes d'équinoxe, conduit à la déposition de cette matière végétale morte sous forme de banquettes sur les plages (Boudouresque & Meinesz, 1982 ; Pergent et al., 1997). Les banquettes sont des accumulations de feuilles mortes de *P. oceanica* et de sédiments pouvant atteindre plusieurs mètres de hauteur (Picard, 1965 ; Vacchi et al., 2017). Ces banquettes ont un rôle essentiel dans la protection des plages contre l'érosion, notamment lors des tempêtes hivernales (Boudouresque & Meinesz, 1982 ; Chessa et al., 1995). Le retrait périodique, qui a lieu dans certaines municipalités pendant la saison estivale dans le contexte du "nettoyage" des plages, est fréquemment associé à un recul significatif du trait de côte (Pergent & Kempf, 1993 ; De Falco et al., 2008). De plus, les banquettes servent également de ressources alimentaires pour les invertébrés détritivores des écosystèmes dunaires et peuvent servir d'engrais et de support de la végétation menant au maintien et au développement de l'arrière-dune (Duarte, 2004).

En raison de ses exigences écologiques strictes et de sa sensibilité aux variations de température, de turbidité, et de pollution, *P. oceanica* est un bioindicateur clé pour évaluer l'état écologique des eaux côtières (Gobert et al., 2009 ; Montefalcone, 2009 ; Pergent-Martini et al., 2005). Elle permet de détecter différentes perturbations à différents niveaux. Au niveau de l'individu, la phénologie et la biométrie foliaire renseignent sur l'état de santé de la plante et sa croissance, alors que la structure de l'herbier (densité, taux de recouvrement, présence de matte morte) reflètent les conditions environnementales et les pressions anthropiques (Pergent et al., 1995 ; Marbà et al., 2006 ; Montefalcone et al., 2008 ; Gobert et al., 2009). L'herbier de *P. oceanica* peut également être étudié au niveau de la communauté, où la composition de la faune et de la flore associées, notamment les épiphytes foliaires, permet de détecter des altérations du milieu (Ruiz et al., 2001, Balata et al., 2007). Ainsi, les magnoliophytes marines, et particulièrement les herbiers de *P. oceanica*, sont utilisés dans le cadre de la Directive-Cadre sur l'Eau (DCE ; '*Water Framework Directive*' en anglais) de l'Union européenne pour l'évaluation de l'état écologique des eaux côtières en Méditerranée (Foden & Brazier, 2007 ; Gobert et al., 2009 ; Lopez y Royo et al., 2011).

La valorisation économique des services écosystémiques est un outil essentiel pour sensibiliser les décideurs et améliorer la gestion des territoires côtiers. Elle permet d'attribuer une valeur monétaire aux bénéfices fournis par les écosystèmes, facilitant ainsi leur intégration dans les stratégies de conservation et de gestion. Dans le cas des herbiers de *P. oceanica*, la valeur économique des services fournis est estimée entre 25,3 et 45,9 millions d'euros par an, soit 283 à 513 €/ha/an (Campagne et al., 2014). Cependant, la dégradation des herbiers de *P. oceanica* au cours du dernier siècle a entrainé une perte économique annuelle estimée entre 1,11 et 2 millions d'euros. De plus, cette destruction a également entrainé un déclin à long terme de certains services écosystémiques tels que la libération de carbone et de métaux lourds normalement séquestrés dans la matte (Campagne et al., 2014). L'absence de reconnaissance économique des bénéfices rendus par les écosystèmes et la biodiversité a souvent contribué à leur surexploitation et à leur dégradation (MEA, 2005 ; Secrétariat de la CDB, 2010). En réalité, dans de nombreux cas, les décideurs politiques ne peuvent pas prendre correctement en compte ce qui n'a pas de valeur économique, justifiant ainsi la nécessité d'une évaluation économique des écosystèmes (Costanza et al., 1997). Il est donc primordial d'identifier, mesurer et suivre le capital naturel et d'intégrer cette évaluation économique dans les politiques de gestion environnementale afin d'assurer une conservation efficace des herbiers de *P. oceanica* (Campagne et al., 2014).

4. Conservation et restauration des herbiers de *P. oceanica*

a. *Pressions anthropiques et régression*

P. oceanica est très sensible à la qualité de l'environnement, ce qui la rend particulièrement vulnérable à diverses pressions anthropiques telles que le changement climatique, l'eutrophisation, la propagation rapide d'espèces invasives et les impacts mécaniques (construction d'infrastructures côtières, ancrage, pêche au chalut) (Holon et al., 2015, 2018 ; Marbà et al., 2014). L'effet cumulatif de ces perturbations a entrainé une régression globale des herbiers de *P. oceanica* à l'échelle de la Méditerranée (Boudouresque et al., 2009). Les estimations des taux de régression varient selon les régions et les méthodes de mesure. En Espagne, on estime une perte annuelle d'environ 7 % de la surface des herbiers au cours des 50 dernières années (Marbà et al., 2014). D'autres régions présentent des déclins plus modérés, comme en Provence-Alpes-Côte d'Azur, où une diminution de 13% a été observée en 85 ans, malgré une forte pression anthropique côtière (Holon et al., 2015). Une estimation récente, basée sur des données cartographiques, suggère une réduction d'environ 34 % au cours des 50 dernières années à l'échelle de la Méditerranée, bien que cette valeur repose sur une portion limitée de la distribution des herbiers de *P. oceanica* et sur des méthodes de calcul variables (Telesca et al., 2015). Les différences dans ces estimations s'expliquent en partie par l'évolution des techniques de cartographie et de leur précision (Abadie et al., 2015; Bonacorsi et al., 2013 ; Boudouresque et al., 2021 ; Telesca et al., 2015).

Toutefois, la tendance générale reste une régression marquée, en particulier autour des principaux ports méditerranéens (Barcelone, Marseille, Gênes) et en mer de Ligure (Italie) (Boudouresque et al., 2021 ; Peirano et al., 2005 ; Telesca et al., 2015). La construction d'infrastructures côtières, notamment les ports, a un impact important sur les herbiers de *P. oceanica* (Holon et al., 2015 ; Meinesz et al., 1991). L'urbanisation rapide des zones côtières, due au développement touristique et à l'accroissement de la population, a entraîné une multiplication des infrastructures artificielles. Celles-ci provoquent un ensevelissement des herbiers lors des travaux de construction et modifient les conditions environnementales locales (changement de l'hydrodynamisme, augmentation de la sédimentation, pollution) (Ruiz & Romero, 2003). Parmi les impacts mécaniques subis par les herbiers de *P. oceanica*, l'ancrage des grands navires (longueur supérieure à 24 m) a montré une augmentation en raison du développement de la navigation de plaisance au cours des dernières décennies (Abadie et al., 2016 ; Deter et al., 2017 ; Montefalcone et al., 2008). Malgré la taille relativement faible de la mer Méditerranée, plus de la moitié de la flotte mondiale de grands navires de plaisance fréquentent les eaux méditerranéennes pendant au moins huit mois par an (Carreño et Lloret, 2021), principalement dans le bassin occidental (Côte d'Azur, Ligurie, sud de la Corse, nord de la Sardaigne) (Pergent-Martini et al., 2022). Concentré dans les eaux peu profondes, l'ancrage entraîne des dommages directs et indirects lors du déploiement et de la récupération de l'ancre, et du mouvement de la chaîne/corde attachée à l'ancre. L'ancrage à l'intérieur des herbiers de *P. oceanica* semble avoir divers degrés d'impact en fonction de sa densité, de sa fréquence, du type d'ancre, de la profondeur et de la taille des bateaux (Boudouresque et al., 2012). Ainsi, l'ancrage répété des navires de plaisance, à des profondeurs supérieures à 15 m, entraîne des dégradations à grande échelle des herbiers (Abadie et al., 2015). Cela peut entraîner des modifications chimiques du sédiment, notamment par l'intrusion de sulfures d'hydrogène (H_2S) (Abadie et al., 2016), un composé toxique pour *P. oceanica* (Calleja et al., 2007; Holmer et al., 2003; Marbà et al., 2006). Les zones de matte morte succédant à l'action de l'ancrage (Figure 1.11) deviennent des substrats favorables à l'installation et à la prolifération d'espèces invasives, comme *Caulerpa cylindracea* (Katsanevakis et al., 2010; Kiparissis et al., 2011). Cette macroalgue augmente les concentrations de sulfures d'hydrogène dans les sédiments, réduisant ainsi davantage encore le potentiel de recolonisation de la matte morte par *P. oceanica* (Garcias-Bonet et al., 2008; Holmer et al., 2009).

Figure 1.11. *Sillon de matte morte laissé par la chaine d'un navire dans un herbier de P. oceanica dans la baie de Calvi (Corse, France).*

La recolonisation naturelle des herbiers endommagés est généralement considérée comme impossible à l'échelle humaine en raison de la croissance extrêmement lente de *P. oceanica* (Marbà et al., 2002). Il est donc impératif d'adopter des mesures strictes de protection et de gestion des activités anthropiques, afin d'empêcher toute dégradation supplémentaire des herbiers de *P. oceanica* (Boudouresque et al., 2021 ; Holon et al., 2015 ; Telesca et al., 2015).

b. Historique de l'évolution des mesures de conservation

Face au constat de la régression des herbiers de *P. oceanica* provoquée par l'impact des activités humaines, les mesures de conservation et de gestion ont considérablement évolué ces dernières décennies. Aujourd'hui, *P. oceanica* figure parmi les principales priorités en matière de protection et de gestion de l'environnement marin en Méditerranée. (Boudouresque et al., 2012). Au niveau européen, la Directive Habitats (92/43/CEE) classe les herbiers de *P. oceanica* parmi les habitats prioritaires (type d'habitat 1120 : Herbiers de *Posidonia oceanica* - *Posidonion oceanicae*). De plus, la Convention de Barcelone leur consacre un plan d'action spécifique, renforçant leur statut de protection en Méditerranée. L'Union européenne a également mis en place un cadre juridique visant à concilier la préservation des écosystèmes tout en permettant un développement économique durable dans les zones côtières. Deux directives majeures encadrent cette gestion : (1) la Directive Cadre sur l'Eau (DCE, 2000/60/CE) qui impose l'atteinte du « bon état écologique » des eaux de surface de l'UE ; (2) la Directive Cadre Stratégie pour le Milieu Marin (DCSMM, 2008/56/CE) qui étend cette approche aux milieux marins en évaluant leur état écologique global. Pour atteindre cet objectif, il a d'abord été nécessaire de mettre en place plusieurs mesures, notamment définir le "bon état écologique", puis limiter les impacts et enfin évaluer l'état écologique des masses d'eau concernées. Chaque État membre a ainsi défini sa propre méthode d'évaluation de l'état écologique des herbiers de *P. oceanica* selon la DCE (2000/60/CE) (Gobert et al., 2009). Dans le cadre de la DCSMM, l'évaluation ne se limite plus uniquement aux caractéristiques fonctionnelles de la plante, mais intègre l'ensemble des composantes de l'écosystème qu'elle soutient (Figure 1.12). Cette approche a conduit au développement d'un indice de qualité écologique basé sur l'écosystème (*Ecosystem Based Quality Index*, EBQI), qui prend en compte l'état de conservation de chaque compartiment fonctionnel associé aux herbiers (Personnic et al., 2014).

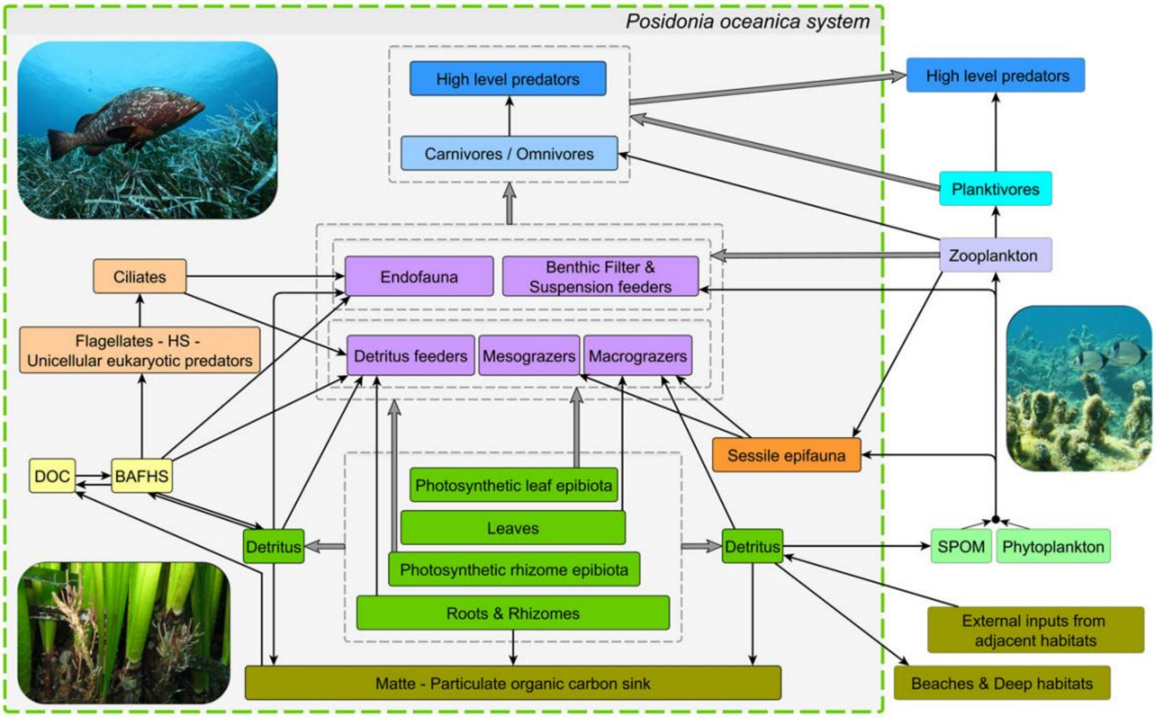


Figure 1.12. *Modèle conceptuel illustrant le fonctionnement de l'écosystème soutenu par les herbiers de P. oceanica, composé de différents compartiments fonctionnels (Giakoumi et al., 2015).*

En France, *P. oceanica* est reconnue comme une espèce protégée depuis plusieurs décennies (décret n° 19/07/1988), ce qui interdit tout prélèvement de la plante, qu'elle soit vivante ou morte, sans autorisation préalable. Par ailleurs, plusieurs pressions anthropiques ont fait l'objet de réglementations spécifiques, notamment à travers la politique des eaux usées de l'Agence française de l'eau et la loi Littoral de 1986 encadrant les constructions côtières (Bockel et al., 2023). Plus récemment, les autorités françaises ont adopté de nouvelles réglementations strictes pour réguler l'ancrage des navires dans les herbiers de *P. oceanica*. Depuis 2016, un décret de la Préfecture maritime française (décret n° 155/2016) interdit l'ancrage des navires de plus de 80 m de longueur dans les zones où des espèces végétales marines protégées sont présentes. Cette réglementation a été complétée en juin 2019 par un second décret (décret n° 123/2019) étendant cette interdiction aux navires de plus de 24 m de longueur. A la suite de l'application de ce dernier décret, le nombre de grands navires (>24 m) ancrant dans les herbiers de *P. oceanica* a considérablement diminué. Une étude récente a estimé que l'application rigoureuse de cette nouvelle réglementation, basée sur des cartographies d'habitats précises, a permis d'éviter la destruction d'herbiers séquestrant entre 134 et 217 tonnes de carbone en 2022 (Bockel et al., 2023). Toutefois, ces réglementations demeurent rares à l'échelle de la Méditerranée, malgré quelques exceptions notables en Croatie et en Espagne (Pergent-Martini et al., 2021b). Aux îles Baléares, par exemple, un décret adopté en 2018 (Bulletin officiel des îles Baléares, décret n° 25/2018 du 28 juillet 2018) interdit strictement l'ancrage des bateaux de toute taille sur les herbiers de *P. oceanica*. Seul l'amarrage sur des systèmes de bouées respectueux de l'environnement à faible impact sur le fond marin y est autorisé.

En parallèle des réglementations, des programmes de surveillance ont été mis en place pour suivre l'état écologique des herbiers. Parmi eux, le Réseau de Surveillance Posidonie (RSP), actif depuis 1984, et le réseau de suivi des herbiers de *P. oceanica* par télémétrie acoustique (TEMPO), depuis 2008, permettent d'évaluer l'état de santé des herbiers de *P. oceanica* et leur rôle en tant qu'indicateur de la qualité environnementale (Holon et al., 2013 ; Pergent et al., 2015). Les coûts annuels de surveillance et de protection des herbiers de *P. oceanica* en France sont estimés à 4,8 millions d'euros, soit environ 0,11 à 0,23 % de la valeur totale des services écosystémiques qu'ils fournissent. Cependant, malgré ces efforts de conservation, la destruction continue de ces habitats engendre des pertes économiques annuelles comprises entre 1,11 et 2,00 millions d'euros, sans compter la perte irréversible de certains services écosystémiques à long terme (Campagne et al., 2014). L'ensemble de ces réglementations et programmes de suivi témoigne de la prise de conscience croissante de l'importance écologique et économique des herbiers de *P. oceanica*, ainsi que des efforts déployés pour leur conservation et restauration.

c. Restauration des herbiers de P. oceanica : rétrospective sur 35 ans

Une fois que les facteurs de destruction des herbiers sont éliminés, leur recolonisation naturelle reste un processus extrêmement lent, en raison de la lente croissance de *P. oceanica* (Boudouresque et al., 2006). En effet, la croissance moyenne d'un rhizome plagiotrope, croissance horizontale du rhizome permettant la colonisation, est de 6 cm/an (Molenaar et al., 2000). De plus, l'arrêt d'une pression anthropique ne garantit pas toujours un démarrage immédiat du processus de recolonisation. Par exemple, trois ans après l'arrêt des activités d'une ferme aquacole à Minorque, la régression des herbiers de *P. oceanica* persistait (Delgado et al., 1999). Cependant, certaines améliorations ont été observées dans des contextes où la qualité du milieu a été restaurée : dans la région Provence-Alpes-Côte d'Azur, le RSP a documenté une progression de certaines limites d'herbiers, coïncidant avec l'amélioration de la qualité de l'eau suite au traitement des eaux usées par les stations d'épuration (Boudouresque et al., 2000). Etant donné que la régénération naturelle des herbiers est extrêmement lente, il peut être nécessaire d'accélérer cette dynamique à travers des projets de restauration écologique. De nombreuses initiatives ont ainsi vu le jour au cours des trente dernières années (Boudouresque et al., 2021 ; Pansini et al., 2022).

Depuis la première expérimentation documentée en 1989 (Pansini et al., 2022), le nombre d'études sur la restauration des herbiers de *P. oceanica* a augmenté de manière croissante, atteignant son maximum en 2019 avec 13 travaux publiés (Pansini et al., 2022). Toutefois, la répartition des efforts de recherche et des actions de restauration varie selon les pays méditerranéens. La France, très active lors des premières décennies, a cessé de jouer un rôle majeur après 2007. En revanche, l'Espagne a nettement intensifié ses efforts ces cinq dernières années, passant de 2 à 9 publications en 2020, tandis que l'Italie reste le pays ayant produit le plus grand nombre d'études sur le sujet (Figure 1.13) (Pansini et al., 2022). D'un point de vue géographique, 89 % des interventions de restauration ont été réalisées en Méditerranée occidentale (Figure 1.14) (Pansini et al., 2022). Parmi les études recensées, 68 % avaient un objectif expérimental visant à évaluer les performances de boutures de *P. oceanica* transplantées face à un facteur environnemental ou à tester des techniques spécifiques de transplantation (Figure 1.14). Les 32 % restants concernaient des opérations de restauration visant à compenser la perte d'habitat dans un site donné (Figure 1.14). Les surfaces concernées par ces interventions de transplantation variaient de 1 m² à 2 ha, les échelles micro (<10 m²) et méso (10 - 10 000 m²) étaient plus représentées que l'échelle macro (>10 000 m²) dans l'ensemble des études de cas (Figure 1.14) (Pansini et al., 2022).

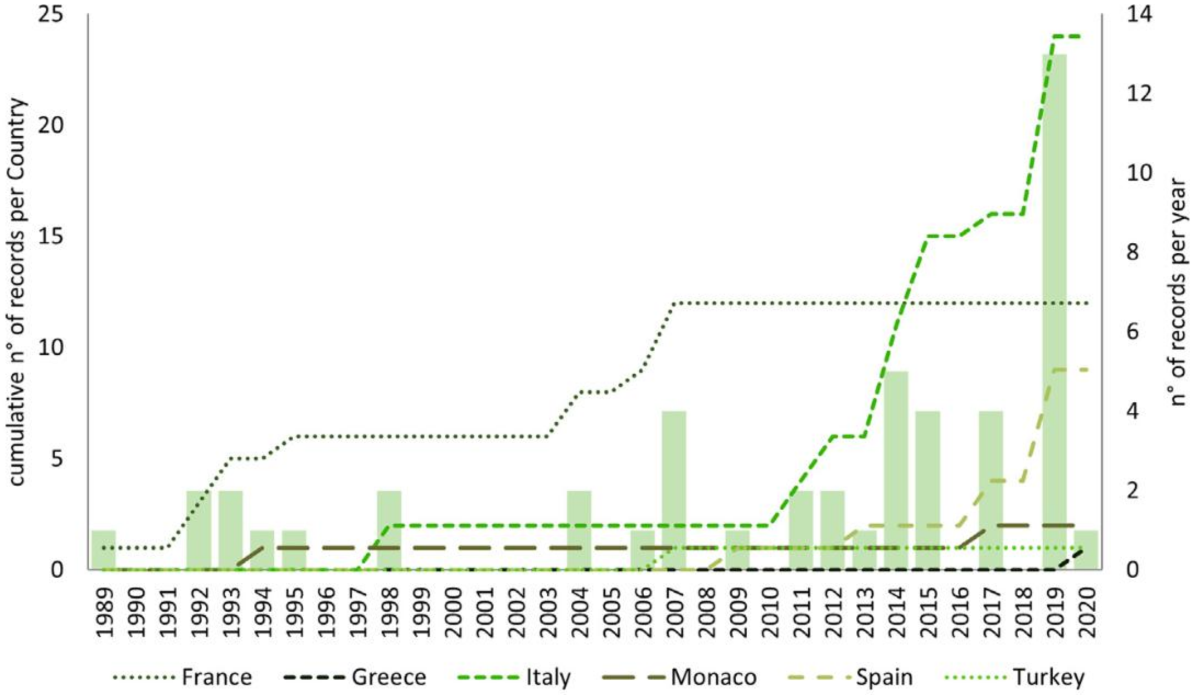


Figure 1.13. *Évolution temporelle des actions de restauration de herbiers de P. oceanica de 1989 à 2020. Les lignes représentent le nombre cumulatif d'interventions par année et par pays (Pansini et al., 2022).*

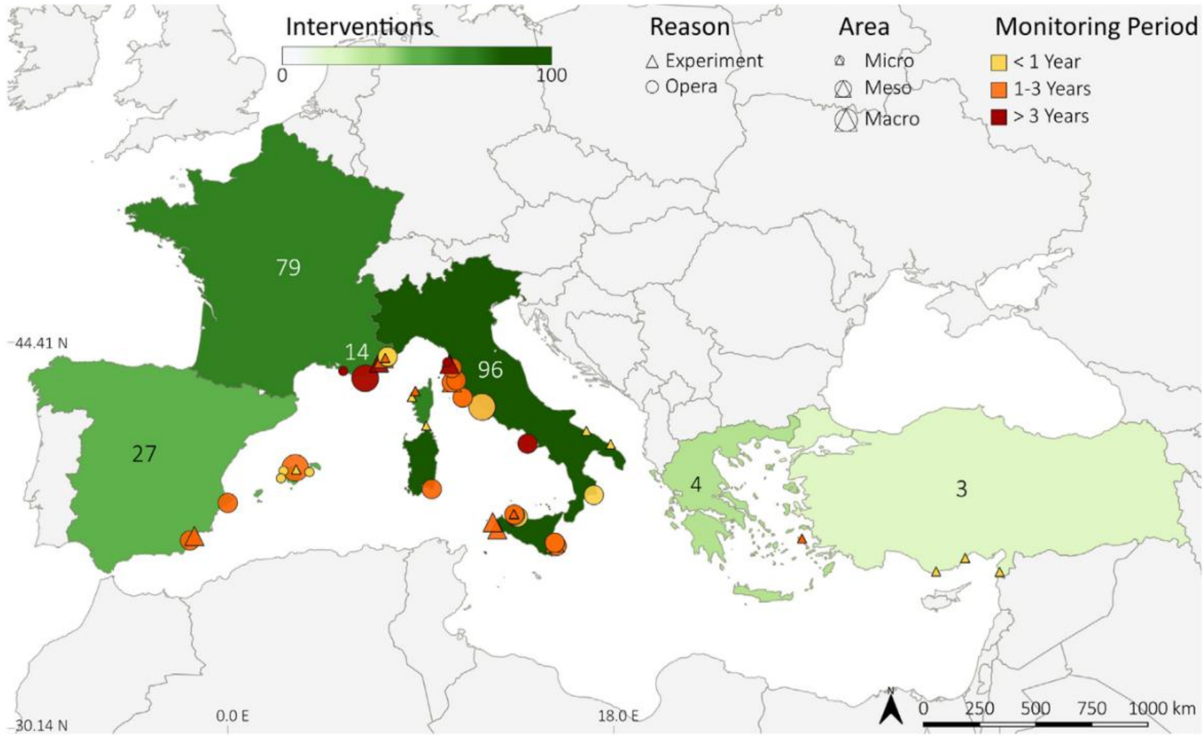


Figure 1.14. *Distribution géographique des opérations de restauration des herbiers de P. oceanica, classées selon le type d'opération, la taille de l'opération et la durée de suivi (Pansini et al., 2022).*

Une augmentation générale du nombre d'études et de publications sur les actions de transplantation de *P. oceanica* a été observée ces dernières années, reflétant l'intérêt croissant porté à la conservation et à la restauration du milieu marin. Toutefois, la majorité des interventions menées restent à petite échelle, souvent dans un cadre expérimental, plutôt que de véritables opérations de restauration à grande échelle. Ces essais expérimentaux avaient pour objectif de tester différentes techniques de transplantation, en variant les substrats, les matériaux d'ancrage, les densités de plantation et les profondeurs. Cette approche expérimentale est cohérente avec le fait que la restauration des herbiers marins est une discipline encore récente et en plein développement (Wood et al., 2019). Elle s'inscrit dans le cadre de nombreux accords internationaux visant à stopper le déclin des espèces et habitats prioritaires (Convention on Biological Diversity, UNEP, 2021 ; UN Decade on Ecological Restoration 2021–2030, United Nations Environment Agency, 2019). Ainsi, ces essais pilotes de recherche et développement sont essentiels pour combler les lacunes méthodologiques avant d'envisager des projets à plus grande échelle (Pansini et al., 2022). Cependant, la restauration des herbiers de *P. oceanica* ne peut être envisagée isolément, elle doit s'intégrer dans une stratégie de gestion globale à l'échelle d'une baie ou d'une région (Boudouresque et al., 2006 ; 2021). Cette stratégie doit prendre en compte plusieurs éléments (Figure 1.15) : (i) la priorité doit être donnée au potentiel de recolonisation naturelle, la restauration active ne doit être envisagée que si la régénération naturelle est insuffisante; (ii) le coût de la restauration par rapport à un investissement équivalent dans l'amélioration des conditions environnementales (zones de mouillages écologiques, traitement des eaux usées, interdiction du chalutage et de l'ancrage, etc.) ; et (iii) l'impératif de ne replanter que dans des zones historiquement colonisées par l'herbier de *P. oceanica* (Figure 1.15) (Boudouresque et al., 2021).

Enfin, il est fondamental de rappeler que la capacité technique de restaurer les herbiers de *P. oceanica* ne doit en aucun cas être utilisé comme mesure compensatoire pour servir d'alibi à de nouvelles destructions (Boudouresque et al., 2006 ; 2021). Cependant, dans certains cas exceptionnels, lorsque des décisions politiques entrainent la destruction inévitable d'herbiers, par exemple lors de l'expansion du port de Piombino (Ligurie) ou du projet d'urbanisation en mer dans l'Anse du Portier à Monaco, une opération de transplantation afin d'empêcher la destruction totale de l'herbier peut être envisagée pour limiter les pertes écologiques (Bedini et al., 2020 ; Descamp et al., 2017, 2025).

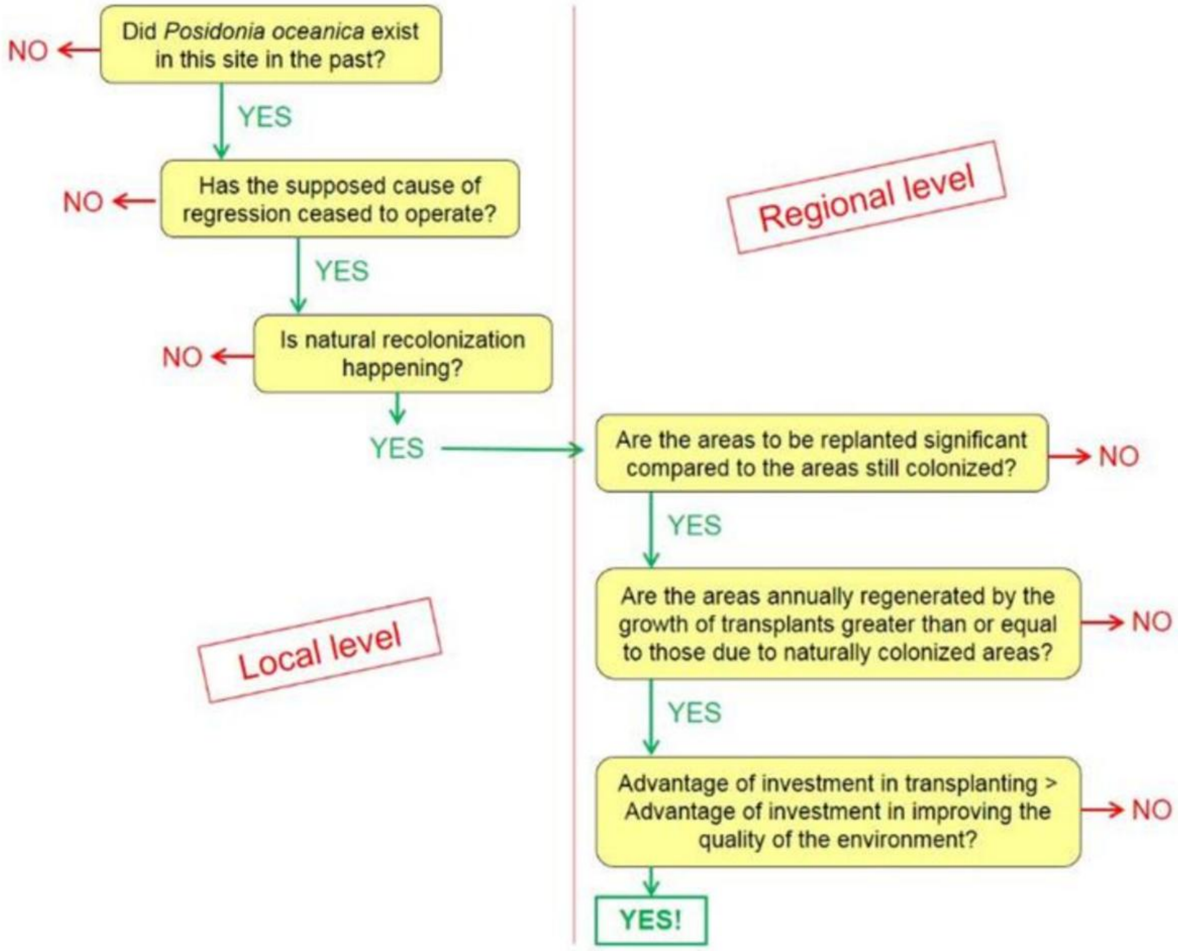


Figure 1.15. *Stratégie de prise de décision pour la transplantation de P. oceanica (Boudouresque et al., 2021).*

Les efforts de restauration des herbiers de *P. oceanica* reposent sur diverses techniques qui peuvent être classées en trois grandes catégories selon l'origine biologique du matériel utilisé pour la transplantation :, (i) des parcelles ou mottes d'herbiers comprenant le sédiment et la matte sous-jacente (Figure 1.16), (ii) des graines (Figure 1.17) ou (iii) des boutures individuelles (Figure 1.18).

La transplantation de parcelles, ou mottes, entières d'herbiers avec le sédiment et la matte sous-jacente a principalement été mise en œuvre dans des projets de grande envergure (plusieurs centaines de m²) mis en place comme mesures compensatoires suite à la construction d'infrastructures côtières (Bedini et al., 2020 ; Descamp et al., 2017, 2025; Sánchez-Lizaso et al., 2009). En 2017, à la demande du gouvernement monégasque, le protocole SafeBent a été développé pour déplacer environ 500 m² d'herbiers menacés par un projet d'aménagement côtier. Des parcelles d'herbiers (*P. oceanica* + sédiment et matte sous-jacent) 0,8 m²/60 cm d'épaisseur ont été récoltées à l'aide d'une grue sur une plateforme de forage. Ces parcelles ont été ensuite placées dans des paniers métalliques recouverts d'un tapis en fibres végétales (fibres de coco) et replantées dans des trous préalablement creusés dans un fond sableux (Figure 1.16). Cette méthode permet un taux de transplantation élevé, pouvant atteindre jusqu'à 32 m² par jour (Descamp et al., 2017, 2025). Une opération similaire a été réalisée lors de l'extension du port de Piombino (Ligurie), mais avec une variante : Les parcelles d'herbiers ont été déposées directement sur une intermatte sableuse naturelle, sans creusement préalable (Bedini et al., 2020). Les principaux avantages de cette technique résident dans sa capacité à permettre la transplantation simultanée de milliers de fragments de *P. oceanica*, tout en préservant la structure de l'herbier ainsi que la faune invertébrée et le microbiome associés à la matte et au sédiment (Bedini et al., 2020 ; Descamp et al., 2017, 2025).

Figure 1.16. *Transplanteur utilisé dans le protocole SafeBent pour la transplantation de parcelles d'herbier de P. oceanica (Descamp et al., 2017).*

Une autre alternative repose sur l'utilisation de graines de *P. oceanica* (Figure 1.17) comme matériel biologique pour des opérations de restauration par transplantation. Cette approche présente l'avantage d'être non-destructrice pour les herbiers existants, notamment lorsque les graines sont récoltées flottant en mer ou échouées sur les plages (Boudouresque et al., 2021). Plutôt que de transplanter directement des graines, certaines études ont d'abord fait germer les graines en aquarium avant de les transplanter sous forme de plantules, permettant ainsi un meilleur contrôle des conditions initiales et une augmentation des chances de survie (Escandell-Westcott et al., 2023 ; Piazzi et al., 2000). Bien que l'herbier se reproduise principalement par reproduction végétative (stolonisation ou dispersion de boutures par le courant), la dispersion des graines et leur recrutement jouent un rôle important dans le maintien de la diversité génétique des herbiers (Montefalcone et al., 2013). Cependant, de nombreuses opportunités de germination et de recrutement sont perdues en raison de divers facteurs : (i) l'avortement des fruits, près de 10 % des fruits flottants sont trop petits et non viables (Belzunce et al., 2005), (ii) la libération des graines sur des substrats inadaptés, empêchant la germination (plages ou zones trop profondes) et (iii) la prédation par des herbivores, en particulier la saupe *Sarpa salpa*, pouvant endommager jusqu'à 58 % des graines collectées (Belzunce et al., 2005). Le principal obstacle à l'utilisation des graines dans le cadre de projets de restauration reste l'imprévisibilité de la floraison, qui varie fortement selon les régions et ne donne lieu à des épisodes massifs (i.e. impliquant la plupart des herbiers des différentes régions de la Méditerranée) qu'environ tous les 10 ans (Montefalcone et al., 2013). Cette faible disponibilité des graines explique le nombre limité de projets de restauration basés sur cette méthode et les lacunes scientifiques qui persistent à ce sujet (Escandell-Westcott et al., 2023).

Figure 1.17. *Plantules de P. oceanica un mois après germination en milieu naturel. Les graines ont été disposées à même la matte morte (© GIS Posidonie).*

Cependant, la majorité des actions de restauration des herbiers de *P. oceanica* ont été réalisées par transplantation de boutures. Certains travaux antérieurs ont validé des méthodologies permettant d'optimiser la survie et le développement des boutures transplantées. Parmi ceux-ci, les travaux de recherche de Heike Molenaar ont montré qu'une plantation des boutures en forte densité (i.e., 5-10 cm d'espacement) augmente les taux de survie des boutures sur le long terme (Molenaar & Meinesz, 1995). De plus, la transplantation lors des phases métaboliques les plus actives de *P. oceanica* (i.e., printemps) de rhizomes plagiotropes avec un minimum de trois faisceaux et d'un rhizome d'une longueur supérieure à 10 cm permet d'obtenir les meilleurs taux de survie (Molenaar, 1992 ; Molenaar et al., 1993 ; Molenaar & Meinesz, 1995). Concernant la méthode de transplantation, les boutures peuvent être fixées au substrat à l'aide d'une large variété de techniques d'ancrage (e.g., Figure 1.18), pour lesquels il ne semble pas y avoir de consensus (Pansini et al., 2022). Dans les expérimentations conduites jusqu'à ce jour, les méthodes d'ancrage individuel des boutures impliquaient l'utilisation de crochets, de piquets, d'agrafes métalliques tandis que les techniques d'ancrage modulaires nécessitaient l'utilisation de grillages plastiques ou métalliques, de treillis en fibres végétales, de gabions ou de structures en béton (Alagna et al., 2019 ; Boudouresque, 2001 ; Boudouresque et al., 2006 ; 2021 ; Cooper, 1982 ; Genot et al., 1994 ; Gobert et al., 2005 ; Meinesz et al., 1992; Molenaar, 1992; Molenaar et al., 1993; Molenaar & Meinesz, 1995 ; Pansini et al., 2022). Les méthodes de transplantation basées sur l'utilisation de grilles (plastiques, métalliques ou en fibres naturelles) ainsi que les méthodes d'ancrage individuel (agrafe ou piquet) ont généralement donné de bons résultats en termes de transplantation (Calvo et al., 2021 ; Genot et al., 1994 ; Mancini et al., 2021 ; Molenaar & Meinsez, 1995 ; Piazzi et al., 2021 ; Scannavino et al., 2014).

Figure 1.18. *Boutures de P. oceanica attachées sur une structure polymérique en amidon de pomme de terre (éléments BESE).*

Malgré les nombreuses expérimentations de transplantation de boutures de *P. oceanica* menées au cours des 35 dernières années, des lacunes importantes persistent (Boudouresque et al., 2021 ; Pansini et al., 2022 ; Pergent-Martini et al., 2024), notamment en ce qui concerne les méthodes de fixation des transplants (Pergent-Martini et al., 2024). La fixation des boutures sur le fond marin constitue en effet une étape déterminante pour assurer le succès de la transplantation et la persistance à long terme des herbiers transplantés. Plusieurs méthodes ont été testées et se sont révélées efficaces dans certaines conditions environnementales, mais seules certaines sont considérées comme écologiquement durables (Bacci & La Porta, 2021 ; Boudouresque et al., 2021).

Une autre limite majeure concerne la disponibilité du matériel biologique pour la transplantation. Les boutures de *P. oceanica* peuvent être obtenues par prélèvements destructifs dans un herbier naturel, ce qui engendre un impact direct sur les populations existantes. Une alternative consiste à collecter les fragments naturellement détachés des herbiers naturels par l'hydrodynamisme (appelés boutures-épaves) (Balestri et al., 2011; Gobert et al., 2005; Ward et al., 2020). Bien que ces fragments dérivants présentent une faible probabilité de réimplantation naturelle (Meinesz & Lefèvre, 1984), ils conservent la capacité de s'ancrer, se rétablir et former de nouveaux patchs par expansion clonale (Boudouresque & Meinesz, 1982; Boudouresque et al., 1990; Almela et al., 2008; Di Carlo et al., 2005). Leur utilisation présente ainsi un double avantage : une disponibilité abondante de matériel transplantable et un impact écologique nul sur les populations existantes, contrairement aux prélèvements destructifs (Balestri et al., 2011). Cependant, les performances (survie, croissance) des boutures-épaves, comparées à celles des boutures issues d'herbiers naturels, restent encore très peu documentées (Balestri et al., 2011 ; Pergent-Martini et al., 2024).

5. Objectifs et structure de la thèse

En février 2024, le Parlement européen a adopté la Loi sur la restauration de la nature, établissant des objectifs progressifs et contraignants pour les États membres de l'Union européenne : restaurer 30 % des habitats terrestres et marins en mauvais état d'ici 2030, 60 % d'ici 2040 et 90 % d'ici 2050. Parmi les écosystèmes marins concernés, la majorité de la communauté scientifique s'accorde sur la nécessité et la légitimité de restaurer les vastes zones d'herbiers de *P. oceanica* dégradés (Boudouresque et al., 2021).

Toutefois, la mise en place de projets de restauration des herbiers à grande échelle, ainsi que la pérennité des transplantations, nécessitent des méthodes de transplantation à la fois efficaces, écologiques, et adaptées aux conditions environnementales locales. D'autre part, la disponibilité des boutures représente une contrainte majeure. Il est donc essentiel de déterminer si les boutures-épaves présentent des performances biologiques et écologiques similaires aux boutures prélevées directement dans l'herbier, afin d'éviter des prélèvements destructeurs non justifiés. De plus, un consensus sur les protocoles de suivi permettant d'évaluer le succès des transplantations fait encore défaut (Pansini et al., 2022). La définition la plus couramment admise considère qu'une opération de transplantation est réussie lorsque le taux de survie des transplants et le taux de progression des rhizomes dépassent 50 % (Campbell, 2000). Cependant, le taux de survie (variable binaire vivant/mort) bien qu'indicateur central, reste insuffisant à lui seul car il ignore les altérations physiologiques pouvant précéder les signes morphologiques visibles (Pansini et al., 2022). Or, face à différentes pressions, plusieurs paramètres physiologiques se sont révélés être des indicateurs précoces de stress, mais demeurent encore peu intégrés en écologie de la restauration (Cozza et al., 2019 ; Pérez et al., 2008 ; Roca et al., 2014). L'adoption de tels descripteurs dans les protocoles de suivi permettrait un suivi plus fin et dynamique de l'acclimatation des herbiers transplantés (Ceccherelli et al., 2018; Cooke & Suski, 2008; Roca et al., 2014) et donc une réévaluation continue des opérations de restauration.

Dans ce contexte, cette thèse poursuit les objectifs suivants:

1) **Caractériser les conditions environnementales et la dynamique de recolonisation naturelle de l'herbier de *P. oceanica* sur les zones de matte morte dégradées**. Des lacunes persistent quant à la capacité de recolonisation naturelle de l'herbier de *P. oceanica*, alors qu'il est crucial d'évaluer la pertinence d'une intervention de restauration active par rapport à cette dynamique naturelle (Cunha et al., 2012). Les conditions environnementales facilitant ou contraignant la recolonisation

naturelle, et donc la pérennité des transplants, sont également très peu documentées. Cet objectif sera poursuivi par des acquisitions photogrammétriques annuelles pour suivre la dynamique de recolonisation naturelle. La caractérisation des conditions environnementales sera réalisée par des mesures de température, granulométrie, teneur en matière organique et structure des communautés microbiennes dans la matte morte. La présence d'autres macrophytes marines (invasives ou natives) se développant sur la matte morte sera également documentée.

2) **Evaluer l'efficacité relative de trois méthodes de transplantation utilisant des matériaux biodégradables, en fonction des conditions environnementales locales**. Cet objectif visera à déterminer si la rigidité et la structure tridimensionnelle du matériel de transplantation favorisent l'établissement à long terme des transplants. Les transplantations seront réalisées à deux profondeurs différentes, représentatives de conditions environnementales différentes de luminosité et d'hydrodynamisme. Le suivi de cet objectif sera réalisé par un suivi du taux de survie, de la production foliaire, de la morphologie foliaire et racinaire, ainsi qu'en termes de rapport cout-bénéfice apporté par chacune de ces méthodes de transplantation. Afin de ne pas limiter les conclusions à un suivi des variables morphologiques des transplants, le suivi de cet objectif sera complété par un suivi des communautés microbiennes foliaires et racinaires, ainsi que la mesure de traits physiologiques et biochimiques des transplants.

3) **Déterminer si les boutures-épaves présentent des performances similaires aux boutures prélevées dans un herbier naturel** dans le cadre de leur utilisation comme matériel donneur pour la transplantation. Comme pour la discrimination entre les méthodes de transplantation, le suivi de cet objectif sera réalisé par un suivi morphologique, microbiologique, physiologique et biochimique des deux origines de boutures.

4) **Comparer, trois ans après la transplantation, les traits des transplants avec ceux des herbiers naturels.** Cette analyse permettra d'évaluer la convergence morphologique, microbiologique, physiologique et biochimique des transplants vers les herbiers naturels.

Bien que la démarche scientifique développée dans cette thèse s'articule autour de quatre objectifs principaux, l'organisation du manuscrit répond à une logique complémentaire, visant à présenter d'abord le cadre général et l'état des connaissances, avant de détailler les approches expérimentales et d'ouvrir sur des perspectives plus larges. Ainsi, le manuscrit est structuré en quatre chapitres :

Chapitre I : Présenter le cadre général de l'étude à travers une synthèse sur la restauration écologique, la mer Méditerranée, la biologie et l'écologie de *P. oceanica*, les pressions anthropiques qui pèsent sur cette espèce, son statut de protection, ainsi que l'état actuel des connaissances sur la restauration des herbiers de *P. oceanica*.

Chapitre II : Évaluer si les facteurs influençant la dynamique de recolonisation naturelle, dans des zones de matte morte dégradées par l'ancrage, conditionnent également la performance des différentes méthodes de transplantation et des origines de boutures.

Chapitre III : Utiliser des descripteurs microbiologiques, physiologiques et biochimiques pour analyser les mécanismes limitant la croissance et l'acquisition des nutriments essentiels, et comprendre en quoi ces contraintes expliquent les différences de performances observées entre méthodes de transplantation et origines de boutures.

Chapitre IV : Proposer une synthèse et une réflexion globale, en mettant en évidence l'apport de ces travaux et en envisageant des perspectives de restauration à l'échelle régionale et sur des sites à plus grande échelle spatiale.

Chapitre II

Résilience et restauration de l'herbier de *Posidonia oceanica* après dégradation par l'ancrage : comparaison entre recolonisation naturelle et restauration active par transplantation

Ce chapitre est composé de deux articles :

Article 1 : Boulenger, A., Chapeyroux, J., Fullgrabe, L., Marengo, M., & Gobert, S. (2025). Assessing *Posidonia oceanica* recolonisation dynamics for effective restoration designs in degraded anchoring sites. *Marine Pollution Bulletin*, 216, 117960. https://doi.org/10.1016/j.marpolbul.2025.117960

Article 2 : Boulenger, A., Marengo, M., Boissery, P., & Gobert, S. (2025). Comparative assessment of transplantation methods and donor sources for the restoration of *Posidonia oceanica* meadow. *Science of the Total Environment*, 1000, 180488. https://doi.org/10.1016/j.scitotenv.2025.180488

Assessing *Posidonia oceanica* recolonisation dynamics for effective restoration designs in degraded anchoring sites

Arnaud Boulenger[a,b], Juliana Chapeyroux[a], Lovina Fullgrabe[b], Michel Marengo[b], Sylvie Gobert[a,b]

[a] Laboratory of Oceanology, MARE Centre, UR FOCUS, University of Liege, 11 allée du six août, 4000, Liege, Belgium

[b] STAtion de REcherche Sous-marines et Océanographiques (STARESO), 20260 Calvi, France

Marine Pollution Bulletin (2025)

Abstract

The Mediterranean seagrass species *Posidonia oceanica* forms extensive meadows that provide numerous ecological and economic services. Among the human activities threatening these meadows, boat anchoring causes severe degradation resulting in meadow fragmentation, exposure of the dead matte, and sediment disruption. In this study, we assessed the natural recolonisation dynamics of *P. oceanica* in anchoring-degraded sites focusing on both shallow and deep sites. Over two years, photogrammetry was employed to monitor recolonisation dynamics with a focus on patchs' edges expansion and storm-fragments accumulation. Our results show distinct recolonisation patterns between shallow and deep sites, with shallow patches displaying more variable dynamics of erosion and recolonisation, while deep patches showed slower but more consistent recovery. Additionally, the abundance of storm-fragments, primarily in shallow areas, suggests potential for enhanced recovery through natural trapping structures. Despite recent regulations reducing anchoring pressures, recolonisation rates remain insufficient to counteract the extent of degradation in a reasonable timespan. These findings underline the importance of designing tailored restoration strategies based on site-specific recolonisation potential: high-density transplantation with durable anchoring structures in shallow areas to withstand hydrodynamic forces, and more cost-effective solutions like iron staples in deeper areas. Additionally, the study supports the use of trapping substrates to retain storm-fragments in shallow sites to boost natural recolonisation. This approach is crucial for enhancing seagrass meadow resilience, especially within a context of climate change and increasing pressures on coastal ecosystems.

Keywords

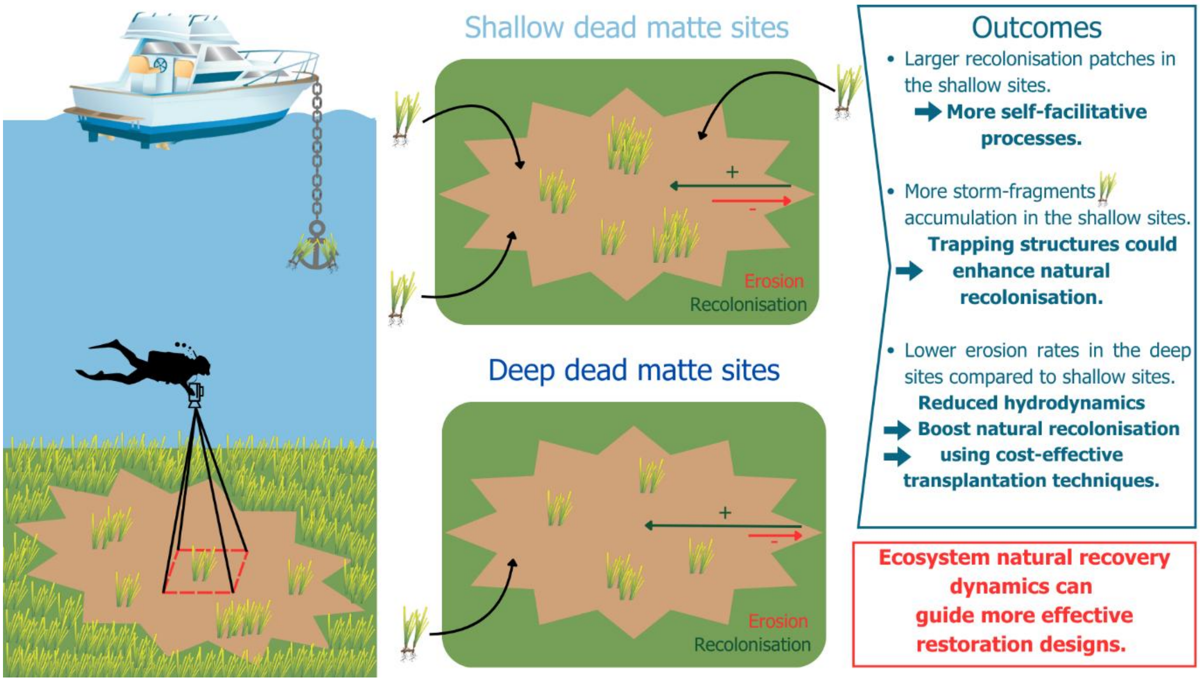
Seagrass, Anthropogenic patches, Photogrammetry, Restoration, Recovery

ACKNOWLEDGMENTS

This work was supported by the University of Liege and the Fonds National de la Recherche Scientifique – FNRS (grants ASP 40006932 and CDR J.0076.23). This study is part of the STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) project funded by the Territorial Collectivity of Corsica and by the Rhone-Mediterranean and Corsica Water Agency. Authors are grateful to the STARESO for facilities and field assistance. The authors have no conflicts of interest to declare.

AUTHORS' CONTRIBUTION

Arnaud Boulenger: Conceptualization, Field sampling, Methodology, Formal analysis, Investigation, Writing, Visualization; Juliana Chapeyroux: Methodology, Formal analysis, Investigation, Writing, Visualization; Lovina Fullgrabe: Formal analysis, Writing, Visualization; Michel Marengo: Conceptualization, Methodology, Writing, Supervision, Funding; Sylvie Gobert: Conceptualization, Methodology, Writing, Supervision, Funding.



Outcomes

- Larger recolonisation patches in the shallow sites.
  ➡ **More self-facilitative processes.**
- More storm-fragments accumulation in the shallow sites.
  ➡ **Trapping structures could enhance natural recolonisation.**
- Lower erosion rates in the deep sites compared to shallow sites.
  Reduced hydrodynamics
  ➡ **Boost natural recolonisation**
  ➡ **using cost-effective transplantation techniques.**

Ecosystem natural recovery dynamics can guide more effective restoration designs.

Graphical abstract used for publication.

1. Introduction

Posidonia oceanica (L.) Dellile is a seagrass species endemic to the Mediterranean Sea, where it forms extensive meadows of critical importance due to the wide range of ecosystem services they offer, acting as nurseries (Campagne et al., 2014), serving as carbon sinks (Monnier et al., 2022; Pergent-Martini et al., 2021), and providing protection against coastal erosion (Gacia et al., 1999; Gacia & Duarte, 2001). *P. oceanica* thrives at depths between 0.5m and 40m in low turbid waters (Gobert et al., 2006) and is sensitive to strong hydrodynamic conditions, such as wave-induced physical damage and matte erosion (Boudouresque et al., 2006; Ruju et al., 2018). This seagrass species can tolerate temperatures ranging from 9 to 29°C (Boudouresque & Meinesz, 1982), although this species is sensitive to increases in water temperature and growth is limited above 27°C (Guerrero-Meseguer et al., 2017; Stipcich et al., 2022a). Besides the rising seawater temperature, global warming also poses significant threats to *P. oceanica* meadows through sea level rise, and the introduction of exotic species leading to the displacement of seagrass communities (Pergent et al., 2014; Stramska & Aniskiewicz, 2019). Moreover, human activities can affect *P. oceanica* meadows (Boudouresque et al., 2009; Giakoumi et al., 2015) both indirectly, by deteriorating water quality (Bockel et al., 2024; Montefalcone et al., 2007), and directly, through habitat destruction caused by coastal development (Holon et al., 2015; Mancini et al., 2023), trawling (Kiparissis et al., 2011), or anchoring (Abadie et al., 2016, 2019; Pergent-Martini et al., 2022). Over half of the world's large (i.e. > 24m length) recreational boats spend the summer months in Mediterranean waters (Carreño & Lloret, 2021), primarily in the North-Western Mediterranean (Pergent-Martini et al., 2022). Anchoring activities, mainly in shallow waters, cause both direct and indirect damage during the process of anchor deployment and retrieval, as well as from the movement of the chain or rope. Repeated anchoring of recreational boats at depths greater than 15m results in widespread degradation of these meadows (Abadie et al., 2015). The removal of the *P. oceanica* foliar canopy and the belowground organs lead to the exposure of the matte, which consists solely of the remaining rhizomes, roots, and sediment particles: this formation is referred to 'dead matte'. Beyond the physical damage, this can also cause chemical alterations in the sediment. Initially, carbonate sediments, which may be oxygenated by the presence of living plants, gradually transition to fine particles that fill gaps within decaying organic matter, resulting in an anoxic bare mat (Boudouresque et al., 2016; Mateo & Romero, 1997; Pergent et al., 2014). This change in substrate quality can lead to the infiltration of hydrogen sulphide (H_2S) (Abadie et al., 2016), a chemical compound that inhibits the growth and development of *P. oceanica* (Calleja et al., 2007; Holmer et al., 2003; Marbà et al., 2006). Furthermore, the dead matte left behind by anchoring provides an ideal environment for invasive species like *Caulerpa cylindracea* to establish and spread (Casoli et al., 2021; Katsanevakis et al., 2010;

Kiparissis et al., 2011). This macroalga increases hydrogen sulphide concentrations in the sediments, further hindering the potential for *P. oceanica* to recolonize the dead matte (Garcias-Bonet et al., 2008; Holmer et al., 2009).

The implementation of French prefectorial decrees banning anchoring in *P. oceanica* meadows for boats over 45 meters in 2016 (Préfecture maritime de Méditerranée, 2016) and for boats over 24 meters in 2019 (Préfecture maritime de Méditerranée, 2019) has led to a significant reduction in the degradation of these meadows (Bockel et al., 2023)**.** Once the causes of meadow regression have ceased, natural recolonisation can occur through the expansion of a meadow's edge due to the growth of plagiotropic rhizomes, the dispersal of cuttings by currents (especially during storms) (Boudouresque et al., 2021)., and the recruitment and establishment of seagrass patches via seedlings (Balestri et al., 2017; Boudouresque et al., 2021). However, while the detrimental effects of anthropogenic pressures and declining environmental conditions on seagrass beds are well-documented, the phenomenon of natural recolonisation has received little attention (Almela et al., 2008; Bockel et al., 2024; Kendrick et al., 2005; Olesen et al., 2004). The lack of studies on this topic is particularly concerning given the increasing number of *P. oceanica* meadow restoration projects in recent years (Boudouresque et al., 2021; Pansini et al., 2022). Indeed, the assessment of natural recolonisation following a disturbance is essential when considering seagrass restoration operations (Boudouresque et al., 2021; Cunha et al., 2012). Active restoration is justified only after observing natural recolonisation at the intended restoration site, demonstrating the return of suitable environmental conditions (Boudouresque et al., 2021; Descamp et al., 2025; Mancini et al., 2022; Unsworth et al., 2024). Additionally, comparing transplanted areas with the site's natural recolonisation capacity is crucial for an objective assessment of the restoration efforts' costs and benefits (Boudouresque et al., 2021). In May 2022, a pilot project was launched in the Bay of Calvi (North-Western Corsica, France) to restore *P. oceanica* meadows by transplanting cuttings onto dead matte resulting from anchoring degradation (Boulenger et al., 2024). Alongside these transplantation trials, the natural recolonisation of the *P. oceanica* at the restoration sites is being monitored using photogrammetry, an accurate and low-cost mapping technique (Marre et al., 2020). Specifically, the main objective of this study was to assess the recovery dynamics of *P. oceanica* within degraded areas at shallow and deep sites in North-Western Corsica. Our aims were to assess whether there were differences in recolonization rates between shallow and deep sites and to determine the relative importance of patch recruitment rate in *P. oceanica* spatial colonization by measuring the inter-annual variation in patch size distribution, as well as the accumulation and establishment of drifting vegetative fragments. Additionally, differences in environmental conditions, including temperature and sediment characteristics, that may influence recolonization dynamics at the two study depths, were measured. Finally, the implications of *P. oceanica* recovery dynamics for improving seagrass restoration designs are discussed.

2. Material and Methods

2.1 Study area

The study sites were located in Alga Bay (8°43′52′′ E; 42°34′20′′ N), a sub-bay of the Bay of Calvi in North-Western Corsica (France) (Figure 2.18). Alga Bay covers a total area of 1 km² and features a specific characteristic known as 'return river,' a large sandy area where seagrass meadows are unable to thrive, likely due to bottom currents generated by surface currents reflecting off the coast (Boudouresque & Meinesz, 1982). An extensive *P. oceanica* meadow, covering a total area of 78 ha and located between 3 and 37m depth, is present in Alga Bay (Figure 2.18) (Abadie et al., 2016). This meadow has a good ecological status based on the PREI index (Gobert et al., 2009). The only anthropogenic stressor is the anchoring of leisure boats (Fullgrabe et al., 2022). Indeed, intensive anchoring activity over the last decades has led to severe seagrass meadow loss due to mechanical destruction, with an estimated 8 ha lost in Calvi Bay between 2012 and 2018 (Fullgrabe et al., 2022). Since the enforcement of the new decree in 2019, the proportion of boats over 20m in length dropping anchors in *P. oceanica* meadows has decreased by 43.8% (Figure S2.1). The bare areas of dead matte are heavily colonized by the invasive macroalga *C. cylindracea* (Figure 2.19A), and to a lesser extent by the native macroalga *C. prolifera* (Figure 2.19B).

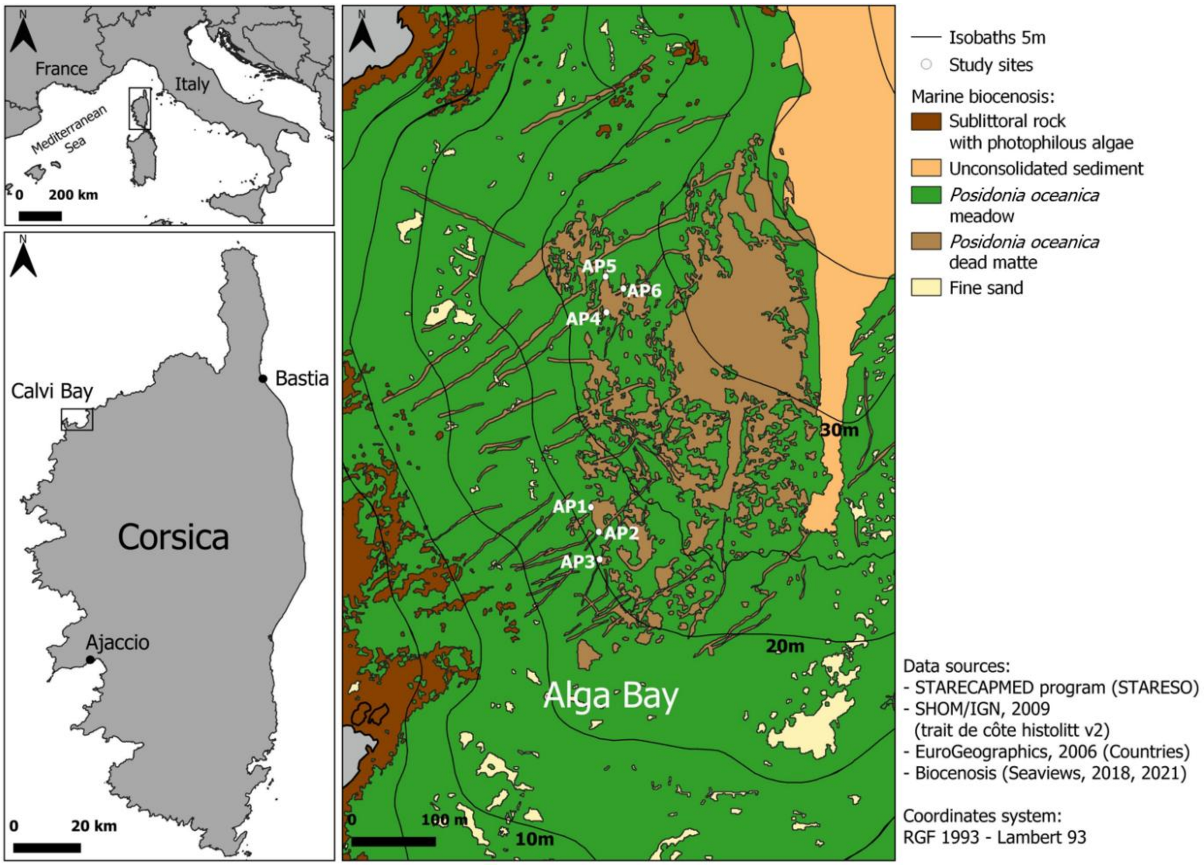


Figure 2.18. *Location of the study area. The top left figure shows a wider view of Corsica and surrounding countries. The bottom left figure displays a more detailed view of Corsica and the location of Calvi Bay. The figure on the right shows Alga Bay with associated marine biocenosis, the isobaths every 5m depth and the six study sites (AP1 – AP6).*

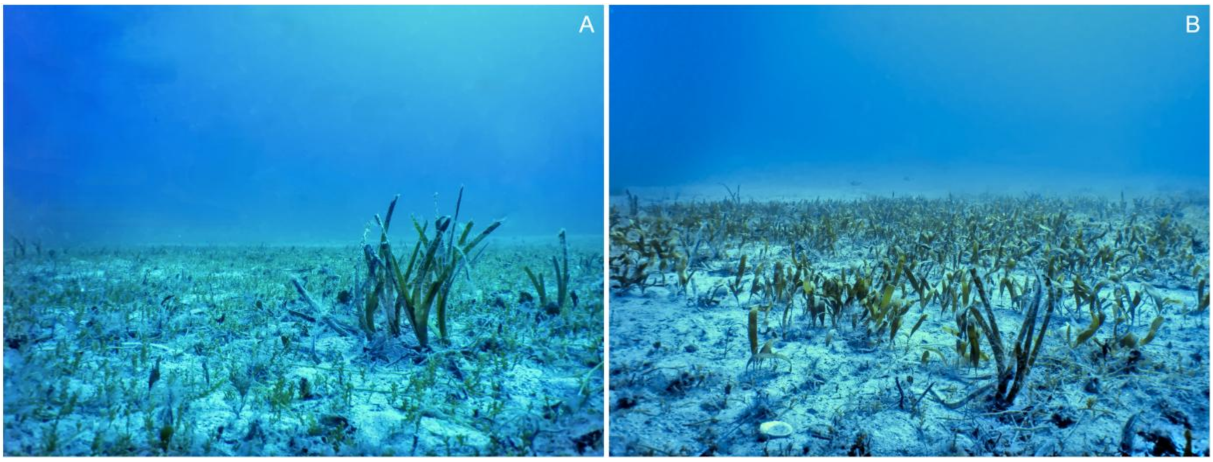


Figure 2.19. *Thin layer of sand covering the dead matte with a few scattered P. oceanica shoots. This area provides a suitable substrate for colonization by two different macroalgae species: (A) a dense network of C. cylindracea, (B) a canopy of C. prolifera but the presence of C. cylindracea in the forefront of the picture can also be noticed. Both photos were taken at 28m depth in close vicinity to the study site AP6.*

Three patches of dead matte were selected as study sites at two different depths: 20 m (sites AP1, AP2 and AP3) and 28 m (sites AP4, AP5, and AP6) depth, since most of the degradation caused by anchoring pressure occurs in the deeper portion (20-35 m) (Figure 2.18). The patches of dead matte selected in this study were chosen because they correspond to restoration sites used in the *REPAIR* project, where *P. oceanica* cuttings were transplanted in spring 2022 (Boulenger et al., 2024). Thus, recolonisation can be compared between natural and transplanted areas, providing a more objective assessment of the effectiveness of the tested restoration techniques (Boudouresque et al., 2021).

2.2 Environmental parameters

Seawater temperature was monitored to determine whether warming affects *P. oceanica* recovery dynamics at shallow and deep sites by influencing the expansion or reduction of seagrass patches' size and the recruitment rates of new seagrass patches. To achieve this, seawater temperature was recorded at the study depths using HOBO loggers (HOBO Pendant Temperature Data Logger, Onset Computer Corporation) at 10-minute intervals from May 2022 to May 2024. The loggers were placed just above the dead matte at the six study sites. Due to recording errors by the data loggers, no data were recorded at 28m for the time intervals from 2022/05/01 to 2022/08/16, and from 2024/03/27 to 2024/04/15. At 20 m, data were missing only for the time interval from 2024/03/27 to 2024/04/15.

Besides seawater temperature, sediment characteristics also play a key role in the spatial colonisation dynamics of *P. oceanica*. More specifically, the organic matter buried in the sediment is an important source of nutrient to seagrasses (Evrard et al., 2005; Fraser et al., 2016; Kilminster et al., 2006), and sediment granulometry partly determine porewater nutrient availability (de Boer, 2007; Holmer et al., 2001). Therefore, sediment cores (20cm depth x 5cm diameter) were collected in May 2024 to characterize the sediment granulometry and organic matter content of the seafloor at the six study sites. Five samples were taken in the dead matte from each of the six study sites (n=30). In addition, five samples were also collected in nearby *P. oceanica* meadows at both study depths (n=10). After field collection, samples were stored at -20°C and sent to MicroPolluants Technology SA (Saint Julien Les Metz, France). Sediment cores were dried at 60°C and passed through a 2mm diameter sieve to remove the coarser fraction of the sediment. The percentage of organic matter in each sediment sample was determined by loss on ignition (LOI), combusting at 550°C for 4 hours (Heiri et al., 2001). Inorganic carbon was then removed from the samples by combustion at 950°C for 2 hours (Heiri et al. 2001) before grain size determination. A small amount of sediment from each sample, with organic matter and inorganic carbon removed, was placed in circulating water under continuous sonication for sediment composition analysis (i.e. the percentages of clay, silt, and sand) and median grain size measurements using laser-diffraction

(Pratica LA-960, HORIBA Ltd, Kyoto, Japan). Finally, sediment classification was performed according to Wentworth's grain size classes (Wentworth, 1922).

2.3 Monitoring of P. oceanica natural recolonisation dynamics by photogrammetry

2.3.1 Underwater images acquisition

To monitor natural recolonisation of *P. oceanica* on dead matte, photographic surveys were conducted at each site (n=6) by a scuba-diver at an average distance of 5m from the sea bottom conducting parallel, regularly spaced transects. Surveys were carried out at a relatively low swimming speed of 20-25 m.min^{-1}, with a time-lapse of approximately 1s between pictures (photo density: 4–5 photos/m²) as recommended by Marre et al. (2019). The photographs were taken with a pure nadiral orientation using a 20.1 Mega Pixels Sony Cyber-Shot RSC-RX100 Va in a waterproof Nauticam housing, with the following settings: shutter speed = automatic, aperture = F12, sensibility = 400 ISO. In some cases, the settings were slightly adjusted depending on environmental conditions (e.g., lighting and visibility) at the time of sampling. Focus was set automatically before each acquisition. The surveys were conducted over two years (2022 - 2024), each year during the same period (May - June) to avoid differences in leaf growth stages. The total acquisition time per site ranged between 33 and 53 minutes; with a total of 173 to 701 photos collected per site.

2.3.2 Photogrammetric images processing

When necessary, the images were batch-processed for quality improvements using Darktable v4.6.1, enhancing colour contrast, brightness and sharpness. They were then processed with Agisoft Metashape Professional v1.8.4 (Agisoft LLC, 2022), a commercial photogrammetry software extensively used in the scientific community for seagrass meadow monitoring (Abadie et al., 2020; Bockel et al., 2024; Marre et al., 2019, 2020; Piazzolla et al., 2024; Ventura et al., 2022, 2023). This software follows a classic photogrammetric workflow, including images alignment, automatic identification of key points in all photos, bundle adjustment, point cloud densification, mesh building, texturing and orthomosaic production. The specific parameter settings for the different steps of the photogrammetric workflow are provided in Table S2.1. As the photogrammetric process requires known reference distances to produce metric results, reference makers with known distances between them, called scale bars, were used (Rende et al., 2022; Ventura et al., 2022). Scale bars are particularly useful when it is not feasible to place ground control points across the entire site. They offer a more time-efficient alternative, as placing a few accurately measured scale bars is simpler than using specialized equipment to determine the coordinates of multiple markers (Agisoft LLC, 2022).

Six scale bars were installed on the seafloor of each site just before the start of the transects and were used for alignment optimization after bundle adjustment. Scale bars with an error greater than 10cm were removed from the model. Half of the scale bars (n=3) were not included in the alignment optimization but were retained as validation scale bars to assess the accuracy of internal measurements after bundle adjustment (Ventura et al., 2022). The scale bars placed in the mapped area included four coded markers fixed to a 1 x 1m cross-scale bar with the same coded marker consistently oriented north using a compass. This configuration ensured that all models were aligned uniformly (Abadie et al., 2022). Moreover, these scale bars were positioned at the same locations each year using rebars placed into the dead matte. This setup allowed the markers on the scale bars to function as fixed reference points, facilitating relative positioning by aligning the orthomosaics of each site over the two years of monitoring (Abadie et al., 2022). All orthomosaics were exported in a TIFF format with a resolution of 0.0003m to preserve the local metric coordinate system for further GIS analysis.

2.3.3 P. oceanica natural recolonisation monitoring over time

To monitor *P. oceanica* natural recolonisation, four monitoring quadrats (3x3m) near the transplantation units were initially randomly selected on the orthomosaics of each study sites (n=24) (Figure 2.20). This framing approach allowed for smaller-scale analysis and accelerated data processing. Within each of these 9 m² quadrats, the seagrass area (m²) was manually digitized and quantified using QGIS v3.36.1 (Figure 2.20). The seagrass area within each quadrat was measured annually to assess whether there was stability, an increase, or a reduction in the total seagrass area and in the individual surface area of each seagrass patch at the two study depths. Moreover, measuring the inter-annual variations in seagrass area (m²) allowed us to calculate the annual average recolonization and erosion rates (m²/year) for each site. The percentage cover of *P. oceanica* in each quadrat was calculated as the ratio of seagrass area (m²) to the quadrat area (m²). This metric was used to classify the fragmentation level at each study site. The fragmentation level describes the complex spatial patterns used to analyze landscape configuration (Saura, 2002). Different levels of fragmentation were defined by Sleeman et al. (2005) to characterize seagrass seascapes along a continuum of decreasing patch size and increasing isolation, ranging from large continuous seagrass meadows to many small, dispersed seagrass patches. They are five categories based on the proportion of seagrass cover: : many small patches for seagrass cover below 16%, medium patches represent 16%–32%, few large patches cover 32%–45%, fragmented continuous meadows cover 45%–86%, and fully continuous meadows exceed 93% seagrass cover (Sleeman et al., 2005).

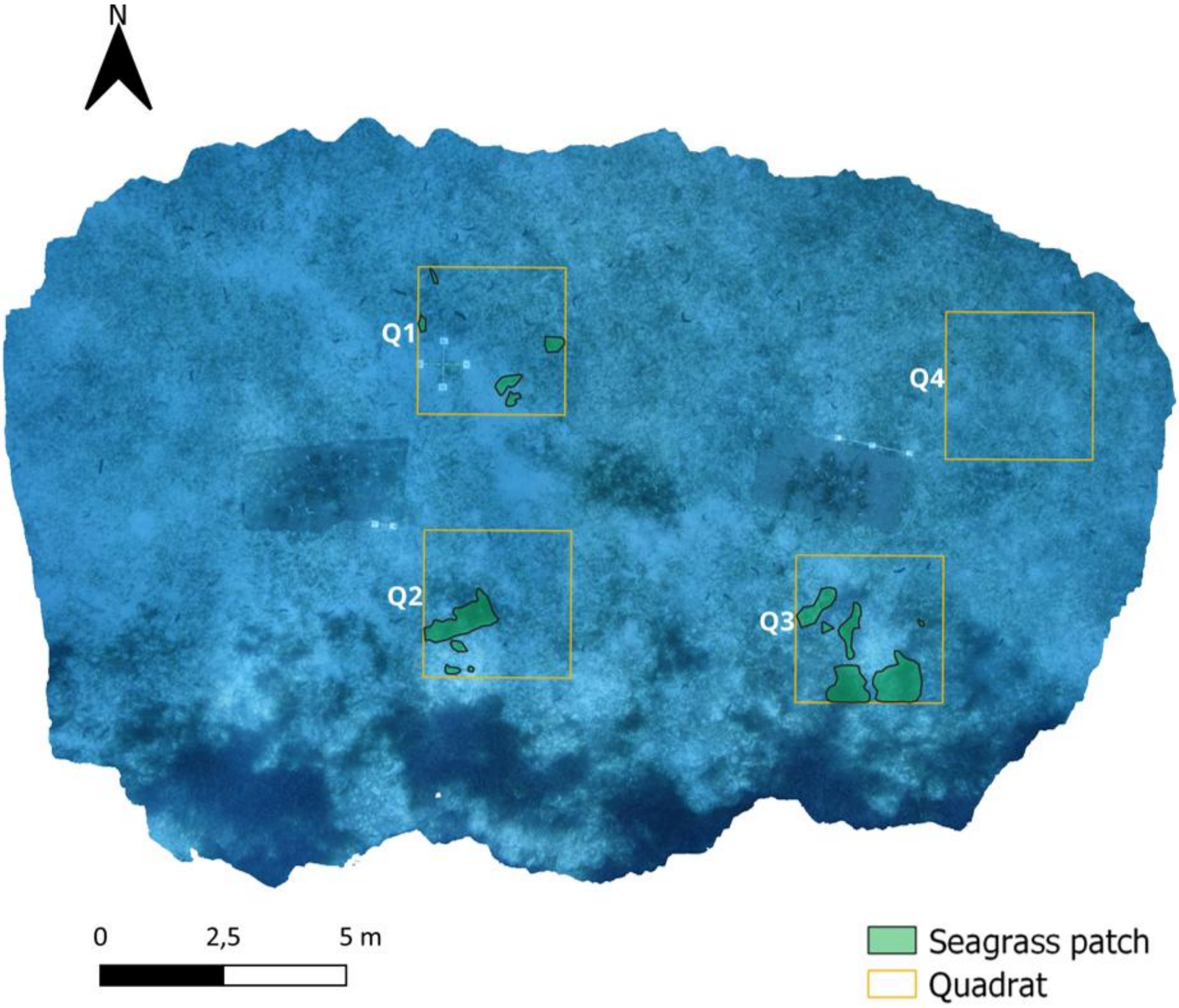


Figure 2.20. *Orthophoto of one of the study sites (AP4 located at 28m depth in May 2024) with the manual delineation of P. oceanica patches within the four quadrats (Q1 to Q4) placed in a dead matte area. The three transplantation sites of the REPAIR project are also visible on the orthophoto (see Boulenger et al., 2024).*

Measuring the inter-annual variations in seagrass area, percent cover, and annual recolonization and erosion rates provided an initial insight into the recovery dynamics of *P. oceanica* at shallow and deep sites. However, to obtain more precise information on recovery dynamics, it is essential to monitor patch population dynamics (Almela et al., 2008). To achieve this, each individual seagrass patch within the monitoring quadrats was assigned to one of ten size classes, with each category defined by a doubling of the previous size (Almela et al., 2008). The temporal evolution of patch size distribution allowed us to determine whether seagrass patches tend to increase in size over time and to assess the recruitment of new seagrass patches. Finally, the number of drifting vegetative fragments (hereafter referred to as storm-fragments) were counted within each quadrat over the two-year monitoring period to assess the potential for new *P. oceanica* patches initiation and recruitment.

2.4 Data analysis

Differences in median grain size, mud content and organic matter content were tested using a two-way ANOVA , with Biocenosis (dead matte vs. *P. oceanica* meadow) and Bathymetry (shallow vs. deep) as independent variables to assess how sediment characteristics vary between degraded and vegetated areas and across depths. Prior to the ANOVA test, normality and linearity of the residuals were assessed by visual inspection of residuals versus fitted values plots and with a Shapiro-Wilks test. Levene's test was used to check for homogeneity of variances. Since the normality assumption was not met for mud content and organic matter content, data were log transformed. Differences in *P. oceanica* meadow area, percent cover, number of patches and number of storm-fragments were tested using the non-parametric Kruskal-Wallis test with Time (0, 12, 24 months) and Bathymetry as factors to evaluate temporal trends in *P. oceanica* recolonization and depth-related differences. A non-parametric test was chosen as normality could not be achieved. Statistical significance was set at $p < 0.05$. Statistical analyses were performed in RStudio v4.3.2 (RStudio Inc., Boston, MA, USA). All values are reported as mean ± standard error.

3. Results

3.1 Environmental parameters

The seawater temperature ranged from a maximum of 27.1°C at 28 m and 26.8°C at 20 m in September 2022 to a minimum of 13.7°C at both 28 m and 20 m in February 2023 (Figure 2.21). Seasonal variations were pronounced, and thermal stratification was observed in spring 2023 and 2024, with temperatures higher at 20 m than at 28 m. This thermal stratification gradually dissipated as summer approached (Figure 2.21).

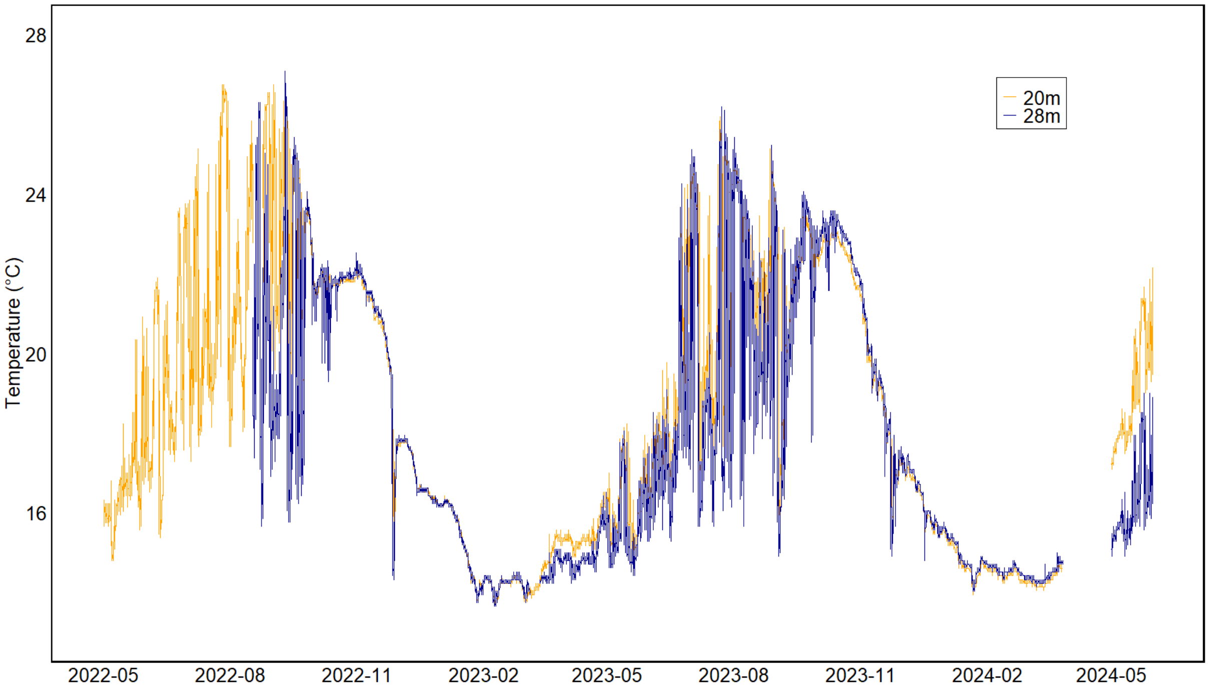


Figure 2.21. *Temporal dynamics of seawater temperature at the shallow (20m) and deep (28m) study sites from May 2022 to May 2024. Missing data were represented as blank spaces in the figure.*

The median grain size was significantly higher at the shallow sites compared to the deeper sites (p = 0.015; F = 6.765), while no significant differences were found between the dead matte and *P. oceanica* meadow (Figure 2.22). The mud content (0.01µm < grain size < 63µm) showed the opposite pattern, with significantly higher values for the deeper sites compared to the shallow sites (p = 0.009; F = 8.103) (Figure 2.22). Both Biocenosis and Bathymetry had a significant influence on the organic matter content, with higher values for the deep sites compared to the shallow sites (p = 0.009; F = 7.977) and higher values in the *P. oceanica* meadow compared to dead matte (p = 0.003; F =11.100) (Figure 2.22).

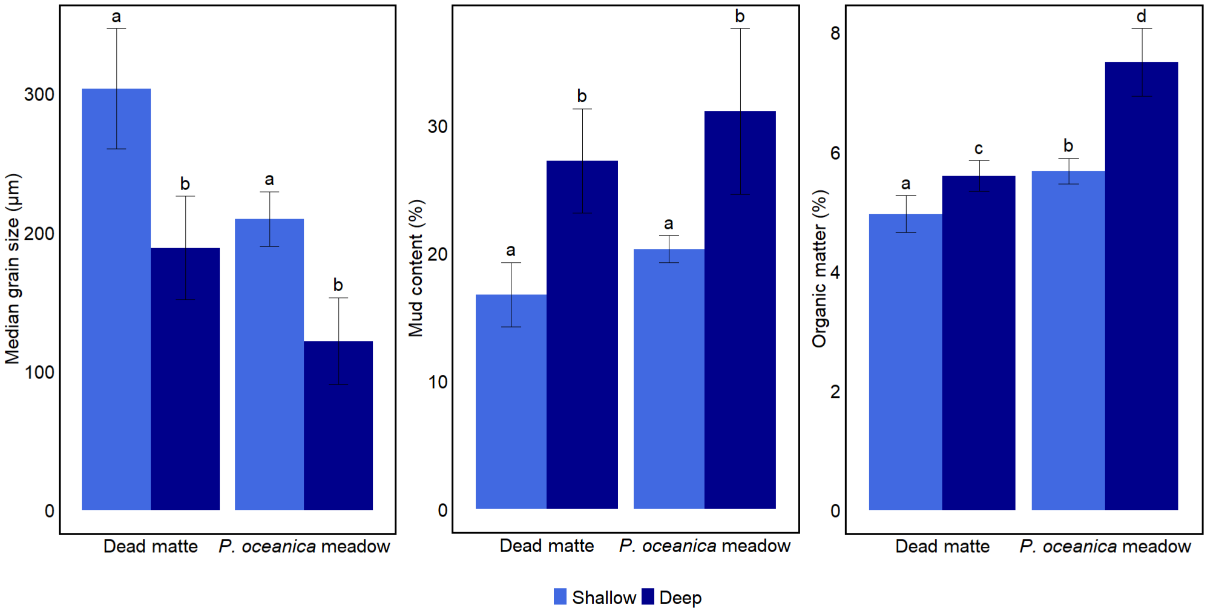


Figure 2.22. *Mean median grain size (left), mud content (centre), and organic matter content (right) in the dead matte and P. oceanica meadow as a function of depth. Vertical error bars represent standard errors. Statistically significant differences are indicated by different letters (a, b, c, and d).*

3.2 P. oceanica meadow recovery dynamics

The six selected patches of dead matte exhibit considerable heterogeneity in area, with no apparent relationship to their depth (Table 2.1). Their average size is 191.5 m², with individual patch areas ranging from 56.6 m² at a depth of 20 m to 319.2 m² at a depth of 28 m (Table 2.1).

Table 2.1. *Initial dead matte patch area (m²) at the beginning of the study.*

Site	Bathymetry	Dead matte patch area (m²)
AP1	Shallow	194.2
AP2	Shallow	300.7
AP3	Shallow	56.6
AP4	Deep	226.9
AP5	Deep	319.2
AP6	Deep	206.1

The two-year monitoring of the area covered by the *P. oceanica* seagrass bed showed high variability within the 24 quadrats across the 6 study sites. The largest seagrass area measured 4.15m² in one of the quadrats at site AP3, located at 20 m depth. Conversely, some quadrats exhibited a seagrass area of 0.00 m², as observed at site AP4 at 28 m and site AP2 at 20 m. The temporal dynamics of *P. oceanica* meadow area and relative cover over the two years of monitoring showed a slight progressive trend for both shallow and deep sites (Figure 2.23).

For the shallow sites, the mean colonised seagrass area increased by 12.90%, from 1.24 ± 0.40 m² at the start of the monitoring to 1.40 ± 0.46 m² two years later (Figure 2.23). A mean progression of 35.13% was observed for the deep sites, from 0.74 ± 0.17 m² at 0 months to 1.00 ± 0.24 m² after two years (Figure 2.23). In terms of *P. oceanica* meadow percent cover relative to the dead matte cover, there was an increase from 13.82 ± 4.46 % initially to 15.57 ± 5.17 % after two years for the shallow sites (Figure 2.23). A similar pattern was observed for the deep sites, with an initial 8.12 ± 1.96 % cover increasing to 11.14 ± 2.66 % two years later (Figure 2.23). According to the fragmentation classification by Sleeman et al. (2005), there was a high level of fragmentation, with the meadow primarily consisting of small sized seagrass patches as the percent cover was always lower than 16% (Figure 2.23). No significant differences were found in seagrass area or percent cover between the three monitoring campaigns ($p = 0.860$; $\chi^2 = 0.300$; p= 0.845; $\chi^2 = 0.337$; respectively) or between sites at the two different depths ($p = 0.937$; $\chi^2 = 0.006$; p= 0.915; $\chi^2 = 0.011$; respectively) (Figure 2.23).

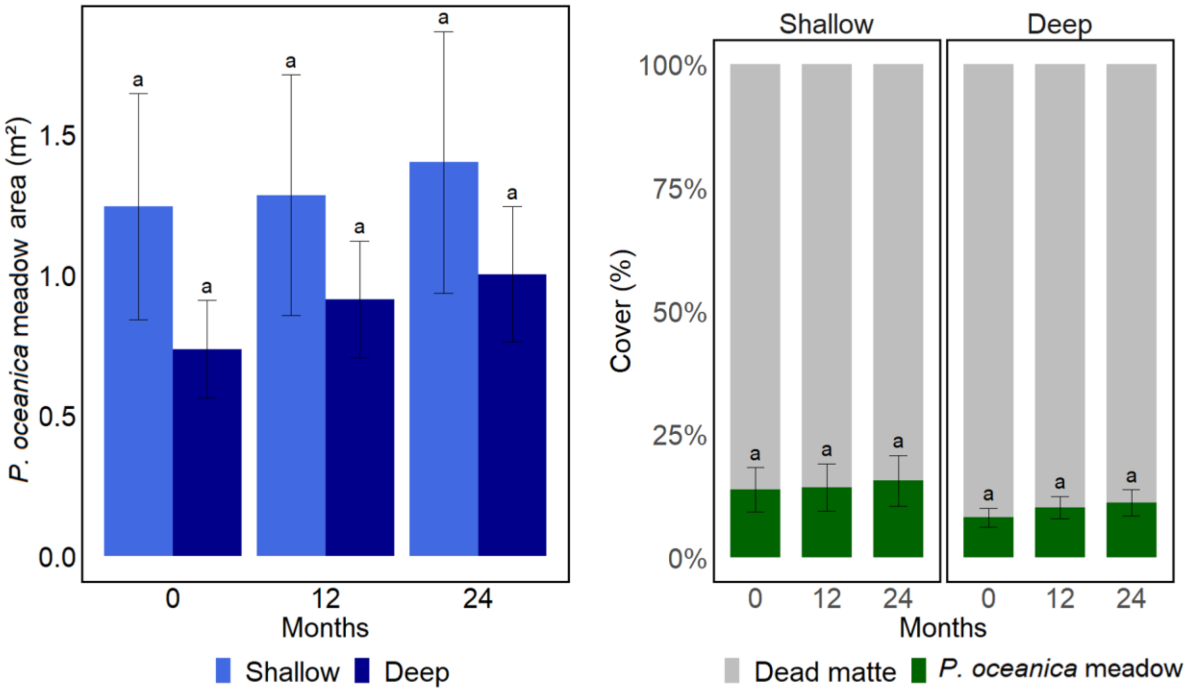


Figure 2.23. *Mean temporal dynamics of P. oceanica meadow area (m²; left figure) and P. oceanica relative cover (%, right figure) as a function of depth. Vertical error bars represent standard errors. The absence of statistically significant differences is indicated by the same letter (a).*

The measurement of the inter-annual variations in seagrass area (m²) allowed us to calculate the annual average recolonization and erosion rates (m²/year) for each depth (Figure 2.24). Both recolonisation and erosion (i.e. regression in seagrass area) processes were observed at each study site, although some sites (i.e., AP3 at 20 m; AP4 and AP5 at 28 m) showed very limited erosion (Table S2.2). Significant disparities were also observed within the quadrats across the different study sites. For the shallow sites, a maximum recolonisation rate of 0.59 m²/year and a maximum erosion rate of 0.25 m²/year were observed (Table S2.2). For the deep sites, a maximum recolonisation rate of 0.81 m²/year and a maximum erosion rate of 0.14 m²/year were recorded (Table S2.2). The mean annual recolonisation and erosion rates exhibit different patterns depending on the depth (Figure 2.24). The deep sites show a higher mean recolonisation rate (0.20 ± 0.06 m²/year) compared to the mean erosion rate (0.07 ± 0.01 m²/year) (Figure 2.24). In contrast, the shallow sites appear to have more balanced processes, with a smaller difference between the mean recolonisation rate (0.16 ± 0.04 m²/year) and the mean erosion rate (0.09 ± 0.03 m²/year) compared to the deep sites (Figure 2.24). It is also important to note that the mean recolonisation rates are consistently higher than the mean erosion rates regardless of the depth (Figure 2.24).

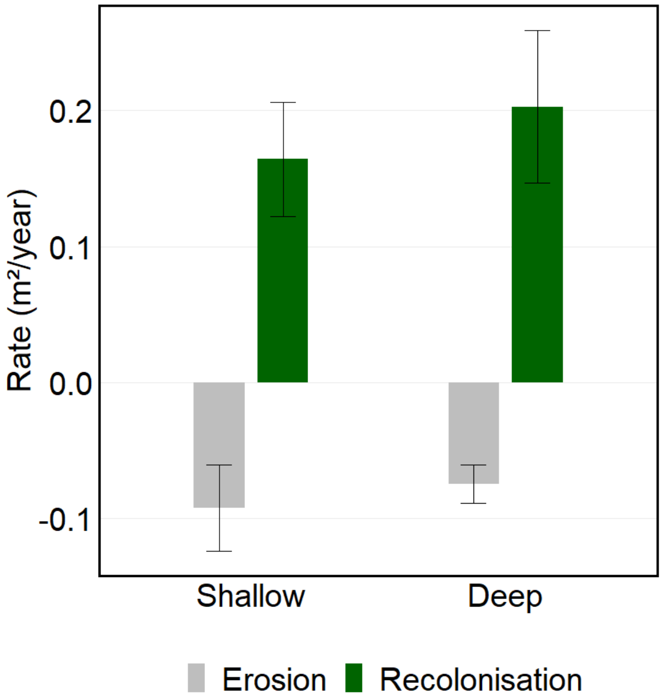


Figure 2.24. *Mean annual recolonisation and erosion rates (m²/year) as a function of depth. Vertical error bars represent standard errors. Positive values indicate recolonisation while negative values indicate erosion.*

Assessing inter-annual variations in seagrass area, cover, and recolonization/erosion rates offers a first glimpse into *P. oceanica* recovery dynamics. However, precise insights require monitoring patch population dynamics. As it was first observed with the seagrass area, high variability was observed in the number of *P. oceanica* patches within the quadrats. Up to 14 distinct seagrass patches were identified within a single quadrat, while some quadrats contained only one seagrass patch or none at all. No significant differences were observed in the number of seagrass patches over time ($p = 0.089$; $\chi^2 = 4.829$) or between the two study depths ($p = 0.337$; $\chi^2 = 0.921$). The mean number of seagrass patches per quadrats was 4.41 ± 2.75, with a mean size of 0.078 ± 0.027 m². In the shallow sites, at the beginning of the study, seagrass patches were primarily distributed across intermediate size classes, notably within the 400–800 cm², 800–1600 cm², and 1600–3200 cm² classes (Figure 2.25). After one year, there was a notable increase in the number of seagrass patches within the small and intermediate size classes, as well as an increase in the number of very large seagrass patches (12,800–25,600 cm²) (Figure 2.25). After two years, seagrass patches in the very small size class (0–100 cm²) disappeared, the number of seagrass patches in the intermediate size classes decreased, and the number of seagrass patches in the 1600–3200 cm² class increased (Figure 2.25). The number of very large seagrass patches (12,800–25,600 cm² and >25,600 cm²) remained constant (Figure 2.25). For the deep sites, the trend differed. The initial distribution was skewed toward smaller seagrass patches compared to the shallow sites, with a dominance of seagrass patches in the 200–400 cm² size class (Figure 2.25). After one year, the number of seagrass patches in the 200–400 cm² class increased, reaching a peak for this period (Figure 2.25). The number of large size class seagrass patches remained lower than in the shallow sites for the same period. After two years, the number of seagrass patches in the small and intermediate size classes decreased, but there was an increase in the number of seagrass patches within certain large size classes (1600–3200 cm², 3200–6400 cm², and 12,800–25,600 cm²). However, no very large seagrass patches (>25,600 cm²) was observed in the deep sites (Figure 2.25). Overall, larger seagrass patches were more prevalent in the shallow sites and appeared to persist over time, whereas the deep sites exhibited a dominance of smaller seagrass patches (Figure 2.25).

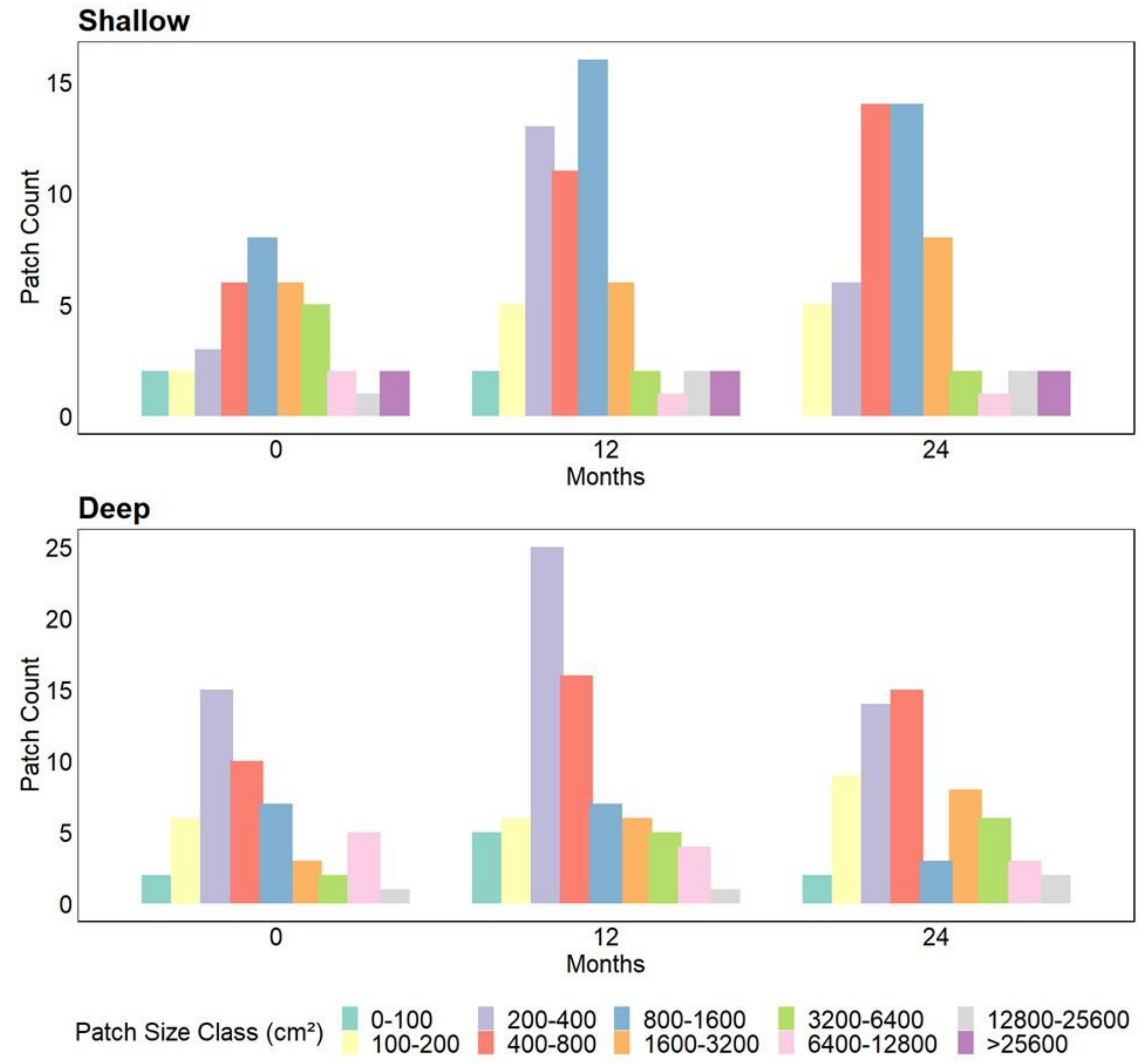


Figure 2.25. *Dynamics of seagrass patch size distribution (number of seagrass patches within each size class, size as cm²) over time for shallow (top) and deep (bottom) sites.*

Finally, the number of storm-fragments was counted within each quadrat. A large disparity in the quantity of storm-fragments among quadrats at different sites was observed (Table S2.3). For instance, a maximum of 25 storm-fragments were recorded in 3% of the quadrats (Table S2.3), while a substantial portion of the quadrats had no storm-fragments at all (Table S2.3). No significant difference in the number of storm-fragments between the three monitoring campaigns was observed ($p = 0.371$; $\chi^2 = 1.981$). However, the difference in the number of storm-fragments was significantly related to the depth of the patch of dead matte, with deeper sites experiencing a significantly lower influx of storm-fragments compared to shallower sites ($p = 0.0003$; $\chi^2 = 12.626$) (Figure 2.26).

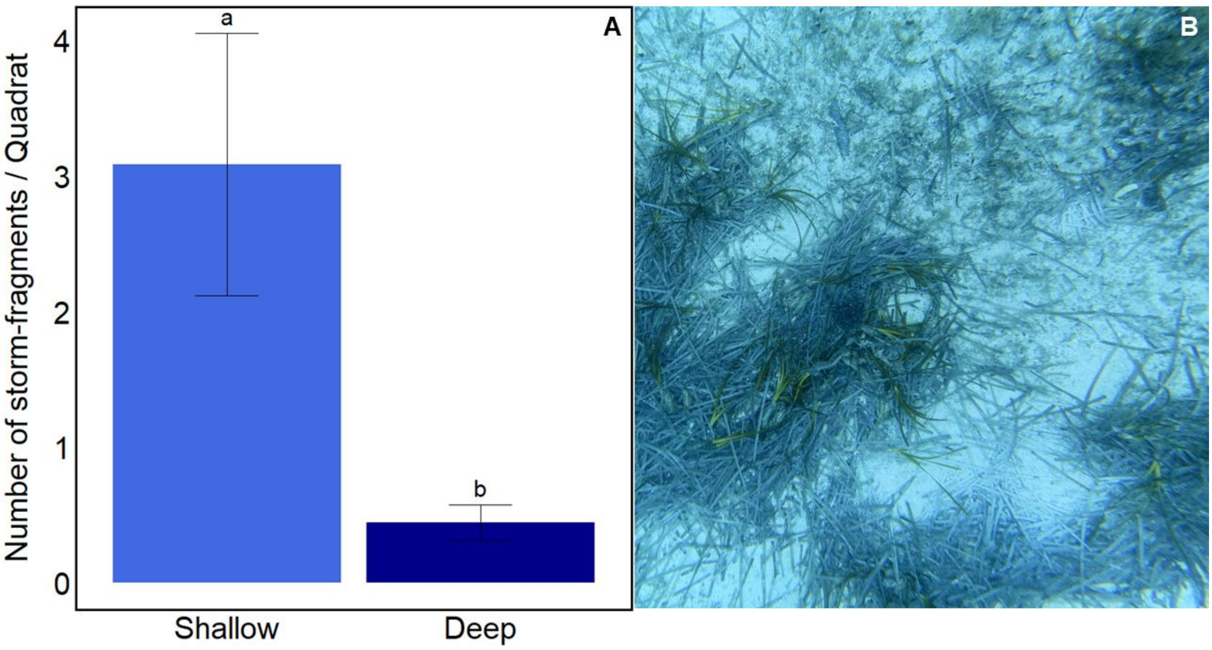


Figure 2.26. *(A) Mean number of storm-fragments accumulation per 9 m² quadrat as a function of depth. Vertical error bars represent standard errors. The statistically significant difference is indicated by different letters (a and b). (B) Important accumulation of storm-fragments on the site AP3 at 20m depth.*

4. Discussion

The establishment of new regulations prohibiting anchoring of large vessels (2016 and 2019), along with an effective reduction in anchoring pressure on the *P. oceanica* meadows and the creation of eco-mooring areas, opens prospects for the ecological restoration of degraded areas. In the past 10 years, there has been a growing number of *P. oceanica* meadow restoration projects throughout the Mediterranean basin (Boudouresque et al., 2021; Pansini et al., 2022), and this trend is set to increase further in the coming years following the adoption of the 'Nature Restoration Law' by the European Parliament in 2024. Most of these restoration projects too rarely evaluate and quantify the dynamics of natural recolonisation at the restored sites (Bockel et al., 2024; Boudouresque et al., 2021). Yet, this data is essential for objectively assessing the cost-benefit ratio of a restoration project compared to natural recolonisation at the study site (Boudouresque et al., 2021).

The natural recolonisation from meadow edges or isolated clumps of *P. oceanica*, as well as the establishment of new patches by storm-fragments, was quite limited in the dead matte areas in this study, which also served as transplantation sites (see Boulenger et al., 2024). Seawater temperature was measured at both shallow and deep sites, as rhizome biomass and length, playing a crucial role in recolonization capacity and speed, are negatively affected by marine heatwaves (MHV) (Pansini et al., 2021; Stipcich et al., 2022b). However, the high temperatures recorded during the summer 2022 MHW (Guinaldo et al., 2023; Marullo et al., 2023) were similar at 20m and 28m depths, suggesting a reduced thermal gradient that may drive differences in *P. oceanica* resilience and recolonization potential between both depths. The depth of the patches of dead matte also showed no significant difference in recolonized area or cover after two years of monitoring, which aligns with findings from other studies that measured natural recolonisation on dead matte at various depths (Abadie et al., 2019; Badalamenti et al., 2011). However, a progressive positive trend in seagrass area is observed for both the shallower sites (35.13% mean progression) and deeper sites (12.90% mean progression). These values are consistent with other recent works conducted in the French Mediterranean, such as the 8-46% progression measured by Marre et al. (2020) or the 5-9.3% progression measured by Bockel et al. (2024). There appears to be an alternation between erosion and recolonisation processes, with erosion being more prominent at shallow sites compared to deep sites, likely due to the reduction in hydrodynamic forces with increasing depth (Vacchi et al., 2012; Uhrin & Turner, 2018). However, colonization remains the dominant process over erosion for both shallow and deep sites, as observed in other studies (Abadie et al., 2019; Bonamano et al., 2021; Marre et al., 2020).

The favorable recolonisation dynamics demonstrate the remarkable resilience of *P. oceanica* exposed to an altered dead matte, with changes in the below-ground chemical processes (Abadie et al., 2016) and microbial communities (Frasca et al., 2024). The analysis of grain size and organic matter content in the anchoring patches monitored in this study showed few differences compared to the adjacent *P. oceanica* meadows. No significant differences were found between shallow and deep dead matte patches. The range of values observed for both median grain size and mud content corresponds to other measurements on matte characterization in the north-western Mediterranean (Serrano et al., 2012). Only the organic matter content differed, with lower organic matter content in the shallow sites compared to the deeps sites, for both *P. oceanica* meadow and dead matte. The lower organic matter content in shallow sites compared to deep sites is primarily driven by increased hydrodynamic activity (Vacchi et al., 2012; Uhrin & Turner, 2018) and temperature-induced organic matter remineralization (Roca et al., 2022; Trevathan-Tackett et al., 2017) has shown by the strong thermal stratification in spring between shallow and deep sites. Moreover, the organic matter content was also significantly higher in the *P. oceanica* meadow compared to the dead matte. *P. oceanica* meadows are known to sequester a significant portion of their primary production within the underlying matte (Boudouresque et al., 2016), making them a substantial reservoir of organic matter due to the recalcitrant nature of *P. oceanica* necromass (Boudouresque et al., 2016; Mateo et al., 1997; Kaal et al., 2018). The decrease in organic matter content within the dead matte is typically linked to erosion after the loss of the canopy, which leads to increased mineralization of previously buried organic material under newly oxygenated conditions (Marbà et al., 2015; Moksnes et al., 2021; Salinas et al., 2020).

It has also been shown that the colonization of dead matte by macroalgae of the genus *Caulerpa* can help preserve sequestered carbon and limit matte erosion by stabilizing the surface layers with their rhizoids (Apostolaki et al., 2022). Additionally, *Caulerpa* species also significantly enrich the sediment with organic carbon through their metabolic production and their ability to trap allochthonous particles (Hendriks et al., 2010; Holmer et al., 2009). However, despite the significant presence of a dense network of *C. cylindracea* (Figure 2.19A) and, to a lesser extent, patches of *C. prolifera* (Figure 2.19B) at the study sites, this does not appear to be sufficient to maintain the original organic matter content in the dead matte as observed in the surrounding *P. oceanica* meadows. It is likely that most of the matte erosion occurs in winter, as both *C. cylindracea* and *C. prolifera* exhibit marked seasonality in leaf area and have a weak rhizoidal structure, leaving the dead matte unprotected during winter storms (Casoli et al., 2021; Hendriks et al., 2010). Nevertheless, the recolonisation rate measured in this study remains far smaller compared to the rates of several m² (Marre et al., 2020), hundreds of m² (Pergent-Martini et al., 2002), or even thousands of m² per year (Boudouresque et al., 2021) reported by some authors. The extreme values presented in the two latter studies both refer to the recolonisation of dead matte at the lower depth limit resulting from poor water quality and increased water turbidity (Boudouresque et al., 2021), a threat that disappeared following the installation of waste-water treatment plants, which restored appropriate environmental conditions (Bockel et al., 2024; Boudouresque et al., 2021; Pergent-Martini et al., 2002). The direct mechanical action of anchoring, as well as trawling, by tearing out plant shoots or sections of the matte, reducing the meadow cover and increasing meadow fragmentation, led to prolonged effects over time and a very slow rate of recolonisation (Abadie et al., 2016, 2019; González-Correa et al., 2005; Kiparissis et al., 2011).

Besides the alteration in the physico-chemical characteristics of the seafloor, the highly fragmented *P. oceanica* meadows, consisting of numerous small patches with low percent cover (<16%), struggle to recover also due to isolation and reduced connectivity between patches. Indeed, the average seagrass patch size across the study sites is small (0.078 m²) which also explain the low recolonisation rates measured in this study. The observed high variability in patch number within quadrats, ranging from 1 to 14 patches, further highlights the fragmented nature of these meadows. Seagrass meadows rely heavily on self-facilitation processes, such as an increase in the density of conspecifics, to enhance survival and promote patch expansion (Almela et al., 2008; González-Correa et al., 2005; Valdez et al., 2020; Vidondo et al., 1997). A fragmented seagrass meadow composed of numerous small patches will initially undergo a period of slow vegetative growth in random directions (Kendrick et al., 2005). In our study, we observed an initial period where small and intermediate-sized patches increased in number in shallow sites, particularly within the 400–800 cm², 800–1600 cm², and 1600–3200 cm² classes. However, after two years, very small patches (0–100 cm²) disappeared, while intermediate-sized patches decreased, and larger patches (1600–3200 cm²) became more dominant, suggesting a gradual coalescence process. With an increase in shoot density and cover, the patches will coalesce into larger units, thus forming a more continuous meadow with a greater spreading rate (González-Correa et al., 2005; Kendrick et al., 2005). This process was particularly evident in shallow sites, where a few patches reached very large sizes (12,800–25,600 cm²). In contrast, deep sites exhibited a different trend, with an initial dominance of smaller patches (200–400 cm²) and a slower transition toward larger patches. Notably, no patches larger than 25,600 cm² were observed at deep sites. Large patches can change local hydrodynamics and create shelter in their surroundings, enhancing the survival of nearby small patches. Additionally, they may produce a greater number of vegetative fragments, boosting the recruitment rate of nearby patches (Almela et al., 2008). The recolonisation process in the deep sites occurs almost exclusively through the progression of patch edges, but many storm-fragments were observed in the shallower sites. These fragments could serve as a source for the establishment of new patches, potentially accelerating long-term recolonisation. This hypothesis was also proposed by Pergent-Martini et al. (2022) to explain the high recolonisation rates observed in their study. Meinesz and Lefèvre (1983) determined that the establishment frequency of storm fragments is 3/ha/year, which represents a minimal contribution to recolonisation compared to meadows margins progression, even when considering the low recolonisation rates measured in our study. Indeed, most storm-fragments lack roots, and the dead matte offers little structural complexity to allow these fragments to be trapped, remain stable in one place and develop (Badalamenti et al., 2011). However, studies on natural recolonisation on calcareous rubbles show a significant contribution of storm-fragments to natural recolonisation, as the crevices between adjacent rubbles offer a pattern of substrate complexity enabling the trapping and persistence of *P. oceanica* vegetative fragments (Almela et al., 2008; Badalamenti et al., 2011; Di Carlo et al., 2005). Regarding sexual recruitment through seed germination and seedling establishment, it predominantly occurs in sheltered areas, at shallower depths than the study sites and more frequently on rocky substrate than dead matte (Balestri et al., 2017; Balestri & Lardicci, 2008; Piazzi et al., 1999).

Implications for Restoration

The overall level of fragmentation in the meadow is a key factor in determining the vulnerability of seagrass meadows (Barcelona et al., 2021). Due to climate change, the increasing frequency and severity of storms (Masson-Delmotte et al., 2019) will impact the integrity of *P. oceanica* meadows, especially fragmented meadows in exposed locations (Marco-Méndez et al., 2024). Additionally, climate-induced rises in sea temperature and sea level, which affect light availability, also threaten the resilience of seagrass meadows at upper and lower depth limits (Pergent et al., 2014). Given the very slow natural recolonization observed in our study, it is crucial to emphasize the fundamental importance of protecting *P. oceanica* meadows from anthropogenic activities to prevent further degradation. Moreover, conservation measures and new regulations must be implemented across the entire Mediterranean basin to prevent the displacement of anchoring-related degradation between countries with differing protection laws. Furthermore, active restoration through the transplantation of cuttings (e.g. Boulenger et al., 2024; Mancini et al., 2022) or seedlings (e.g. Mancini et al., 2024; Zenone et al., 2025) can serve as a valuable tool to reduce fragmentation and facilitate the recovery of highly fragmented seagrass meadows, ensuring their resilience in a changing climate. Restoring *P. oceanica* meadows also helps limit the degradation of the dead matte and the mineralization of organic matter, thereby preserving the integrity of the dead matte—a factor that is particularly important for climate change mitigation (Pergent-Martini et al., 2021). Our study highlights the differing recolonisation dynamics between shallow and deep dead matte patches. The insights gained can inform the design of effective restoration plans tailored to local conditions, which we stress as essential for any restoration project. Given the fluctuations between recolonisation and erosion processes at shallow sites, we suggest that transplantation of cuttings should use anchoring structures with a certain level of resistance to local hydrodynamics (Heide et al., 2021; Temmink et al., 2020). Additionally, transplantation should be carried out at high density in relatively large planting units to benefit from the mutual sheltering effect (Valdez et al., 2020; van Katwijk et al., 2016). Considering the relatively high accumulation of storm-fragments in shallow areas, it would also be beneficial to use structures that facilitate their trapping, such as gabions with ten-centimeter gaps between rocks (Badalamenti et al., 2011; Di Carlo et al., 2005). This approach could support natural recolonisation without the need for transplantation or serve as a complementary strategy. A different restoration design should be applied for deeper sites. The limited erosion rate compared to the recolonisation rate suggests that the type of anchoring structures used for transplanted cuttings will have little impact, making more cost-effective solutions, such as the use of iron staples (Mancini et al., 2021), a suitable alternative. The very low quantity of storm fragments in deep sites suggests that using trapping substrates to promote their retention and establishment would be ineffective.

5. Conclusion

This study assessed the recovery dynamics of *P. oceanica* in areas damaged by anchoring at shallow and deep sites in North-Western Corsica. The findings highlight clear differences in recolonisation patterns between shallow and deep sites. Shallow patches of dead matte exhibited more dynamic processes, with alternating erosion and recolonization, while deeper patches of dead matte showed slower yet steadier recovery. The high presence of storm-fragments in shallow areas suggests their potential in aiding recovery by establishing new seagrass patches. However, despite these dynamics, recolonisation remained limited, likely due to alterations in the dead matte's physico-chemical characteristics, such as reduced organic matter content compared to adjacent meadows. This underscores the inherent challenges in the natural recovery of *P. oceanica* meadows and the importance of protecting those meadows against anthropogenic pressures to prevent further degradation. For seagrass meadows that are already heavily damaged and fragmented, ecological restoration offers a viable solution to facilitate the recovery of degraded areas. The findings stress the importance of incorporating site-specific factors into restoration efforts. At shallow sites, where erosion and fragment accumulation are more pronounced, restoration designs could incorporate structures that facilitate fragment trapping and offer stability against hydrodynamic forces. In contrast, deeper sites, characterized by less erosion and very little storm-fragments accumulation, may benefit from simpler and more cost-effective approaches like the use of iron staples for cuttings' transplantation. Across all depths, ensuring the connectivity between seagrass patches and promoting self-facilitation processes will be critical to accelerate recovery. This research underscores the need to assess and monitor natural recolonisation processes before implementing active restoration measures. By aligning restoration strategies with the specific environmental conditions and recolonisation capacities of degraded areas, it is possible to improve restoration success and long-term resilience of *P. oceanica* meadows.

Comparative assessment of transplantation methods and donor sources for the restoration of Posidonia oceanica meadows

Arnaud Boulenger[a,b], Michel Marengo[b], Pierre Boissery[c], Sylvie Gobert[a,b]

[a] Laboratory of Oceanology, MARE Centre, UR FOCUS, University of Liege, 11 allée du six août, 4000, Liege, Belgium

[b] STAtion de REcherche Sous-marines et Océanographiques (STARESO), 20260 Calvi, France

[c] Agence de l'Eau Rhône Méditerranée Corse, 2, street Henri Barbusse, CS 90464, 13207 Marseille Cedex 01, France

Science of the Total Environment (2025)

Abstract

The restoration of coastal ecosystems, especially seagrass meadows, has become a key priority to support the recovery of ecosystem services. In the Mediterranean Basin, although many projects have been carried out to restore *Posidonia oceanica* meadows over the past 50 years, major knowledge gaps persist. This study is the first to simultaneously compare two donor sources, storm fragments versus donor meadow cuttings, and three sustainable transplantation methods. This three-year experiment involved transplanting 693 cuttings using three distinct transplantation methods (iron staples, coconut fiber mats, and BESE elements) in shallow (20 m) and deep (28 m) dead matte areas of Calvi Bay (Corsica, NW Mediterranean). Performance was assessed through survival, shoot production, leaf and root morphological traits, with particular attention given to root systems development, a critical but often overlooked component in seagrass restoration studies. Storm-fragments performed comparably to donor meadow cuttings, supporting their use as a sustainable, non-destructive source of planting material. Among transplantation methods, iron staples led to the best performance across survival, root development, and cost-efficiency. BESE elements ensured high survival but limited root development, while coconut fiber mats performed poorly overall. Despite encouraging survival rates (>80% under optimal conditions), significant differences in leaf and root traits remained between transplants and natural meadows after 36 months, suggesting incomplete ecological recovery. This comparative approach provides a critical first benchmark for evaluating the feasibility, performance, and economic viability of different restoration techniques in *P. oceanica* meadows.

Keywords

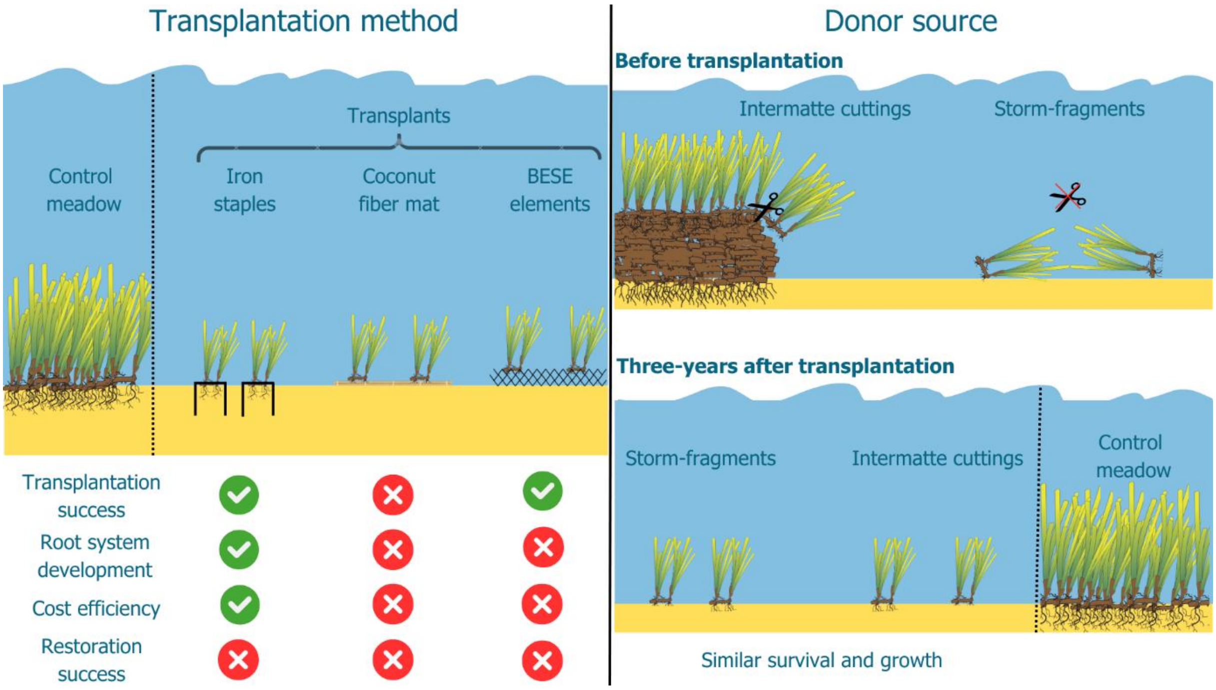
Seagrass, Anchoring, Survival, Root system, Structural complexity

AUTHORS’ CONTRIBUTIONS

Arnaud Boulenger: Conceptualization, Field sampling, Methodology, Formal analysis, Investigation, Writing, Visualization; Michel Marengo: Conceptualization, Methodology, Writing, Supervision, Funding; Pierre Boissery: Methodology, Writing, Funding; Sylvie Gobert: Conceptualization, Methodology, Investigation, Writing, Supervision, Funding.

ACKNOWLEDGMENTS

This work was supported by the University of Liege (grant FSR2021) and the Fonds National de la Recherche Scientifique—FNRS (grants ASP 40006932 and CDR J.0076.23). This study is part of the STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) project funded by the Territorial Collectivity of Corsica and by the Rhone-Mediterranean and Corsican Water Agency. Authors are grateful to the STARESO for facilities and field assistance. Authors are grateful to Karin Didderen and Wouter Lengkeek from BESE Ecosystem Restoration (Culemborg, the Netherlands) for designing the experimental set-up and field assistance for transplantation.



	Iron staples	Coconut fiber mat	BESE elements
Transplantation success	✓	✗	✓
Root system development	✓	✗	✗
Cost efficiency	✓	✗	✗
Restoration success	✗	✗	✗

Graphical abstract used for publication.

1. Introduction

In February 2024, the European (EU) Parliament adopted the *Nature Restoration Law*. Under this legislation, EU member states are required to restore at least 30% of terrestrial and marine habitats in poor condition by 2030, 60% by 2040, and 90% by 2050. Over the past centuries, many human activities, such as land reclamation, coastal development and water pollution, have significantly contributed to the degradation of European coastal habitats (Airoldi & Beck, 2007). This is especially true for the extensive seagrass meadows formed by *Posidonia oceanica* (L.) Delile, which have experienced a well-documented regression throughout the entire Mediterranean basin (Boudouresque et al., 2009; de los Santos et al., 2019; Telesca et al., 2015). These meadows, growing from the surface down to an average 40 m depth, are of considerable ecological and economical importance because of the multitude of ecosystem services they provide such as nursery (Campagne et al., 2014), carbon sink (Monnier et al., 2022; Pergent-Martini et al., 2021) and protection against coastal erosion (Gacia et al., 1999; Gacia & Duarte, 2001). *P. oceanica* meadows are impacted by anthropogenic pressures (Boudouresque et al., 2009; Giakoumi et al., 2015) either indirectly through degradation of water quality (Bockel et al., 2024; Montefalcone et al., 2007) or directly through habitat destruction, such as coastal development (Holon et al., 2015; Descamp et al., 2025), trawling (Kiparissis et al., 2011) or anchoring (Abadie et al., 2016, 2019). Land-based pollution is one of the major anthropogenic threats to coastal ecosystems (Halpern et al., 2008; Holon et al., 2015; Micheli et al., 2013). Wastewater discharges contribute to eutrophication by increasing nutrient and organic matter loads, which reduce water clarity, stimulate algal blooms, and cause sediment accumulation. These processes limit light penetration and ultimately compromise seagrass growth and survival (Waycott et al., 2009). Since 1991, the European Urban Wastewater Treatment Directive (91/271/CEE) has established water quality standards to safeguard receiving ecosystems and required member states to implement action plans to achieve these targets. This directive has since led to substantial improvements in wastewater collection and treatment infrastructure in France, reducing untreated discharges and benefiting *P. oceanica* meadows, particularly at their lower depth limits (Bockel et al., 2024; Boudouresque et al., 2021; Pergent-Martini et al., 2002). Moreover, mechanical damage from anchoring by large recreational boats (hull length > 24 m) (Abadie et al., 2016; Montefalcone et al., 2008; Pergent-Martini et al., 2022) is of growing concern due to the increasing popularity of recreational boating in recent decades (Cappato et al., 2011; Carreño & Lloret, 2021). Anchoring in *P. oceanica* meadows causes both direct and indirect harm through the deployment and retrieval of anchors and the dragging of chains and ropes along the seabed (Milazzo et al., 2004). Repeated anchoring ultimately leads to widespread degradation of the meadows (Abadie et al., 2015; Montefalcone et al., 2006; Pergent-Martini et al., 2022). Although the Mediterranean Sea represents less than 1% of the global ocean surface, it receives more than half of the world's fleet of large recreational vessels, especially during the summer months (Cappato et al., 2011; Carreño & Lloret, 2021). This activity is predominantly concentrated in the northwestern Mediterranean basin (Pergent-Martini et al., 2022).

In mainland France and Corsica, *P. oceanica* meadows are the marine habitat most impacted by anchoring pressure (Deter et al., 2017). In 2016, French authorities implemented a regulation relative to the anchoring of the largest vessels (>80m) (French Naval Prefecture, Decree No. 155/ 2016), reinforced by a new regulation in 2019 prohibiting any anchoring within seagrass meadows for boats longer than 24 m (French Naval Prefecture, Decree No. 123/2019). Although a reduction in large boat anchoring in *P. oceanica* meadows has been observed following the enforcement of these regulations (Fontaine et al., 2024; Bockel et al., 2023), the slow growth rate of *P. oceanica* rhizomes (a few centimetres per year; Caye, 1980) means that natural recolonization of damaged areas is a process that will takes decades or even centuries (Abadie et al., 2019; Boulenger et al., 2025a). To accelerate the recovery of degraded *P. oceanica* meadows and their associated ecosystem services, active restoration measures such as the transplantation of cuttings have been implemented in areas where the source of degradation has been removed or mitigated (Boudouresque et al., 2021). A wide range of anchoring or stabilization techniques have been employed for *P. oceanica* transplantation, including heavy structures such as concrete frames (Bacci et al., 2024; Cooper, 1982), as well as lighter alternatives using metallic, plastic, or biodegradable meshes and wires (De Luca, 2025; Gobert et al., 2005; Piazzi et al., 2021), or cost-effective devices like stakes and staples (Castejon-Silvo & Terrados, 2021; Mancini et al., 2021). Both grid-based systems (plastic, wire, or natural fibers) and individual anchoring methods (metallic or biodegradable staples and pegs) have generally produced good transplantation outcomes (Calvo et al., 2021 ; Genot et al., 1994 ; Mancini et al., 2021; Molenaar & Meinsez, 1995 ; Piazzi et al., 2021; Scannavino et al., 2014), with some techniques proving effective over the long term (Pirotta et al., 2015). Despite increasing experimental trials of *P. oceanica* transplantation over the past decade, significant knowledge gaps remain (Boudouresque et al., 2021; Pansini et al., 2022; Pergent-Martini et al., 2024). These include the need to test different sustainable anchoring methods to attach the cuttings to the seafloor, as well as different donor sources of plant material for transplantation, across a variety of environmental conditions (Boudouresque et al., 2021; Pansini et al., 2022; Pergent-Martini et al., 2024). This study aimed to address these gaps by : (1) Testing biodegradable substrates of varying structural complexity to facilitate transplants' anchoring; (2) Comparing the performance of transplants from two donor sources: fragments of *P. oceanica* rhizomes either harvested from natural meadows and fragments of unknown origin that are found drifting on the seafloor; and (3) transplanting at two different depths (20 and 28 m), corresponding to the bathymetric zones where significant degradation due to anchoring (Abadie et al., 2015) and wastewater discharge (Bockel et al., 2024) is often found.

2. Material and Methods

2.1 Study area

This study was conducted in the sub-bay Alga Bay (8°43′52′′E; 42°34′20′′N), located within Calvi Bay in northwestern Corsica in the northwestern Mediterranean basin. Alga Bay, situated nearby the STARESO research station, covers an area of 1 km² and is colonized by a *P. oceanica* meadow that spans 0.78 km² of the seafloor (Abadie et al., 2016) (Figure 2.27). Prior to the enforcement of the Decree No. 123/2019, this bay experienced several decades of intensive anchoring activity, which resulted in many anchoring scars within the seagrass meadows, corresponding to the abrasion caused by anchor removal (Abadie et al., 2019). However, since the implementation of the new regulation, anchoring by leisure boats (>20 m long) in *P. oceanica* meadows has decreased by 57% in Calvi Bay between 2019 and 2023 (Fullgrabe et al., 2024). The selection of experimental sites followed the decision-making strategy for transplanting *P. oceanica* and other seagrasses proposed by Boudouresque et al. (2021). Seven patches of dead matte, resulting from previous anchoring damage, were selected as experimental sites (Figure 2.27). Three sites were located at an average depth of 20 m (AP1 – AP3, hereafter referred to as "shallow" sites) and four at an average depth of 28m (AP4 – AP7, hereafter referred to as "deep" sites).

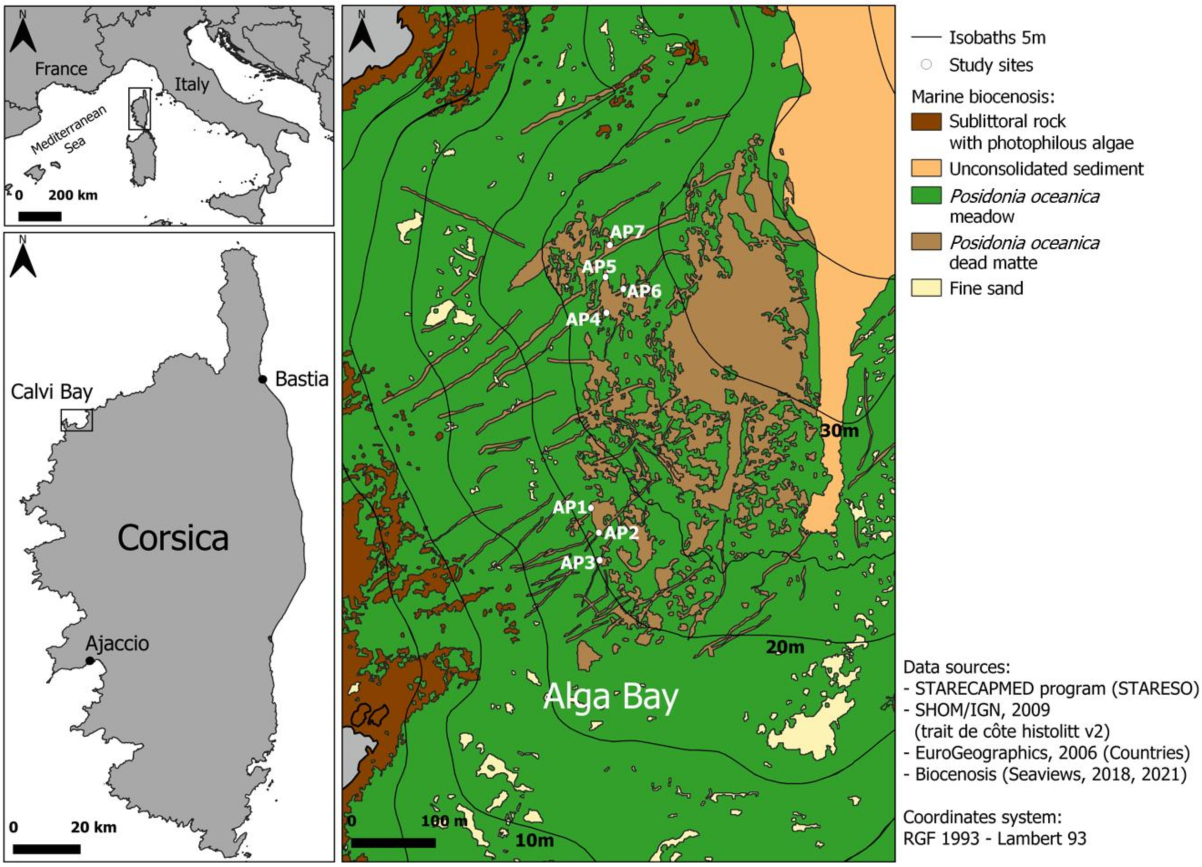


Figure 2.27. *Location of the study area. The top left figure shows a wider view of Corsica Island in the Mediterranean Sea. The bottom left figure displays a more detailed view of Corsica and the location of Calvi Bay. The figure on the right shows Alga Bay with associated marine biocenosis, the isobaths every 5m depth and the transplantation sites (AP1 – AP7). Figure modified from Boulenger et al. (2025).*

2.2 Seagrass transplantation

2.2.1 Transplantation methods

The attachment of cuttings to the seabed is a critical step for the success of seagrass transplantation. Although several methods have been tested in previous studies and proven to be effective, only a subset are considered environmentally sustainable (Bacci & La Porta, 2021; Boudouresque et al., 2021). In this study, three different biodegradable anchoring methods were tested with the aim of ultimately leaving only the natural ecosystem once the transplants have developed a sufficiently robust root system. The first method used U-shaped iron staples (Figure 2.28A), which offered the least protection against hydrodynamic forces. Each staple consisted of a 10 cm straight horizontal section and two 30 cm vertical arms designed to be inserted into the dead matte. Each staple had a thickness of 3 mm. The second anchoring method, commonly used in terrestrial ecological engineering (e.g., for riverbank or dune restoration) (Piazzi et al., 2021), employed a biodegradable mat made of natural coconut fiber, woven into an H2M5 mesh weighing 740 gr/m² (Ecobiotex, Thizy Les Bourgs, France) (Figure 2.28B). The mesh size was 9x9 mm, and the mat had a thickness of 5 mm. The third method used BESE elements (BESE Ecosystem Restoration Products, Culemborg, The Netherlands), composed of biodegradable potato-waste-derived Solanyl C1104M (Rodenburg Biopolymers, Oosterhout, the Netherlands) (Figure 2.28C). This Solanyl biopolymer is officially certified as biodegradable (see Figure S2.2), and it gradually degrades under field conditions over 5 to 10 years, depending on the local environmental conditions (Nitsch et al., 2021). Individual sheets (91.0 x 45.5 x 2.0 cm; 0.44 kg, surface-to-volume ratio 80 m^2/m^3) can be stacked together to form a modular 3D-structure (Figure 2.28C). In this study, three sheets were combined to form a 6-cm high 3D honeycomb-shaped matrix allowing for the expansion of seagrass rhizomes and roots through the structure.

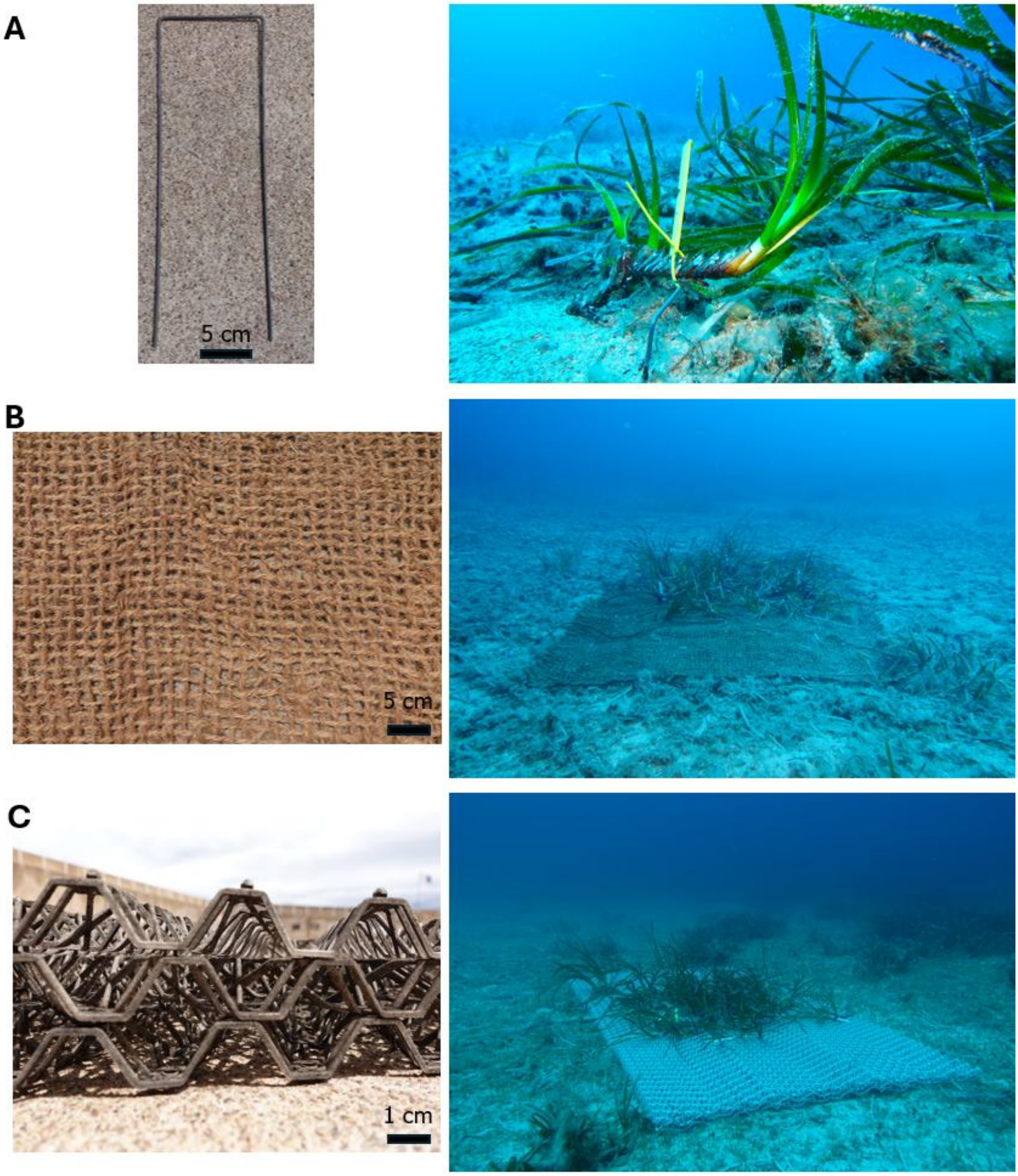


__Figure 2.28.__ The three transplantation methods used in this study: (A) Iron staple, (B) coconut fiber mat and (C) BESE element.

The coconut fiber mats and the BESE elements offer greater structural complexity than the staples. Therefore, we hypothesized that they could facilitate transplant anchoring and enhance natural recruitment by trapping drifting fragments and/or seedlings (Irving et al., 2014; Wear et al., 2010). These methods draw inspiration from the ecological succession theory, wherein pioneer species create a network of roots and rhizomes that capture drifting cuttings and promote their attachment to the substrate (Molinier & Picard, 1952) (Figure S2.3).

2.2.2 Experimental design and field transplantation

A major limitation in *P. oceanica* restoration efforts is the limited availability of planting material, which poses a significant obstacle to large-scale meadow restoration initiatives. To minimize the impact on existing meadows, the use of naturally detached fragments from *P. oceanica* meadows offers a promising, non-destructive alternative to harvesting cuttings directly from donor meadows (Balestri et al., 2011) (Figure 2.29A). Indeed, large quantities of seagrass fragments (hereafter referred to as storm-fragments) of various morphologies are naturally uprooted during storms events (Ewanchuk & Williams, 1996). There is evidence that *P. oceanica* storm-fragments can colonize new habitats, form new patches, and expand clonally over time (Boudouresque & Meinesz, 1982; Boudouresque et al., 1990; Almela et al., 2008; Di Carlo et al., 2005). However, little is known about the performance of those storm-fragments drifting on the seafloor, in terms of survival and growth, compared to the use of cuttings directly extracted from natural meadows (Balestri et al., 2011; Boulenger et al., 2024). Since this comparison is essential for developing ecologically sustainable restoration strategies, both types of cuttings were included in this study (Figure 2.29). The majority of the cuttings (462 fragments) consisted of storm-fragments collected from natural accumulation areas located in natural sandy intermattes within *P. oceanica* meadows (Abadie et al., 2015; Gobert et al., 2016). These were collected during SCUBA dives at depths ranging from 6 to 28 m (Figure 2.29A). A smaller portion (231 fragments) of the total cutting pool was manually excised from the erosion edges of natural sandy intermattes at 15 m depth near STARESO (Gobert et al., 2016) (Figure 2.29B). These intermatte cuttings were specifically included to enable comparison of survival rates and morphological traits between the two donor sources. The harvesting of those cuttings from the erosion edges of natural sandy intermattes was primarily guided by sustainability considerations, aiming to minimize the disturbance to the core of healthy donor meadows. This approach aligns with previously published observations indicating that erosion edges naturally contribute to the production of storm-fragments, as vertical matte notches erode and release pieces of rhizomes (Gobert et al., 2016). Moreover, *P. oceanica* shoots collected from erosion edges do not exhibit significantly lower photosynthetic activity, leaf surface area or leaf biomass compared to those from continuous meadows at 15m depth (Abadie et al., 2017; Lapeyra et al., 2016). Furthermore, erosion edges tend to be dominated by plagiotropic rhizomes (up to 60%; Lapeyra et al., 2016), which are particularly suitable for transplantation due to their horizontal growth form and more rapid growth rates compared to orthotropic rhizomes (Molenaar, 1992; Molenaar & Meinesz, 1995). All fragments (both storm-derived and intermatte cuttings) were harvested within Calvi Bay, in close proximity to the transplantation sites, to ensure the preservation of local genetic integrity.

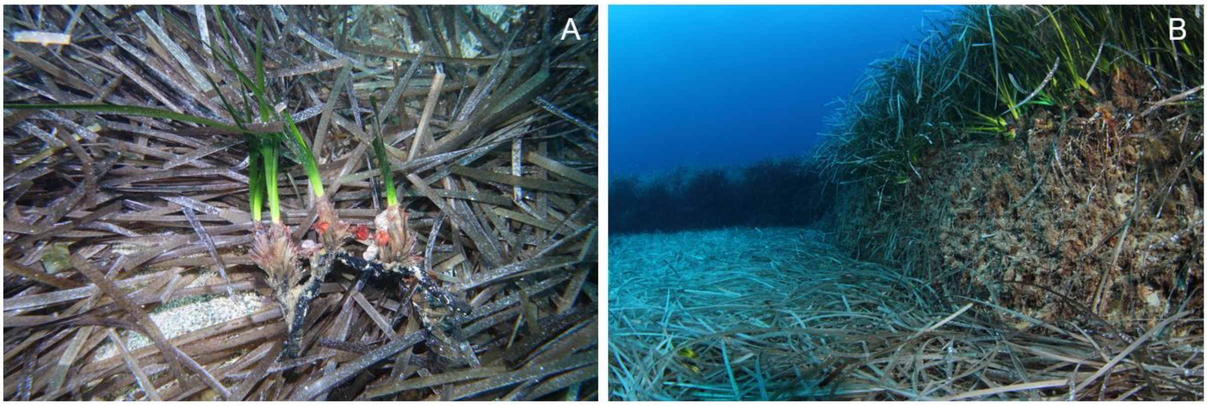


Figure 2.29. *The two donor sources used in this transplantation pilot study: (A) Storm-fragment laying on the seafloor, and (B) erosion edge of a natural sandy intermatte (©STARESO/Arnaud Abadie).*

The cuttings were maintained in outdoor flow-through seawater aquaria until their initial selection, which was based on rhizome length, number of shoots and visually assessed health condition (leaf necrosis and biting marks). Only cuttings bearing at least 3 shoots and a plagiotropic rhizome of at least 15 cm in length were retained, while those exhibiting excessive leaf necrosis were excluded. For each planting unit type (i.e. iron staples, cononut fiber mats and BESE elements) at each experimental site, 33 cuttings were attached using cable ties, comprising 22 storm-fragments and 11 intermatte cuttings. The cuttings were transplanted close together, with rhizomes spaced approximately 10 cm apart, a configuration shown to promote optimal survival and growth (Molenaar & Meinesz, 1995). The planting units were installed approximately 2 m apart from each other. In total, each experimental sites contained 99 transplants, resulting in an overall total of 693 *P. oceanica* transplants. All transplanted cuttings were labelled with small tags attached around the rhizomes by means of cables ties to allow for the monitoring of their survival over time.

2.3 Sampling strategy and morphological traits measurements

Before the start of the transplantation work, , 20 cuttings, including both storm-fragments and cuttings from intermattes, were preserved for biometric measurements and further laboratory analyses. In addition, 20 *P. oceanica* fragments were collected from nearby reference meadows at depths of 20 m and 28 m. This sampling provided a T0 baseline, allowing comparisons between the cuttings and the control meadows prior to transplantation. Following transplantation, six monitoring campaigns were conducted at 3, 12, 15, 24, 27, and 36 months post-transplanting. These campaigns were carried out three times in spring (April-June; 12, 24 and 36 months post-transplanting) and three times in fall (September-October; 3, 15 and 27 months post-transplanting). During each monitoring campaign, the total number of foliar shoots per planting unit was recorded. Survival rate was assessed based on the presence of at least one living shoot per rhizome. Survival data were recorded in a binary format, with (1) indicating a living transplant and (0) indicating a dead one. At each experimental site (n=7), 12 shoots were collected from transplants for biometric measurements, resulting in a total of 84 sampled shoots per campaign. Additionally, 10 control shoots were collected from surrounding meadows at 20 m and 28 m depths and brought back to the laboratory for further examination. All the shoots were sampled using the Non-Destructive Shoot sampling Method (NDSM) as recommended by Gobert et al. (2020). For each sampled shoot, the number of leaves was counted, and the length and width of each leaf were measured. Epiphytes were scraped from all leaves using a ceramic scalpel blade (Dauby & Poulicek, 1995). The leaves were then oven-dried at 60°C for 48 h and weighed to determine their dry biomass.

Furthermore, 36 months after transplantation, six cuttings per experimental site (n=42) were harvested for rhizomes and root morphological traits measurements. The samples were carefully excavated using small hand tools to preserve the integrity of the root systems. The same procedure was applied to five *P. oceanica* fragments collected from the control meadow at 20 m depth and five from the control meadow at 28 m depth. In the laboratory, the collected plants were gently rinsed with seawater to remove fine sediment particles from the root systems. For each sampled individual, the number of primary and lateral roots was counted. The following morphological traits were measured: maximum width (i.e. maximum horizontal spread) and maximum rooting depth (i.e. maximum root length) of the root system, as well as the length of the plagiotropic rhizome (Figure S2.4).

Roots were arranged on grid paper and photographed. The resulting images were analysed using the SmartRoot plugin in ImageJ software version 1.8.0_345 (Lobet et al., 2011). Image data were then used to calculate the total root length of each individual root system. Subsequently, the entire root system was oven-dried for 48 h at 60°C and weighed to determine dry biomass. To account for variation in rhizome lengths among samples, all measurements were normalized to the length of the individual rhizome. Accordingly, the number of primary and lateral roots is expressed per centimetre of rhizome (cm^{-1}), biomass is reported as grams of dry weight per centimetre of rhizome ($gDW.cm^{-1}$), and total root length, maximum horizontal spread, and maximum rooting depth are expressed as dimensionless ratios.

2.4 Cost-efficiency analysis

To identify the most cost-effective transplantation method and assess its potential for upscaling, a cost-efficiency analysis was conducted. Specifically, the planting cost per unit area (€.m^{-2}) was calculated based on the unit price and dimensions of the three materials tested. A planting density of 22 transplants per m^2 was considered, in accordance with the experimental design. This surface-based cost enables direct comparison between the different transplantation materials, assuming an equal number of cuttings transplanted over the same surface area. To account for the effectiveness of each transplantation method, the cost per planted surface area was adjusted by incorporating the survival rate associated with each technique. Assuming an initially planted area of 1 m^2, the remaining area 36 months post-transplantation reflects the survival rate and represents the effectively restored surface. Accordingly, the initial cost per planted m^2 was divided by the survival rate to obtain the cost per m^2 of *P. oceanica* meadow effectively restored after 36 months.

2.5 Data analysis

To assess the effects of the different experimental treatments on the survival of cuttings, the number of shoots per planting unit, and leaf morphological traits, Generalized Linear Mixed Models (GLMMs) were used. These models are well suited for handling discrete, non-negative data such as count data, and allow the inclusion of experimental site as random factor to account for variability among the seven sites (Figure 2.27). A binomial distribution was used for the response variable survival rate. Fixed factors included in the GLMMs were 'Transplantation method' (three levels: iron staple, coconut fiber mat, and BESE element), 'Donor source' (two levels: intermatte cutting and storm-fragment), 'Bathymetry' (two levels: shallow and deep), and 'Months post-transplanting' (six levels: 3, 12, 15, 24, 27, and 36 months). For the response variable average number of shoots per planting unit, a negative binomial distribution was used and the fixed factors used were 'Transplantation levels', 'Bathymetry' and 'Months post transplanting'. A Poisson distribution was used for the number of leaves per shoot, and a Gamma distribution with a log link function was used for the leaf surface area, the maximum leaf length and the dry weight. The same set of fixed factors as in the survival rate model was included in the GLMMs for these leaf morphological traits. Because the experimental sites were nested within the bathymetric levels, a nested random structure (1|Bathymetry/Site) was specified in the GLMMs. GLMMs were built using the *glmer* function from the *lme4* package in RStudio software version 4.3.2 (RStudio Inc., Boston, MA, USA). Model selection was guided by Akaike's Information Criterion (AIC), progressively removing non-significant terms based on statistical criteria until no further variables could be eliminated.

Overdispersion was assessed by comparing the residual deviance to the residual degrees of freedom. To test the statistical significance of differences between treatments, estimated marginal means (EMMs) were computed using the *emmeans* function in RStudio, applying Bonferroni correction to adjust p-values for multiple comparisons.

To compare the leaf morphological traits of the cuttings with the control plants, the normality and linearity of the residuals were assessed by visually inspecting the residuals versus fitted values plot and performing a Shapiro-Wilk test, while the homogeneity of variances was checked using Levene's test. Data assumption checking was conducted using RStudio software. Since the data were not normally distributed, PERMANOVAs were used to compare the transplants to the control meadows over time. PERMANOVAs were computed using the fixed factors 'Transplantation effect' (two levels: control meadow, transplant), 'Months post-transplanting' and their interaction. The root morphological traits were assessed only 36 months after transplanting and compared between transplants and control meadows. PERMANOVAs were also computed for the following root morphological traits: number of adventitious roots, maximum horizontal spread, maximum rooting depth, total root length, and total root biomass (dry weight). Since the factor 'Donor source' and the interaction with 'Transplantation method' and 'Bathymetry' were not significant for any of the root morphological traits, they were excluded from the PERMANOVA design to reduce the number of interactions and model complexity. Therefore, the final PERMANOVA design for the root morphological traits included the fixed factors 'Transplantation method and effect' (four levels: iron staple, coconut fiber mat, BESE element, and control meadows) and 'Bathymetry'. Prior to running the PERMANOVAs, a resemblance matrix based on Euclidean distances was calculated on untransformed data. The effects of the factors on each response variable were assessed using permutation tests applied to the residuals of a reduced model, with analyses based on Type III partial sums of squares. The number of permutations was set to 999 and Monte Carlo tests were performed when the number of permutations was fewer than 100 (Anderson et al., 2008). PERMANOVAs were performed using the PRIMER-E+PERMANOVA software version 7.0.24 (PRIMER-E, Auckland, New Zealand). All the differences were considered statistically significant when $p < 0.05$. All values were reported as mean ± standard error.

3. Results

3.1 Survival rates and shoot counts

The survival rate of *P. oceanica* cuttings transplanted in spring 2022 was monitored over a 36-month period and revealed contrasting responses depending on the experimental treatments. First, the donor source of the cuttings had no significant effect on survival (Figure S2.5), nor did any of its interactions with the three other experimental factors. As a result, this factor was excluded from the final GLMM. Neither the transplantation method nor the bathymetry showed a significant main effect on survival rate. However, significant interaction effects were detected between transplantation method and time since transplanting ($p < 0.001$; $F = 3.85$), as well as between transplantation method and bathymetry ($p < 0.001$; $F = 10.30$) (Figure 2.30A). Finally, time since transplanting had a strong and significant effect on survival rate ($p < 0.001$; $F = 59.52$). Thirty-six months after transplantation, survival rates revealed marked contrasts, with higher survival observed at deep sites compared to shallow ones (Figure 2.30A). At shallow sites, cuttings fixed to BESE elements exhibited significantly higher survival rates than those fixed with iron staples and coconut fiber mats ($p = 0.020$ and $p < 0.001$, respectively). Survival was also significantly different between cuttings attached with iron staples and those with coconut fiber mats ($p < 0.001$) (Figure 2.30A). At deep sites, survival rates were significantly higher for BESE elements compared to coconut fiber mats ($p < 0.001$), but no significant difference was found between BESE elements and iron staples (Figure 2.30A). However, iron staples resulted in significantly higher survival than coconut fiber mats ($p < 0.001$) (Figure 2.30A). Thirty-six months post-transplanting, the highest survival rate was recorded for iron staples at deep sites (82.58 ± 0.03%), while the lowest was observed for coconut fiber mats at shallow sites (31.31 ± 0.05%) (Figure 2.30A).

The total number of shoots per planting unit was monitored across all experimental sites for the entire 36-months monitoring period. The number of months post transplanting had a significant effect on the total number of shoots per planting unit ($p < 0.001$; $F = 33.1944$), as did the interaction between transplantation method and months post transplanting ($p < 0.001$; $F = 4.1121$) (Figure 2.30B). In contrast, bathymetry had no significant influence on the total number of shoots per planting unit. Regardless of the transplantation method used, a general decline in the total number of shoots per planting unit was observed over time (Figure 2.30B). At 36 months post transplanting, shoot counts were significantly higher on BESE elements and iron staples compared to coconut fiber mats ($p < 0.001$ for both). However, there was no significant difference in shoot numbers between BESE elements and iron staples.

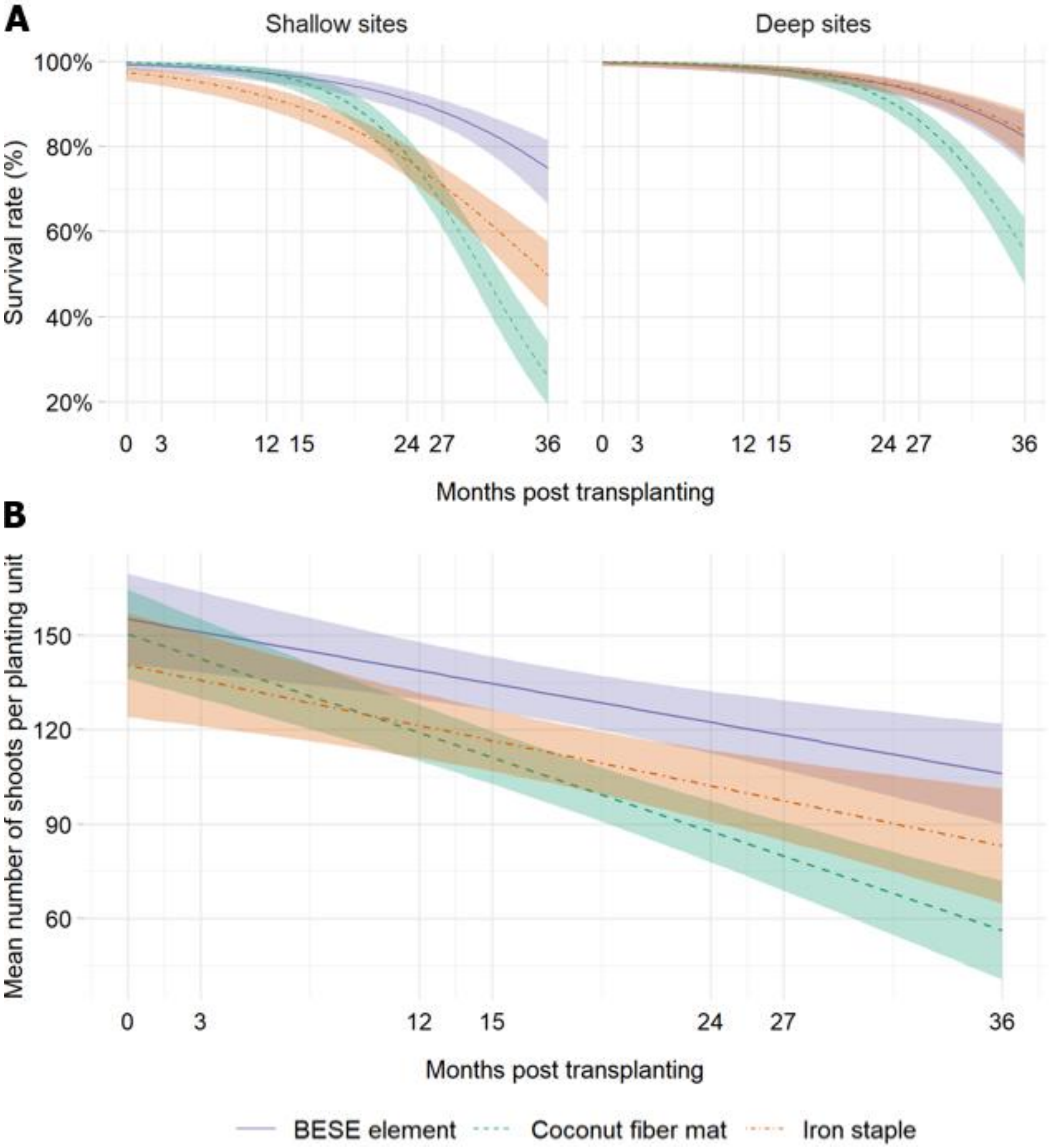


Figure 2.30. *Temporal dynamics of (A) transplanted cuttings' survival rates according to transplantation method at shallow and deep sites and (B) mean number of shoots per planting unit according to transplantation method. Shaded areas around the curves represent 95% confidence intervals.*

3.2 Leaf morphological traits

GLMMs were performed to assess the influence of transplantation method, donor source, bathymetry and months post transplanting on the leaf morphological traits of the *P. oceanica* transplants. For both the number of leaves per shoot and the dry weight, none of the four response variables had a significant effect. The maximum leaf length was only significantly influenced by the months post transplanting ($p < 0.001$; $F = 6.578$), with significant differences each time between spring and fall monitoring campaigns. The leaf surface area was significantly influenced by the bathymetry ($p = 0.014$; $F = 8.4173$) and the months post transplanting ($p <0.001$; $F = 10.4409$), with significant differences each time between spring and fall monitoring campaigns. The PERMANOVAs performed with the factors transplantation effect and months post transplanting highlighted several significant differences between transplants and control meadows over the 36 months of monitoring. The number of leaves, maximum leaf length, leaf surface area and biomass were significantly influenced by the transplantation effect, the months post transplanting, and the interaction between the two factors (Figure 2.31, Table S2.4). The transplants had a higher number of leaves at the time of transplanting compared to control meadows, followed by a similar number of leaves 3 months and 12 months post transplantation. After 15 months, the transplants had less leaves per shoot than the control meadows (Figure 2.31, Table S2.5). The maximum leaf length was always higher for the control meadows compared to the transplants (Figure 2.31, Table S2.5). The leaf surface area was similar for both transplants and control meadows at the time of transplanting, but three months after transplantation the leaf surface area of the transplants was significantly lower than the control meadows (Figure 2.31, Table S2.5). Finally, the control meadows had a significantly higher biomass than transplants for all monitoring campaigns, except 12 months post transplanting when there was no significant difference (Figure 2.31, Table S2.5).

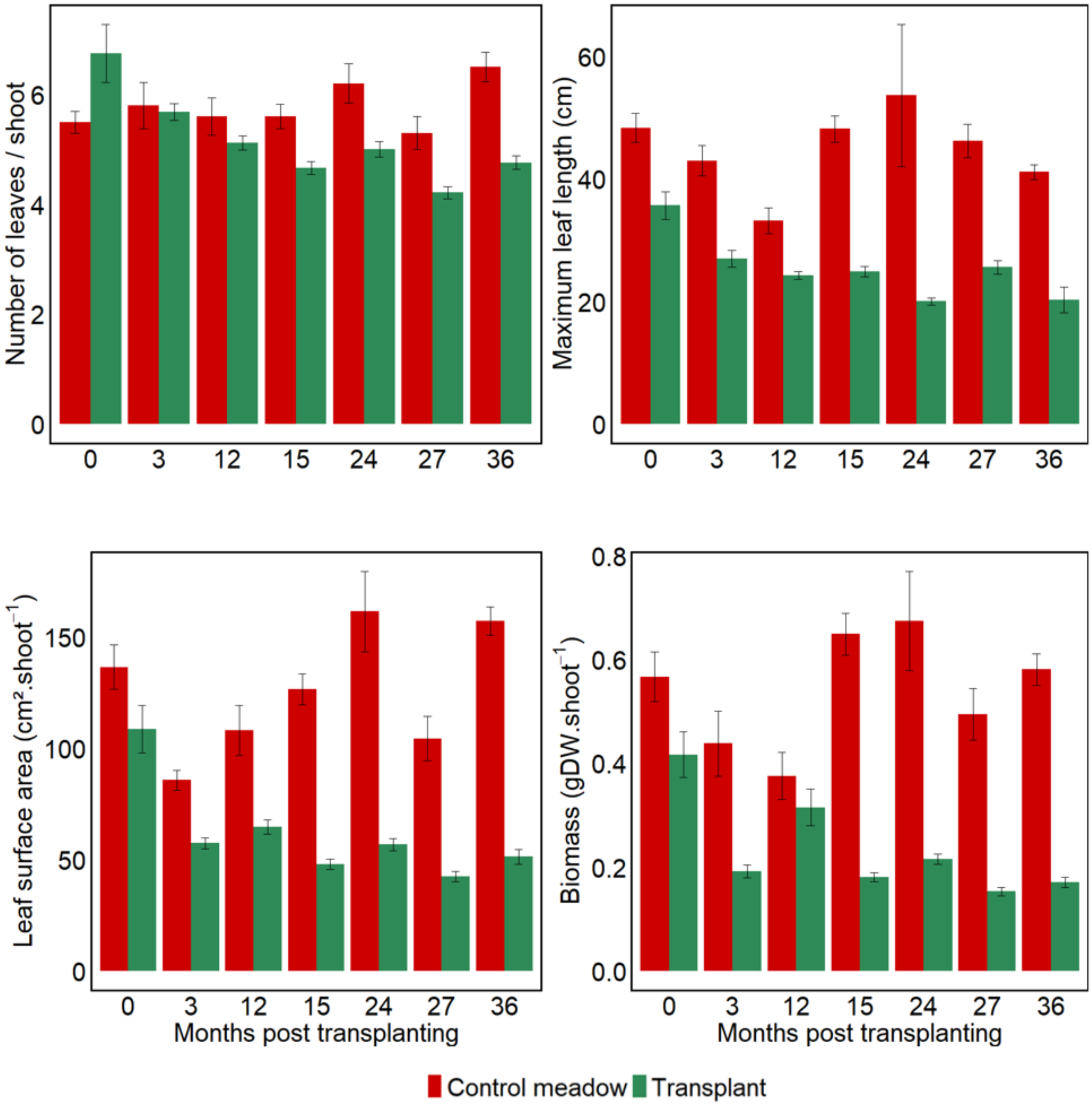


Figure 2.31. *Mean number of leaves, maximum leaf length, leaf surface area and biomass of P. oceanica transplants and control meadows. Vertical error bars represent standard errors.*

3.3 Root morphological traits

The six root morphological traits investigated in this study were all significantly influenced by the transplantation method + effect (Table S2.6). Strong differences were found with control meadows having a much more developed root system than the transplants (Figure 2.32, Table S2.7). Significant differences were also found between the transplants according to the transplantation method, with the transplants attached to the iron staples having a much more developed root system than transplants on the coconut fiber mats or the BESE elements (Figure 2.32, Table S2.7). No significant differences in root morphological traits were found between the transplants on the coconut fiber mats and the BESE elements (Figure 2.32, Table S2.7). The number of primary roots was also significantly influenced by the bathymetry, with higher values at the deep sites (Table S2.6). Moreover, the number of lateral roots was significantly influenced by the interaction between transplantation method + effect and bathymetry (Table S2.6). For the shallow sites, the control meadow plants had a more developed root system compared to the transplants with the iron staples, which had more lateral roots compared to the transplants on the coconut fiber mats and the BESE elements (Figure 2.32, Table S2.7). For the deep sites, there was no significant difference in the number of lateral roots between control meadows and iron staples, but both still had significantly more lateral roots than coconut fiber mats and BESE elements (Figure 2.32, Table S2.7). It is also noteworthy that no lateral roots were observed on transplants attached to BESE elements or coconut fiber mats at shallow sites. A similar pattern was observed at deep sites, except that a very small number of lateral roots were present on transplants anchored with coconut fiber mats (Figure 2.32).

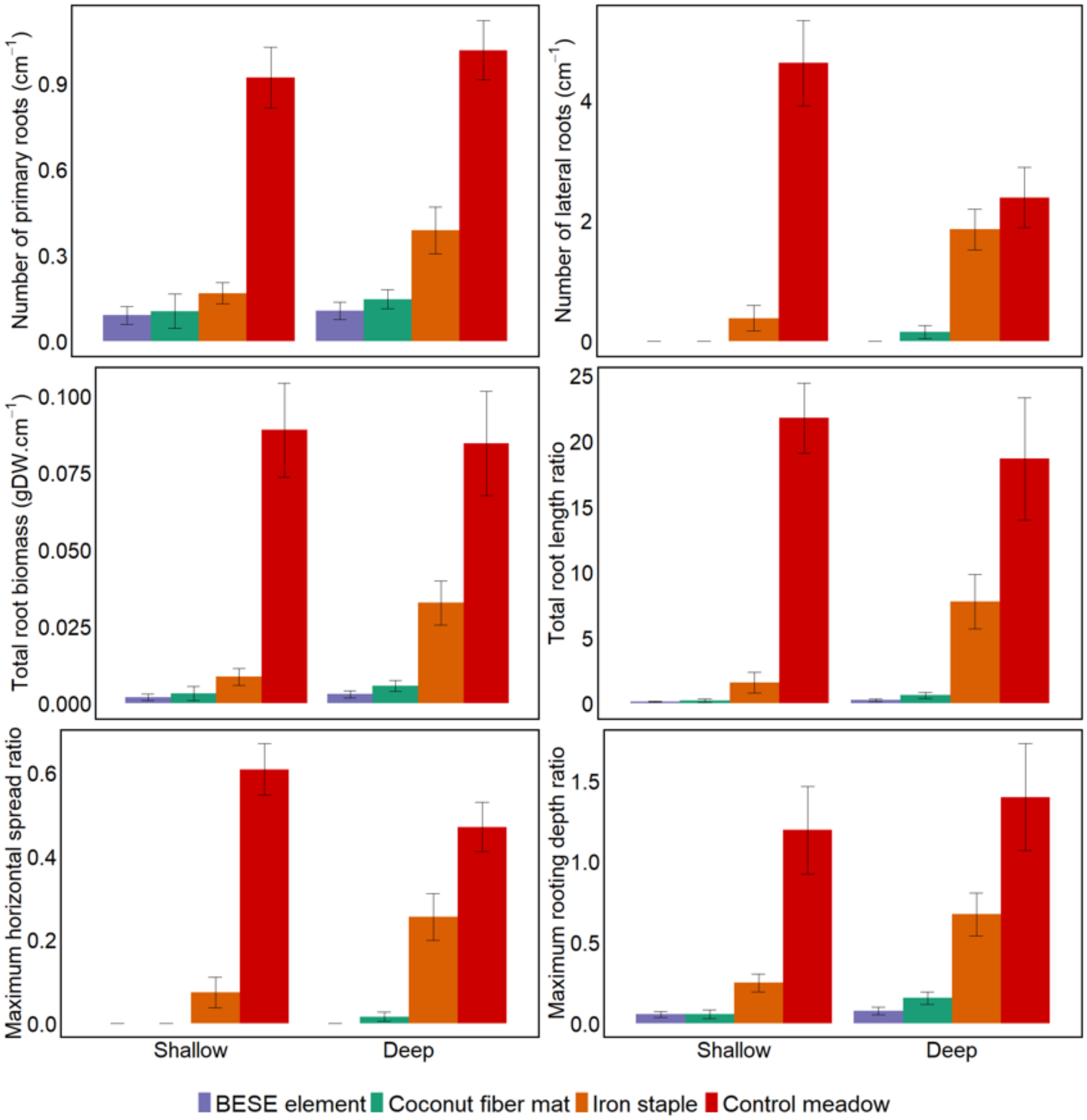


Figure 2.32. *Mean number of primary roots, number of lateral roots, total root biomass, total root length ratio, maximum horizontal spread ratio, maximum rooting depth ratio of P. oceanica transplants (according to the different transplantation methods) and control meadows as a function of bathymetry. Vertical error bars represent standard errors.*

3.4 Cost-efficiency analysis

The unit costs of the three transplantation materials differed considerably, leading to substantial variations in the cost per m^2 transplanted. BESE elements were by far the most expensive, followed by coconut fiber mats, whereas iron staples represented the most economical option. Although survival rates vary among the three transplantation methods, the cost trends remain consistent when comparing both the costs per m^2 transplanted and the cost per m^2 effectively restored after 36 months. Iron staples resulted in the lowest cost per effectively restored m^2 at both shallow and deep sites (Table 2.2).

Table 2.2. *Cost-efficiency comparison of transplantation methods.*

Transplantation method	Unit cost (€)	Unit size (m^2)	Cost/ transplanted surface (€.m^{-2})	Mean survival		Cost/restored surface after 36 months (€.m^{-2})	
				Shallow	Deep	Shallow	Deep
BESE element	21.0	0.42	50.0	0.788	0.818	63.4	61.1
Coconut fiber mat	8.1	1	8.1	0.313	0.538	25.9	15.1
Iron staple	0.3	NA	6.6	0.566	0.826	11.7	8.0

4. Discussion

In recent years, numerous efforts have been made to transplant *P. oceanica*, reflecting a growing interest in the conservation and restoration of marine ecosystems (Boudouresque et al., 2021; Pansini et al., 2022). However, several knowledge gaps remain and need to be addressed to improve the success of *P. oceanica* meadow restoration projects. Among these, the comparison of donor sources for planting material (donor meadow vs storm-fragment) and the development of sustainable methods for transplant fixation on the seafloor are two key research areas (Pergent-Martini et al., 2024) investigated in this study. This experimental work aimed to address these gaps by transplanting a total of 693 *P. oceanica* cuttings onto dead matte at a recipient site in the Bay of Alga (Calvi, Corsica). The three-year monitoring conducted during this initial study identified the most effective transplantation method and donor source, and confirmed the suitability of the site for the implementation of a large-scale restoration project (Boudouresque et al., 2021; Pergent-Martini et al., 2024).

P. oceanica transplantation projects have relied on rhizome fragments either directly harvested from donor meadows (e.g., Bacci et al., 2024; Calvo et al., 2021; Pirrotta et al., 2015) or collected as storm-fragments of unknown origin (i.e., depth, substratum), typically found drifting on the seafloor and accumulating at the edges of meadows or in natural sandy intermattes (e.g., Castejón-Silvo & Terrados, 2021; Mancini et al., 2021; Piazzi et al., 2021a; Ward et al., 2020). To the authors' knowledge, this is the first *P. oceanica* transplantation project to experimentally compare the performance of these two donor sources as planting material under similar environmental conditions. Results from the 36 months of monitoring revealed no significant differences between storm-fragments and intermatte cuttings in terms of survival rate, leaf and root morphological traits. These findings indicate that, when transplanted under similar environmental conditions, the origin of planting material (donor meadow vs storm-fragment) does not significantly influence transplant performance within the studied timeframe. Storm-fragments, once detached from their original meadow, retain the capacity to re-establish and thrive when reintroduced (Almela et al., 2008; Balestri et al., 2011; Di Carlo et al., 2005). As a non-destructive alternative to harvesting fragments from donor meadows, storm-fragments should be prioritized as planting material in restoration initiatives. The use of cuttings collected from donor meadows should only be considered when storm-fragments availability in the study area is insufficient to support the restoration of degraded sites. Furthermore, we recommend prioritizing collection from erosion edges of natural sandy intermattes rather than from other *P. oceanica* seascape types (Abadie et al., 2015, 2018; Gobert et al., 2016). These intermattes undergo natural dynamics of erosion and recolonization, driven by orbital bottom currents eroding the meadow and creating vertical matte edges (Gobert et al., 2016). Harvesting *P. oceanica* cuttings at these erosion edges has a lower ecological impact, as the action of currents in these zones naturally leads to the formation of storm-fragments from exposed rhizomes. As a last resort, and only when the area targeted for restoration is large relative to the total surface area of existing *P. oceanica* meadows in the study area (Boudouresque et al., 2021), harvesting fragments from other parts of the donor meadow may be considered. In such cases, extraction should not exceed the threshold of one rhizome per m² of donor meadow as recommended by Pergent-Martini et al. (2024).

While the donor source had no significant influence on transplant survival 36 months after transplantation, the method used to anchor the cuttings plays a more decisive role, influencing both survival rates and root system development. This study compared the use of biodegradable structures (i.e., BESE elements and coconut fiber mats) as anchoring material with individual iron staples as transplantation methods. The structural complexity provided by BESE elements and coconut fiber mats mimics emergent traits (Van der Heide et al., 2021; Piazzi et al., 2021; Temmink et al., 2020), such as dense aggregations of roots and rhizomes. These traits are known to promote self-facilitation processes naturally generated by established conspecifics (Kendrick et al., 2005), and were hypothesized to reduce physical stress and enhance long-term establishment of the transplants (Temmink et al., 2020). To define if the transplantation methods used were successful or not, transplantation success was defined as a survival rate ≥50% (Bayraktarov et al., 2016; Danovaro et al., 2025) three years after transplantation (Molenaar & Meinesz, 1995). Survival outcomes differed between transplantation depths. At shallow sites, coconut fiber mats did not meet the success threshold, whereas both BESE elements and iron staples were successful, although iron staples had a survival rate (56.57 ± 0.05 %) only slightly above the defined threshold. At deeper sites, both BESE elements and iron staples were highly successful, with survival rates exceeding 80%. Coconut fiber mat also achieved a survival rate above the threshold (53.79 ± 0.04 %), but with a lower survival rate than the other two methods. Similar survival rates for cuttings transplanted on coconut fiber mats were obtained by Piazzi et al. (2021) who tested the same methodology. However, contrary to the recommendations of Piazzi et al. (2021), our results show that coconut fiber mats were the least effective method at both tested transplanting depths. In addition to lower survival rates, coconut fiber mats also resulted in significantly lower shoot abundance compared to both BESE elements and iron staples. Finally, the cost-efficiency analysis does not support the use of coconut fiber mats, which should therefore be excluded from consideration in large-scale restoration projects under similar environmental conditions.

Although BESE elements and iron staples exhibited similar survival rates at deep sites 36 months after transplantation, iron staples showed lower survival rates at shallow sites, whereas BESE elements maintained similar survival rates across both depths. Previous evidence indicate that the survival rate of *P. oceanica cuttings* transplanted at different depths is quite variable. For instance, higher survival rates were reported at shallower depths (8 - 12 m) compared to deeper sites (17 - 21 m) in Mancini et al. (2021), suggesting that increased light irradiance may promote higher survival at shallower depths (Ruiz & Romero, 2001). However, a large-scale transplantation project using iron staples on dead matte reported 79% survival two years after transplantation, with no significant differences between shallow (8-10 m) and deep sites (18 – 23 m) (Mancini et al., 2022). Similarly, Castejón-Silvo and Terrados (2021) observed no differences in survival rates between cuttings transplanted at 15, 20 and 25 m depth. Our results suggest that differences in light intensity between shallow and deep sites did not significantly influence cutting survival 36 months after transplantation, highlighting *P. oceanica* ability to acclimate to a range of light conditions as observed in previous studies (Boulenger et al., 2024; Dattolo et al., 2017; Ruiz & Romero, 2003; Stipcich et al., 2023). It is also noteworthy that most of our cuttings were transplanted at deeper depths than their origin. Specifically, intermatte cuttings were collected at 15 m, while storm-derived fragments originated from a depth range of 6 m to 28 m. Previous studies have shown that transplanting *P. oceanica* cuttings to deeper waters than their origin may result in reduced survival (Genot et al., 1994; Molenaar, 1992; Molenaar & Meinesz, 1992). However, more recent research reports contrasting outcomes, with high survival rates even at increased depths (Boulenger et al., 2024; Mancini et al., 2022; Piazzi et al., 1998), and with evidence of photosynthetic acclimation to ambient light conditions (Boulenger et al., 2024). Interestingly, studies reporting lower survival rates relied on orthotropic rhizomes (Genot et al., 1994; Molenaar, 1992; Molenaar & Meinesz, 1992), whereas those showing little or no depth-related impact used plagiotropic shoots (Boulenger et al., 2024; Mancini et al., 2022; Piazzi et al., 1998). These findings suggest that while transplanting cuttings at similar depths may facilitate acclimation due to pre-existing physiological adaptations, it is not a strict requirement, particularly when using plagiotropic rhizomes and when donor and recipient sites are located within the same bay and experience comparable environmental conditions. It is therefore likely that the lower survival of cuttings anchored with iron staples at shallow sites is due to increased hydrodynamic stress. Shallower areas typically experience higher hydrodynamic energy, which decreases with depth (Bonamano et al., 2010; Uhrin & Turner, 2018; Vacchi et al., 2017). This was further supported by monitoring *P. oceanica* natural recolonization, which showed more pronounced erosion at shallow sites compared to deeper ones (Boulenger et al., 2025a). Iron staples possess lower structural rigidity compared to BESE elements, potentially explaining their reduced performance under high-energy conditions, while their similar performance at deeper sites could be attributed to the reduced hydrodynamic forces (Table 2.3). Moreover, the cuttings were initially secured to the horizontal section of the iron staples using plastic cable ties wrapped around the rhizome. However, this method proved detrimental, as wave action and currents could cause the ties to cut into the rhizome, leading to transplant damage and loss. After observing these negative effects, we replaced this approach by simply positioning the iron staple directly over the rhizome without using cable ties. This adjustment minimized shear stress and resulted in improved anchorage stability over time. These results emphasize the importance of testing different transplantation methods under varying environmental conditions, and the need of site-specific restoration designs as no universally optimal solution exists.

Another key difference between transplantation methods lies in the development of root morphological traits. Establishing a functional root system is crucial for transplants to provide nutrient supply to meet physiological needs (Gobert et al., 2005; Lepoint et al., 2002, 2004), withstand hydrodynamic stress (Infantes et al., 2011), and ensure long-term survival (Balestri & Lardicci, 2006; Lepoint et al., 2004; Vangeluwe et al., 2004). For all six root traits measured three years after transplantation, significantly greater development was observed in cuttings anchored with iron staples (Table 2.3). Initially, the cuttings lacked a developed root system, and three years' post-transplantation, little to no root development was observed in cuttings fixed with BESE elements or coconut fiber mats. Previous studies have shown that root formation in *P. oceanica* rhizome fragments primarily occurs during spring and summer and typically takes from 3 to 12 months (Balestri et al., 2011; Meinesz et al., 1992). However, Castejón-Silvo and Terrados (2021) reported that the development of a fully functional root system in transplanted cuttings may take up to two years. Our findings indicate that under certain conditions, it can take more than three years to grow an effective root system, as observed in transplants anchored with BESE elements and coconut fiber mats (Table 2.3). This delay could be attributed to a limited availability of internal reserves in the transplants on the coconut fiber mats and BESE elements, which may impede root formation (Lepoint et al., 2004; Vangeluwe et al., 2004). Nonetheless, this limitation is thought to primarily affect orthotropic rhizomes and may not apply to plagiotropic rhizomes as used in our study (Castejón-Silvo & Terrados, 2021).

Moreover, there was no differences in the number of leaves, maximum leaf length, leaf surface area and leaf biomass between transplantation methods. Sandy substrates appear to promote the development of *P. oceanica* root systems (Balestri et al., 2015). Cuttings anchored with iron staples had direct contact between their rhizomes and the sediment, including the underlying dead matte (Figure 2.28). In contrast, cuttings placed on coconut fiber mats are physically separated from the seafloor by the 5 mm thickness of the mat (Figure 2.28). Similarly, BESE elements create a 6 cm elevation, resulting in a substantial gap between the transplants' roots and the dead matte (Figure 2.28). These differences in spatial positioning likely modulate the degree of interaction between the roots and the surrounding sediment microbial pool, which is known to influence the recruitment and establishment of root-associated bacterial communities. Plants selectively recruit their microbiome from the surrounding soil or sediment, and the composition of this initial microbial pool plays a critical role in shaping root microbial assembly and plant performance (Bonito et al., 2014; Cúcio et al., 2016; Haney et al., 2015; Hartman et al., 2018). Cuttings in closer proximity to the sediment may benefit from greater exposure to beneficial sediment-associated bacteria, whereas elevated cuttings are subjected to altered oxygen and nutrient gradients that could favour distinct microbial assemblages (Boulenger et al., 2025b). Moreover, plant exudates released by the roots in the surrounding sediment promote microbial colonization through chemotaxis, attracting beneficial microbial partners that enhance plant fitness within the seagrass rhizosphere (Crump et al., 2018; Sogin et al., 2022; Zhang et al., 2024). However, such interactions may be diminished in cuttings placed on coconut fiber mats and BESE-elements due to their reduced initial contact with the sediment, potentially limiting early microbiome recruitment and establishment (Boulenger et al., 2025b). Given the potentially beneficial role of microbial interactions in seagrass ecology, further research is needed to better understand their influence on transplantation success and to assess how different transplantation methods may shape associated bacterial communities during the early stages of transplants' establishment (Corinaldesi et al., 2023; Valdez et al., 2020). Metagenomic approaches offer valuable tools to investigate these dynamics at both phylogenetic and functional levels. Expanding *in situ* studies across habitats and environmental conditions will be crucial to fully integrate microbiome knowledge into effective restoration practices (Corinaldesi et al., 2023; Mohapatra et al., 2024).

Based on these results, the use of iron staples is recommended as a transplantation method for *P. oceanica* restoration projects conducted on dead matte. Although BESE elements achieved survival rates high enough to be considered successful, the near absence of root development in transplants raises concerns about their long-term persistence. Moreover, the cost-efficiency analysis revealed that the use of BESE elements was the least cost-effective among the three tested methods, primarily due to their high material costs (Table 2.3). In comparison, iron staples were found to be nearly ten times more cost-effective. Nevertheless, BESE elements may represent a suitable solution under specific environmental conditions. Castejón-Silvo and Terrados (2021) demonstrated that iron staples do not support the successful recovery of *P. oceanica* meadows when transplantation is carried out on sandy or gravel substrates lacking underlying dead matte (Table 2.3), such as areas disturbed by underwater infrastructure works (e.g., power line installation). In cases where such interventions are unavoidable and affect matte areas, the addition of calcareous stones has been proposed to promote natural recolonisation (Badalamenti et al., 2011; Di Carlo et al., 2005) and to serve as a substrate for transplantation (Alagna et al., 2019). An alternative method, tested by Bacci et al. (2024), involved the use of cement blocks combined with metallic grid frames to transplant *P. oceanica* cuttings onto sandy sediments following pipeline installation. However, both methods present notable disadvantages, including high economic costs, greater handling difficulty due to the weight of the structures, and the introduction of substantial amounts of exogenous material (i.e., stones, cement) into the marine environment. In contrast, BESE elements may offer a suitable transplantation method for restoring *P. oceanica* meadows in areas where the matte has been degraded (Table 2.3). This approach has the advantage of using a fully biodegradable artificial substrate and allows for easier handling due to the lightweight nature of the material. Further research is needed to test the use of BESE elements in degraded matte areas and to determine whether the survival rates observed in our study are consistent under those environmental conditions.

Table 2.3. *Summary of the main limitations and site-specific advantages of iron staples and BESE elements as transplantation methods for P. oceanica restoration.*

Transplantation method	Limitations	Site-specific advantages
Iron staple	– Lower performance under high hydrodynamic conditions – Requires manual insertion in dead matte; limited applicability on unconsolidated substrates	– High survival rates on intact matte substrates under low hydrodynamics. – Promotes robust root development, enhancing anchorage and belowground biomass accumulation – High cost-efficiency (10× cheaper than BESE) – Minimal introduction of exogenous material into the environment – Can be removed after a couple of years when root system is sufficiently developed
BESE element	– Delayed root development (limited root traits after 36 months) – High material costs – Physically elevate cuttings (~6 cm above dead matte), possibly reducing plant–microbe interactions and root system development	– Consistent survival across depth gradients (20–28 m), including high-hydrodynamic zones → Biodegradable structure mimics natural root-rhizome matrix and may facilitate conspecific aggregation/self-facilitation in high-stress areas (e.g., Temmink et al., 2020) – Suitable for degraded matte or unstable sandy/gravel substrates where staples are not applicable

The results of this study indicate that both iron staples and BESE elements achieved transplantation success after three year, as defined by survival thresholds. However, the same conclusion cannot be drawn regarding restoration success, which refers to the re-establishment of the structural and functional characteristics of the transplanted meadow in alignment with those of a natural reference site. Three main categories can be used to comprehensively assess restoration success : (1) seagrass structural attributes, such as canopy height, shoot density, and biomass; (2) ecological functions, including fish and invertebrate diversity and abundance, epiphytic colonization, and nursery habitat provisioning; and (3) biogeochemical functions, encompassing parameters such as water temperature, porewater nutrient concentrations, sediment granulometry, organic matter content, and sedimentation rate (Beheshti et al., 2021; Boulenger et al., 2025a; Castro-Fernandez et al., 2025; Orth et al., 2020; Pergent-Martini et al., 2024).

Among these, seagrass structural complexity is a particularly important driver, as it directly supports the recovery of both ecological and biogeochemical functions (Beheshti et al., 2021; Castro-Fernandez et al., 2025). Thirty-six months after transplantation, significant differences in both leaf and root morphological traits were observed between transplants and control meadows. Leaf morphological traits remained relatively stable throughout the monitoring period, showing neither clear progression or regression when compared to the control meadow. The reduced leaf traits may reflect suboptimal environmental conditions, a possible relocation of internal resources from leaf production to rhizome horizontal growth (Gobert et al., 2005; Lepoint et al., 2004), or increased leaf breakage due to greater exposure to water movement within a sparsely vegetated canopy, as typically occurs in transplanted areas lacking the structural buffering of dense natural meadows (Collier et al., 2009). These findings are consistent with those of Pansini et al. (2024), who reported that transplants across five different sites exhibited a consistently lower number of leaves and reduced leaf growth rates compared to reference meadows, persisting up to 36 months post-transplantation. Notably, even six years after transplantation, the maximum leaf length was still lower than control meadows. Similarly, a long-term study conducted 10 years after transplantation on disturbed sandy sediment showed that transplanted shoots had shorter and narrower leaves compared to natural meadows (Bacci et al., 2024). However, these results contrast with the findings of Calvo et al. (2021) and Mancini et al. (2021), who reported higher primary production (i.e., leaf growth rate, leaf length, shoot density) in transplanted meadows compared to control meadows within the first 48 months following transplantation. As suggested by Pansini et al. (2024), this may be due to the use of *P. oceanica* cuttings with an intact root system, which was lacking in our cuttings at the time of transplantation. This likely constrained early shoot development in our study, consistent with previous findings (Lepoint et al., 2004; Vangeluwe et al., 2004). This hypothesis is further supported by the root traits data, which revealed that transplants exhibited significantly smaller root systems compared to control meadows. The only exception was the number of lateral roots, which was comparable between transplants anchored with iron staples and control plants at deep sites. Longer-term monitoring is required to determine whether the leaf and root morphological traits of transplants will eventually converge with those of control meadows over time.

The various transplantation methods tested to date highlight persistent challenges in achieving full restoration success. This includes both the failure to reach the structural characteristics of natural meadows over the long term and the incomplete recovery of associated ecological functions and ecosystem services. In particular, the reduced leaf traits observed in transplants result in lower habitat complexity, thereby diminishing their nursery and habitat functions for numerous fish species (Castro-Fernández et al., 2025). To date, the simultaneous recovery of all three components of *P. oceanica* restoration success, namely structural attributes, ecological functions, and biogeochemical functions, has never been demonstrated in any transplantation study (Pergent-Martini et al., 2024). This highlights the urgent need for further research that integrates all of these dimensions into long-term monitoring programs (>10 years), in order to determine the time frame required for the full recovery of *P. oceanica* ecosystem functions following transplantation. Furthermore, it may be valuable to develop new assessment tools or adapt existing ones, such as the Ecosystem-Based Quality Index (EBQI; Personnic et al., 2014), to enable standardized and replicable evaluation of both structural and functional recovery in transplanted *P. oceanica* meadows.

Moreover, to promote the recovery of ecosystem functions, it is recommended to transplant with high shoot densities to enhance structural complexity. Furthermore, implementing large-scale planting is also advised, as it has been shown to improve restoration outcomes (van Katwijk et al., 2016) by enabling new transplants to overcome negative feedbacks in the system (e.g. hydrodynamic stress, sediment resuspension) (Maxwell et al., 2017). However, large-scale transplantation should only be considered after a thorough assessment of the local environmental conditions (e.g., hydrodynamics, substrate type, light availability, sedimentation, etc) and the natural recolonization dynamics at the restoration site (Boudouresque et al., 2021; Boulenger et al., 2025a). It is essential to first conduct a pilot trial using a limited number of cuttings to identify the most suitable restoration method(s) under the site-specific conditions. Only after validating the effectiveness of the selected approach(es) should large-scale transplantation be implemented. Finally, it is important to emphasize that the slow growth rate of P*. oceanica* makes direct comparisons with the restoration of other seagrass species challenging. A better management of anthropogenic pressures and the prioritization of the conservation of existing *P. oceanica* meadows must remain the central objectives. In addition, large-scale (>1 ha) restoration of *P. oceanica* is particularly difficult due to the high costs and workload involved, as well as the limited availability of donor material. For effective restoration, efforts should instead focus on reducing the fragmentation of degraded meadows, thereby boosting natural recolonization processes.

5. Conclusion

This study provides new insights into *P. oceanica* restoration by comparing the performance of different transplantation methods and donor sources at shallow and deep sites. The results demonstrate that storm-fragments are as effective as donor meadow cuttings in terms of transplant survival and morphological development, supporting their use as a sustainable, non-destructive alternative for *P. oceanica* restoration projects. Among the tested transplantation methods, iron staples emerged as the most cost-effective and biologically effective solution. BESE-elements, while yielding comparable survival rates, presented limitations in root development and economic feasibility. Coconut fiber mats, despite their biodegradability, performed poorly across most performance indicators and are not recommended for larger scale operations under similar environmental conditions. This study emphasizes the importance of conducting pilot experiments before any large-scale planting, in order to select the most appropriate method based on the environmental conditions of the degraded site. Moreover, the study reveals that transplantation success does not necessarily equate to ecological restoration success. Significant differences in both root and leaf morphological traits between transplants and reference meadows persisted after three years, potentially affecting habitat complexity and ecosystem function recovery. These findings highlight the importance of monitoring beyond survival metrics, focusing on long-term structural and functional convergence with natural meadows. Finally, given the species' inherently slow growth and limited donor material availability, restoration should remain a complementary tool to conservation, and not a substitute. The protection and long-term management of existing *P. oceanica* meadows must remain the priority.

Chapitre III


©Fabrice Dudenhofer

Réponses microbiologiques, physiologiques et biochimiques de *Posidonia oceanica* à la transplantation : implications pour la réussite des projets de restauration

Ce chapitre est composé de trois articles :

Article 3 : Boulenger, A., Aires, T., Engelen, A.H., Muyzer, G., Marengo, M., & Gobert, S. (2025). Microbiome matters: how transplantation methods and donor origins shape the successful restoration of the seagrass *Posidonia oceanica*. *Environmental Microbiome*, 20(99). https://doi.org/10.1186/s40793-025-00764-9

Article 4 : Boulenger, A., Roberty, S., Lopez Velosa, M. M., Marengo, M., & Gobert, S. (2024). The use of photo-biological parameters to assess the establishment success of *Posidonia oceanica* cuttings after transplantation. *Water*, 16(12), 1702. https://doi.org/10.3390/w16121702
Seule la méthodologie a été reprise dans cet article et utilisée pour venir compléter l'article 5.

Article 5 : Boulenger, A., Acuna, A.L.A., Roberty, S., Lepoint, G., Marengo, M., & Gobert, S. (2026). Physiological and biochemical performance indicators of seagrass restoration success. *Marine Pollution Bulletin,* 223, 119036. https://doi.org/10.1016/j.marpolbul.2025.119036

Microbiome matters: How transplantation methods and donor origins shape the successful restoration of the seagrass Posidonia oceanica

Arnaud Boulenger, Tânia Aires[c], Aschwin H. Engelen[c], Gerard Muyzer[d], Michel Marengo[b], Sylvie Gobert[a,b]

[a] Laboratory of Oceanology, MARE Centre, UR FOCUS, University of Liege, Liege, Belgium

[b] STAtion de REcherche Sous-marines et Océanographiques (STARESO), Calvi, France

[c] Centro de Ciências do Mar (CCMAR), Centro de Investigação Marinha e Ambiental (CIMAR), Universidade do Algarve, Faro, Portugal

[d] Microbial Systems Ecology, Department of Freshwater and Marine Ecology, Institute for Biodiversity and Ecosystem Dynamics, University of Amsterdam, Amsterdam, Netherlands

Environmental Microbiome (2025)

Abstract

Posidonia oceanica forms extensive seagrass meadows in the Mediterranean Sea, providing key ecosystem services. However, these meadows decline due to anthropogenic pressures like anchoring and coastal development. Transplantation-based restoration has been explored for decades, yet the role of the plant-associated microbiome in restoration success remains largely unknown. 16S rRNA gene amplicon sequencing was used to investigate how different transplantation methods and donor origins influence the bacterial communities of *P. oceanica* cuttings two years post-transplantation. We tested three transplantation methods, iron staples, coconut fiber mats, and BESE elements, and compared them with control meadows and donor populations from two different origins: naturally uprooted storm-fragments and intermatte cuttings manually harvested from established meadows. Our results show that transplantation methods strongly shape bacterial communities in seagrass roots. Iron staples promoted microbial assemblages most similar to natural meadows, likely due to direct sediment contact enhancing recruitment of key functional bacterial orders such as *Chromatiales* and *Desulfobacterales*. In contrast, BESE elements and coconut fiber mats displayed dissimilar bacterial communities compared to control meadows, likely due to material composition and physical separation between the cuttings and the sediment. Donor origin had only subtle effects on bacterial communities' structure, although intermatte cuttings showed higher abundances of *Candidatus* Thiodiazotropha, a genus thought to be involved in sulfur oxidation and nitrogen fixation. Our results demonstrate that transplantation methods strongly influence root-associated bacterial communities. Limited sediment contact in elevated substrates delayed the establishment of key functional bacteria, highlighting the importance of direct interaction with the sediment microbial pool. These results imply that restoration strategies should prioritize methods enhancing sediment–root interactions to support microbial recovery. Incorporating microbiome considerations, such as optimized substrates or microbial inoculation, could improve the resilience and long-term success of *P. oceanica* restoration.

Keywords

Holobiont, Microbiome, Rhizosphere, Restoration, Seagrass, Symbiosis, Transplantation

ACKNOWLEDGMENTS

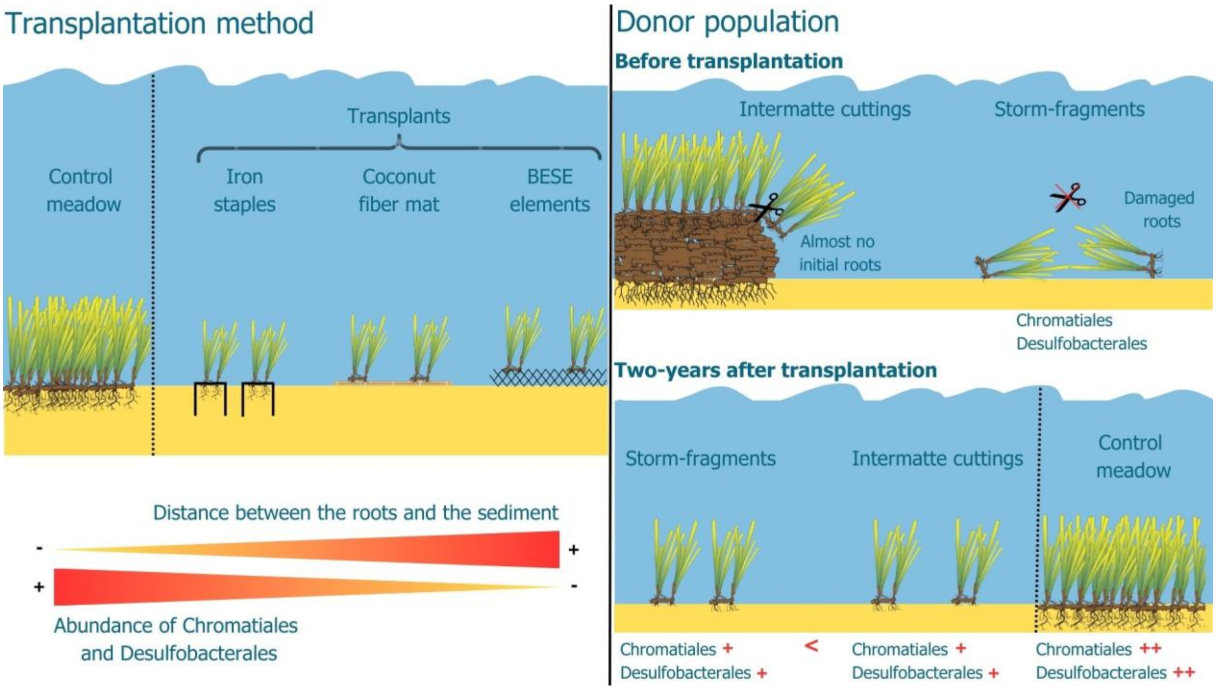
This work was supported by the University of Liege (grant FSR2021) and the Fonds National de la Recherche Scientifique — FNRS (grants ASP 40006932 and CDR J.0076.23). This study is part of the STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) project funded by the Territorial Collectivity of Corsica and by the Rhone-Mediterranean and Corsican Water Agency. GM was supported by the BioDiversa project RESTORESEAS, which was funded by the Dutch Ministry of Agriculture, Nature, and Food Quality. This study also received Portuguese national funds from FCT through projects UIDB/04326/2020 and LA/P/0101/2020 and CCMAR/ID/16/2018 to AE. The authors are grateful to STARESO for facilities and field assistance.

AUTHORS’ CONTRIBUTIONS

AB: Conceptualization, Field Sampling, Methodology, Formal Analysis, Investigation, Writing, Visualization; TA: Conceptualization, Methodology, Formal Analysis, Investigation, Writing; AHE: Conceptualization, Methodology, Formal Analysis, Investigation, Writing, Visualization, Supervision; GM: Conceptualization, Methodology, Formal Analysis, Investigation, Writing, Visualization, Supervision; MM: Conceptualization, Writing, Supervision, Funding; SG: Conceptualization, Writing, Supervision, Funding.

AVAILABILITY OF DATA AND MATERIALS

The sequence reads from all samples collected during this study were deposited in the NCBI data bank (BioProject accession number PRJNA1221124).



Graphical abstract used for publication.

1. Introduction

Seagrasses are marine flowering plants that colonised the aquatic environment about 100 million years ago and are widely distributed in coastal waters worldwide, except in Antarctica (den Hartog & Kuo, 2006; Hemminga & Duarte, 2000). They are key benthic ecosystem engineers (sensu Wright & Jones, 2006) that form three-dimensional meadows providing essential habitats and nursery grounds for marine life (Beck et al., 2001; Jeyabaskaran et al., 2018; Jiang et al., 2020). Those meadows stabilize soft sediments and diminish wave intensity and turbulence, offering coastal protection against erosion (Ackerman & Okubo, 1993; Gambi et al., 1990). Furthermore, they sequester large amounts of CO_2, thus mitigating anthropogenic emissions (Duarte et al., 2010; Hemminga & Duarte, 2000; Turschwell et al., 2021). Despite the ecological and economic significance of seagrass meadows, climate change, and human activities, such as agricultural activities, coastal urbanization, dredging, trawling, and anchoring, have severely impacted those ecosystems (Turschwell et al., 2021; Waycott et al., 2009). These ongoing reductions in seagrass coverage are especially detrimental to large slow-growing seagrass species such as *Posidonia oceanica* (L.) Delile, which forms extensive meadows in the Mediterranean Sea. The alarming global decline of seagrass meadows has prompted a surge in restoration efforts (Rezek et al., 2019; van Katwijk et al., 2016). For *P. oceanica* meadows active restoration, it can be achieved through sod transplantation, which has shown promising results (Descamp et al., 2025). One of the major advantages of this technique lies in preserving the sediment and the underlying matte of the meadow, along with its associated microbiome (Descamp et al., 2025). The second active restoration technique involves transplanting seeds or cuttings into degraded areas. The challenge of transplanting cuttings lies in their long-term anchoring and adaptation to new environmental conditions, such as a modified substrate (Abadie et al., 2016; Abadie et al., 2019; Boulenger et al., 2025). Despite several decades of seagrass restoration research, the role of microbial communities in these processes remains largely overlooked (Corinaldesi et al., 2023). Microbial communities that reside within (endophytic) and on the surface of (epiphytic) plants' tissues can act as functional drivers for their host by forming complex co-associations, impacting terrestrial plant health and productivity (Averill et al., 2022; Bacon et al., 2016; Batista et al., 2021). These microorganisms enhance nutrient availability through nitrogen fixation and the mineralization of organic compounds, produce phytohormones that stimulate root and shoot development, and help alleviate plant stress (Mantelin et al., 2004; Vessey, 2003; Zhou et al., 2024). Yet, our understanding of plant-microbial interactions in marine environments is still limited (Valdez et al., 2020). However, recent studies in salt marshes highlight the potential significance of these interactions. Daleo et al. (2007) found that mycorrhizal fungi enhance nutrient uptake in dense-flowered cordgrass (*Spartina densiflora*). Likewise, seagrasses form symbiotic relationships with various microorganisms both above and below ground (Cúcio et al., 2018; Fuggle et al., 2023; Garcias-Bonet et al., 2016; Mohr et al., 2021; Tarquinio et al., 2019; Valdez et al., 2020; Vohník et al., 2019;). For example, seagrasses are associated with sulfide-oxidizing bacteria to reduce toxic sulfide accumulation (Cúcio et al., 2016; Martin et al., 2020). Additionally, some bacteria on seagrass leaves and roots produce antimicrobial molecules that may protect the plants by selectively targeting pathogens and biofouling organisms (Graham et al., 2024; Tasdemir et al. 2024).

However, marine restoration is a more recent scientific discipline than terrestrial restoration (Saunders et al., 2020). In terrestrial ecosystems, there is evidence that the core microbiota plays a crucial role in maintaining the functional stability of soil microbiomes, nutrient cycling, and plant establishment in reforested areas. This microbiota should also be considered in marine restoration plans' policy and management strategies (Jongen et al., 2024). Considering the potentially beneficial microbial interactions in seagrasses, further research is required to understand better their implications for restoration efforts' success or failure (Corinaldesi et al., 2023; Valdez et al., 2020). A recent guide on *P. oceanica* restoration has emphasized the need for further research on plant-sediment interactions, particularly regarding associated bacterial communities (Pergent-Martini et al., 2024). Notably, recent reviews fail to mention the role of microbial communities in *P. oceanica* restoration (Boudouresque et al., 2021; Pansini et al., 2022), highlighting a critical knowledge gap. While microbial studies have been conducted on some temperate (Christiaen et al., 2013) and tropical (Li et al., 2024) seagrass species, *P. oceanica* remains largely unstudied in this context. In our study, *P. oceanica* cuttings collected from donor populations from two different origins were transplanted onto various biodegradable materials. Two years after transplantation, leaf and root samples were collected from the transplants and nearby control meadows for bacterial community characterization. We hypothesized that transplantation methods would shape distinct bacterial communities due to the material composition and physical structure of the transplantation supports, as well as their proximity to the sediment, with methods allowing closer sediment contact favouring communities more similar to natural meadows. We further hypothesized that donor origin would influence initial bacterial community composition, but that these differences would diminish over time as communities adapt to the transplantation site. Finally, we hypothesized that bacterial communities in transplants would gradually converge towards those of natural meadows over time, reflecting a progressive recovery of the microbiome after transplantation.

2. Material and Methods

2.1 Study area

Samples were collected by SCUBA-diving in May 2024 in a sub-bay of Calvi Bay, Alga Bay (8°43′52′′ E; 42°34′20′′ N), located in front of the oceanographic research station STARESO (Calvi, Western Corsica, France) (Figure 3.33). This bay harbours extensive seagrass meadows of *P. oceanica*, spanning around 0.78 km² at depths ranging from 3 m to 37 m (Abadie et al., 2019). Significant anchoring activity in the area has caused a substantial reduction in these seagrass meadows (Fullgrabe et al., 2022), and restoration efforts by cuttings' transplantation on dead matte took place in the spring of 2022 (Boulenger et al., 2024). As the availability of donor material for transplanting is one of the main constraints in *P. oceanica* meadows restoration, donor populations of two different origins were used as planting material: naturally uprooted seagrass fragments drifting on the seafloor (referred to as storm-fragments) and fragments of *P. oceanica* rhizomes manually extracted from donor meadows. The storm-fragments were collected from various locations near STARESO during scuba dives ranging from 6 to 28 m depth. The manual extraction of *P. oceanica* fragments from donor meadows was performed on a healthy *P. oceanica* meadow located on the erosion side of a natural sandy intermatte at 15m depth (Figure S3.6) (Gobert et al., 2016). The later cuttings are hereafter referred to as 'intermatte cuttings'. The cuttings were attached to the seafloor using three different types of biodegradable artificial structures: (i) iron staples, (ii) biodegradable mat in natural coconut fibre woven mesh (referred to as coconut fiber mat), and (iii) BESE-elements® (BESE Ecosystem Restoration Products, Culemborg, The Netherlands) (Figure 3.34). The storm-fragments and intermatte cuttings were spatially interspersed within each structure, and the structures were spaced approximately 3 m apart. This experimental design was replicated in seven sites, and two control meadows were selected in close vicinity to the experimental sites.

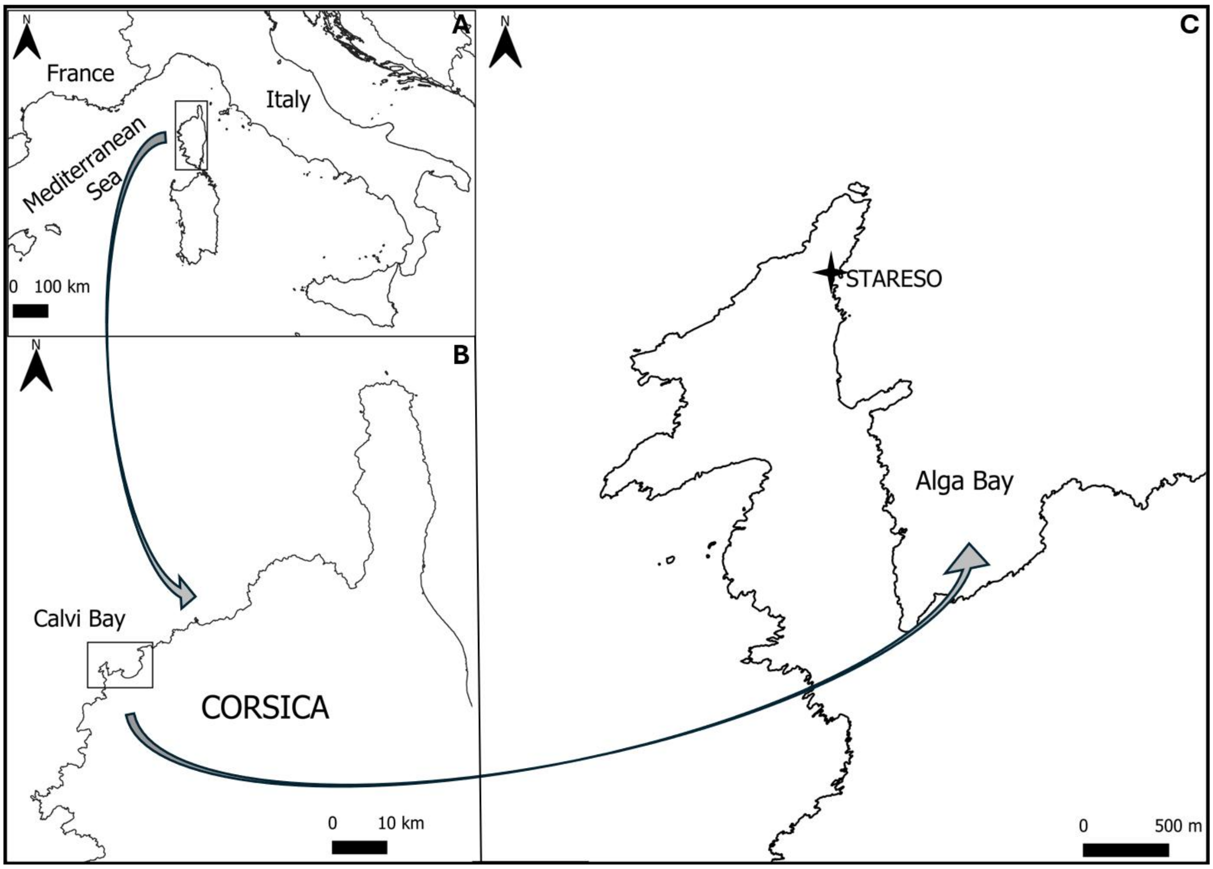


Figure 3.33. *The location of the study area : (**A**) Corsica Island in the North-Western part of the Mediterranean Sea; (**B**) Northern part of Corsica and Calvi Bay; (**C**) Location of the STARESO marine station and Alga Bay (Calvi, Corsica) where the samples were collected.*

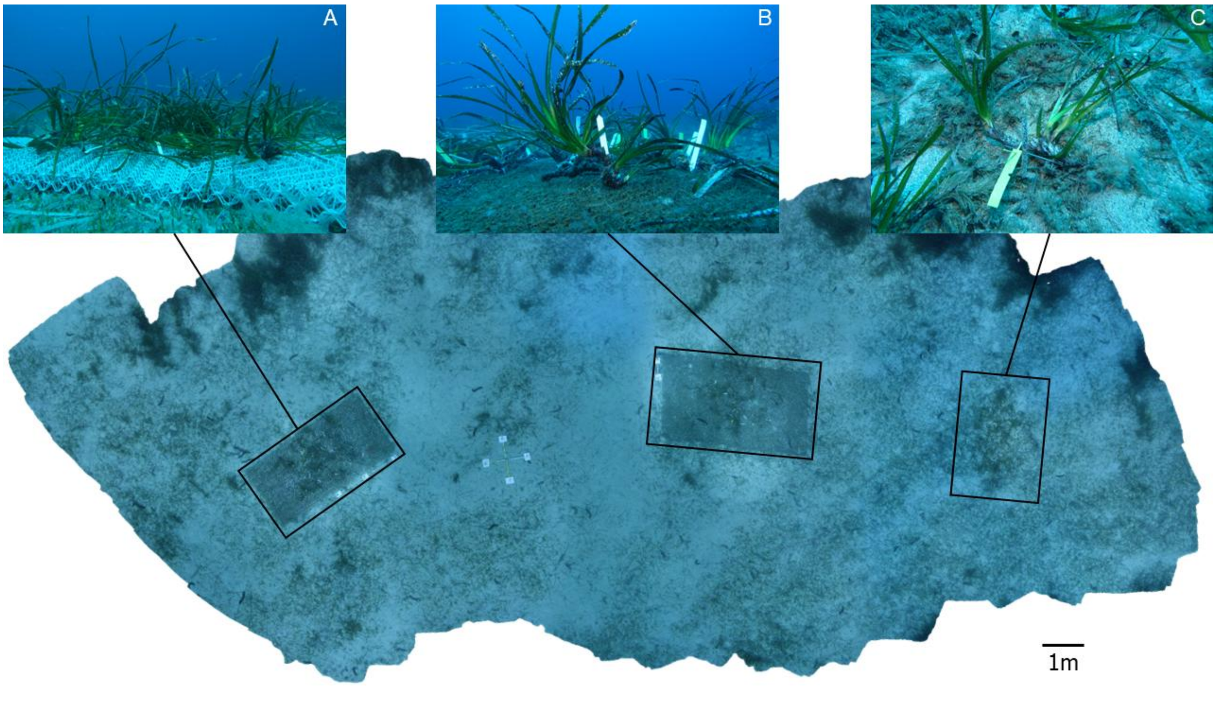


Figure 3.34. *Orthomosaic of one of the seven experimental sites. It represents a dead matte area with the three different P. oceanica transplantation methods tested in this study (black rectangles): (A) BESE elements, (B) coconut fiber mat, and (C) iron staples.*

2.2 Sampling strategy

Transplanted *P. oceanica* fragments were collected two years after transplantation along with environmental samples (i.e., sediment and seawater). Seven replicates of *P. oceanica* transplants were collected for each combination of transplantation method and donor origin. Five individual seagrass fragments were collected at the two control sites, also with environmental samples. This resulted in a total of 52 fragments and a total of 104 plant samples as leaves and roots were separated. Each seagrass fragment (cutting or control plant) was uprooted and washed with seawater from the sampling location to remove sediment, epiphytes and any loosely attached material. The seawater in excess was shaken off. A portion of approximately 1 cm² in the middle section of the second most external leaf was collected from one sampled shoot per individual fragment. If present, pen roots and hair roots were sampled. Sediment cores (20 cm depth x 5 cm diameter) were collected from the dead matte in close vicinity to the experimental restoration sites (n=20) and control meadows (n=10). From those cores, sediment samples of a volume of approximately 1 mL were collected at a depth of 1-10 cm, representing the seagrass's root depth. Seawater samples with a volume of 120 mL were collected above the dead matte at each of the seven experimental restoration sites, with two replicates per site (total n=12), and inside the seagrass meadows' canopy for the two control meadows, with three replicates per meadow (total n=6). The seawater was filtered using 0.22 µm Sterivex^{TM} unit with a sterile 120 mL syringe (MF-Millipore Membrane, Merck KGaA, Darmstadt, Germany). The leaves, roots, sediment samples, and filters were directly preserved in DNA/RNA Shield (ZymoResearch, California, USA) and stored at −20 °C until DNA extraction. Environmental contaminants were removed from the dataset using the above-mentioned sediment and seawater controls to ensure only seagrass-associated bacterial communities were retained for diversity analyses.

2.3 DNA extraction and amplicon sequencing

DNA was extracted from all samples using the Quick-DNA^{TM} Miniprep Kit (ZymoResearch, California, USA) following the manufacturer's instructions for 'Solid Tissue Samples' (page 6 of the manual). Filters from the Sterivex^{TM} casing were removed according to Cruaud et al. (2017) that demonstrated significantly increased DNA yields. For all the samples, including sediment and seawater filters, in the lysis step, tungsten beads, and an automatic homogenizer (Vortex-Genie® 2, Scientific Industries) (for 10 min at a maximum speed) were used for a more efficient mechanical lysis. After DNA extraction, the samples were sent to Novogene GmbH (Munich, Germany) for DNA amplification and sequencing. PCR was performed on extracted

DNA to amplify the V5–V7 region of 16S rRNA gene using the primer pairs 799F and 1193R (forward primer, 5′-AACMGGATTAGATACCCKG-3′; reverse primer, 5′-ACGTCATCCCCACCTTCC-3′) (Bodenhausen et al., 2013). The samples were pooled in equal proportions based on their molecular weight and DNA concentrations (using Qubit Invitrogen®) and purified using magnetic beads. The sequencing libraries were generated, and paired-end (2×250 bp) sequencing was performed on an Illumina MiSeq PE250 system following the manufacturer's guidelines.

2.4 Bioinformatic analysis

The raw dataset consisted of a total of 6,360,321 sequences. The sequences were depleted from barcodes and primer sequences and were trimmed for quality with the fastp (version 0.23.1) software. Sequences with ambiguous base calls, as well as chimeras, were removed. The de-duplicated or unique sequences were denoised using DADA2 (Callahan et al., 2016) to obtain initial ASVs. Taxonomy was assigned to ASVs using the SILVA reference database (version 138). From the resulting ASV table, eukaryotic organelle sequences (i.e., chloroplasts and mitochondria) and unassigned sequences were removed. The resulting absolute ASV table was used for all downstream analyses. Rarefaction curves were used to assess sampling depth (Figure S3.7). Due to the important differences in the number of sequences among samples (7312 – 70,118 sequences), the samples were normalized by rarefaction to the minimum number of sequences (7312) per sample to adjust for those differences (Figure S3.7). Library size normalization is required for meaningful alpha and beta diversity analysis. Therefore, the rarefied ASVs table resulted in 1,118,736 high-quality sequences, clustered in 40,028 ASVs.

2.4.1 Bacterial community richness and diversity: alpha diversity analysis

Before calculating alpha diversity indices, all the ASVs with a relative abundance above 0.01% in seawater and sediment samples were classified as 'environmental bacteria' and removed from the rarefied ASVs table. Bacterial community richness was assessed using the number of ASVs (S), while diversity was evaluated using the Shannon (H′) and Simpson (1-λ′) indices. The exponential function was applied to the Shannon's diversity index to determine the true Shannon diversity (i.e., the effective number of species), following the methodology outlined by Lundberg et al. (2012). The seagrass samples within the 'donor population of intermatte cuttings' did not have roots, which is why this level within the group factor 'sample tissue' is absent in the following analyses. The normality and linearity of the residuals were tested by visual inspection of the residuals versus fitted values plot and with a Shapiro-Wilks test. The homogeneity of variances was checked using Levene's test. Data visualisation and assumptions were checked using RStudio software version 4.3.2 (RStudio Inc., Boston, MA, USA). As the data were not normally distributed, and to maximize comparability with the beta diversity analysis (as in Aires et al., 2021), PERMANOVAs were used to determine significant differences between samples origins and the transplantation methods, according to sample tissue. Two two-factor PERMANOVAs were performed. The first PERMANOVA was computed with the following factors: 'Sample tissue' (fixed factor with two levels) and 'Transplantation method' (fixed factor with five levels). The second PERMANOVA was computed with the following factors: 'Sample tissue' (fixed factor with two levels) and 'Sample origin' (fixed factor with five levels). All the factors and respective interactions were tested. After square root transformation of the data, the resemblance matrix was constructed based on Euclidean distances, and the number of permutations was set to 999. Monte Carlo tests were performed when permutations were fewer than 100 (Anderson et al., 2008). Community richness, diversity indices and one-way PERMANOVAs were done using the PRIMER-E+PERMANOVA software version 7.0.24 (PRIMER-E, Auckland, New Zealand).

2.4.2 Bacterial community structure : beta diversity analysis

Differences in community structure were visualized with Canonical Analysis of Principal coordinates (CAP), based on a Bray-Curtis dissimilarity matrix after square root transformation of the rarefied ASVs table. CAP analysis was chosen as it allows to constrain the ordination based on explanatory variables, which is a better match for a priori hypothesis testing plots, enabling to assess specific relationships between sample groupings and environmental or experimental factors. PERMANOVAs were used to test for statistical significance of the differences among samples nature, samples origins, and transplantation methods. The same PERMANOVA designs as described in section 2.4.1 were used. Moreover, a one-way PERMANOVA test for the factor "Sample nature" (fixed factor with four levels) was performed to assess the differentiation among the seagrass samples and the environmental samples. Differential abundance analysis using Linear Discriminant Analysis (LDA) Effect Size (LEfSe) (Segata et al., 2011) was performed to identify the top 20 significant orders and ASVs contributing to the differences observed among groups. This analysis employed the Kruskal-Wallis rank test with an adjusted p-value threshold of 0.05. The Log LDA Score was set to 1.0, and significant orders and ASVs were ranked in descending order based on their LDA scores. The CAPs were done using RStudio software version 4.3.2, the PERMANOVAs were done using the PRIMER-E+PERMANOVA software version 7.0.24 and the LEfSe analysis was performed in MicrobiomeAnalyst (Dhariwal et al., 2017).

3. Results

3.1 Taxonomic composition at the order level

The three most abundant bacterial orders for the leaf samples of the control meadows were *Rhizobiales*, *Burkholderiales*, and *Bacillales,* while *Chromatiales*, *Corynebacteriales*, and *Desulfobacterales* were the three most abundant in the root samples of the control meadows. (Figure 3.35, 3.36; Table S3.8, S3.9).

Rhizobiales was also the first most abundant order of the leaves from transplants attached to iron staples, while it was the second most abundant order of the leaves of the transplants on the coconut fiber mats (Figure 3.35; Table S3.8). *Burkholderiales* was the most abundant order for those latter samples, while it was *Bacillales* for the leaves on the transplants on the BESE element and from cuttings of the donor populations (Figure 3.35; Table S3.8). For the roots, *Chromatiales* was the second most abundant bacterial order on iron staple samples, with *Microtrichales* being more abundant (Figure 3.35, Table S3.8). *Rhizobiales* was the most abundant for the coconut fiber mat samples, *Pseudomonadales* for the BESE element samples, and *Enterobacterales* for the donor populations samples (Figure 3.35; Table S3.8). Although *Desulfobacterales* was the third most abundant order in the roots of the control meadows (9.44%), it was only present in the roots of iron staples (0.84%) and coconut fiber mats (0.15%) samples (Figure 3.35; Table S3.8).

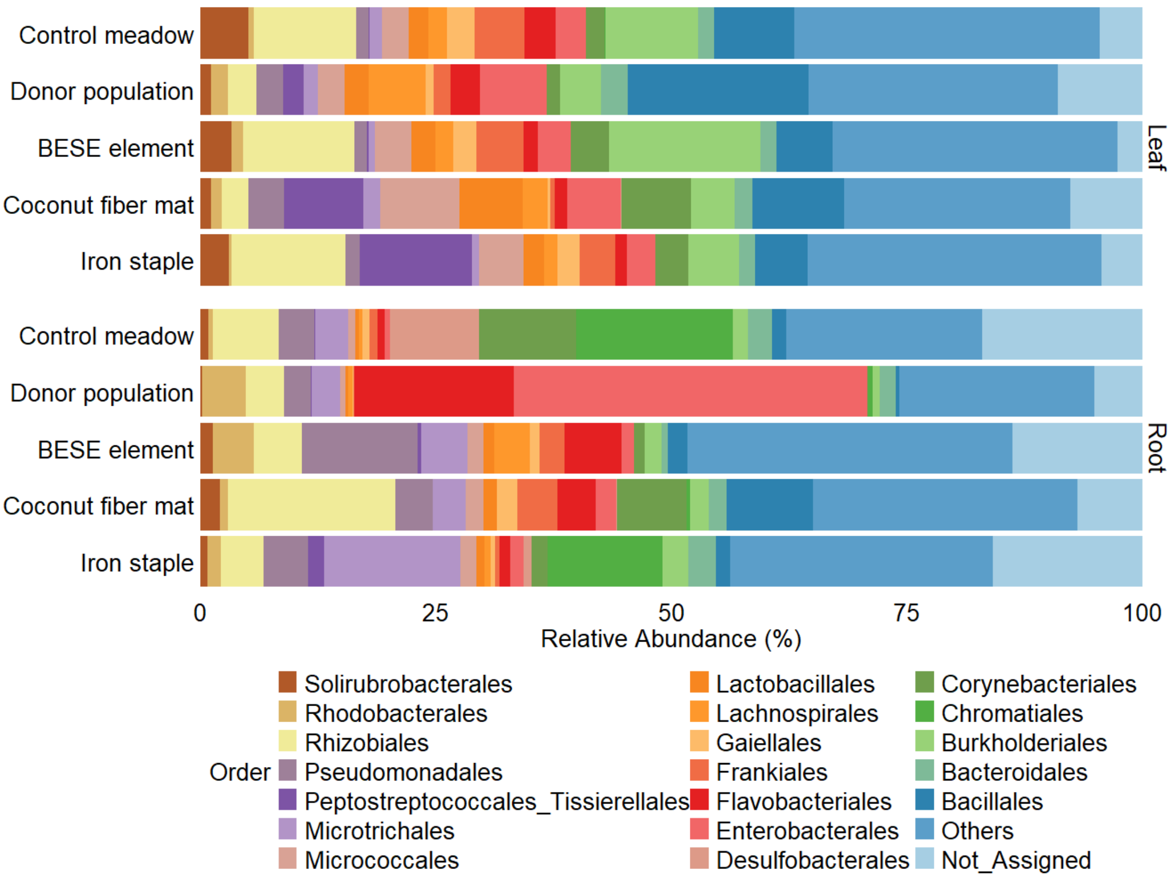


Figure 3.35. *Distribution of bacterial communities, at the order level, associated with the different tissues (leaf and root) of transplanted P. oceanica cuttings and control meadows as a function of transplantation method.*

For the samples grouped according to their origin, *Rhizobiales* was the most abundant order in storm-fragment leaf samples as in the control meadows leaf samples (Figure 3.36; Table S3.9). *Burkholderiales* was the most abundant in the leaves of intermatte cuttings, as well as in the leaves of storm-fragment donor population, while *Bacillales* was the most abundant in the leaves of intermatte cutting donor population (Figure 3.36; Table S3.9). *Microtrichales* were dominating the roots of storm-fragments while *Rhizobiales* were the most abundant in the roots of intermatte cuttings (Figure 3.36; Table S3.9). In those two groups, *Pseudomanadales* was the second most abundant order, followed by *Chromatiales* in the third position while it was the first most abundant order in roots of control meadows (Figure 3.36; Table S3.9). *Enterobacterales* was the most abundant order in the roots of the storm-fragment donor population (Figure 3.36; Table S3.9). Although *Desulfobacterales* was the third most abundant order in the roots of the control meadows (9.44%), it was only present in the roots of storm-fragments at a very low relative abundance (0.89 %) (Figure 3.36; Table S3.9).

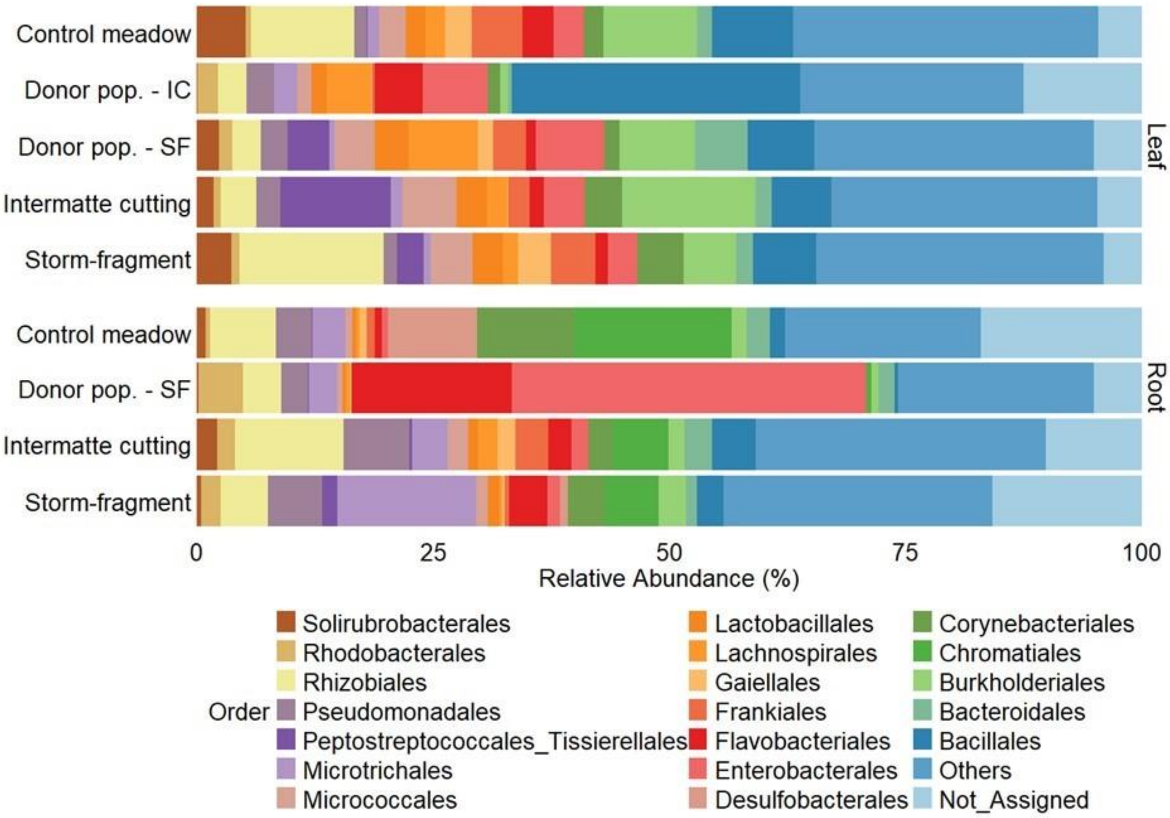


Figure 3.36. *Distribution of bacterial communities, at the order level, associated with the different tissues (leaf and root) of transplanted P. oceanica cuttings and control meadows as a function of sample origin. "Donor pop.-IC" - donor population of intermatte cuttings before transplantation, "Donor pop.-SF" donor population of storm-fragments before transplantation.*

3.2 Bacterial community richness and diversity: alpha diversity analysis

The effects of transplantation method, sample origin, and their interaction with sample tissue (leaf vs. root) on bacterial alpha diversity (number of observed ASVs, exponentiated Shannon index, and Simpson index) were evaluated. Among the three diversity metrics, only the number of observed ASVs showed significant differences for the factors transplantation method (Table S3.10), sample origin (Table S3.15), and their respective interaction with sample tissue (Figure 3.37; Table S3.10, S3.15). In contrast, no significant effects were detected for Shannon or Simpson indices (Figure 3.37B, C, E, F; Table S3.13, S3.14, S3.18, S3.19). Pairwise PERMANOVA tests indicated that the significant differences in the number of observed ASVs were driven exclusively by root samples. Roots from donor populations prior to transplantation exhibited significantly higher number of ASVs compared to roots from transplanted plants (all transplantation methods and origins) and control meadows (Figure 3.37, A, D; Table S3.11, S3.12, S3.16, S3.17). No significant differences were observed for leaves (Figure 3.37, A, D; Table S3.11, S3.12, S3.16, S3.17). All the *p*–values for the alpha diversity statistical analysis are reported in Supplementary Tables S3.10–S3.19.

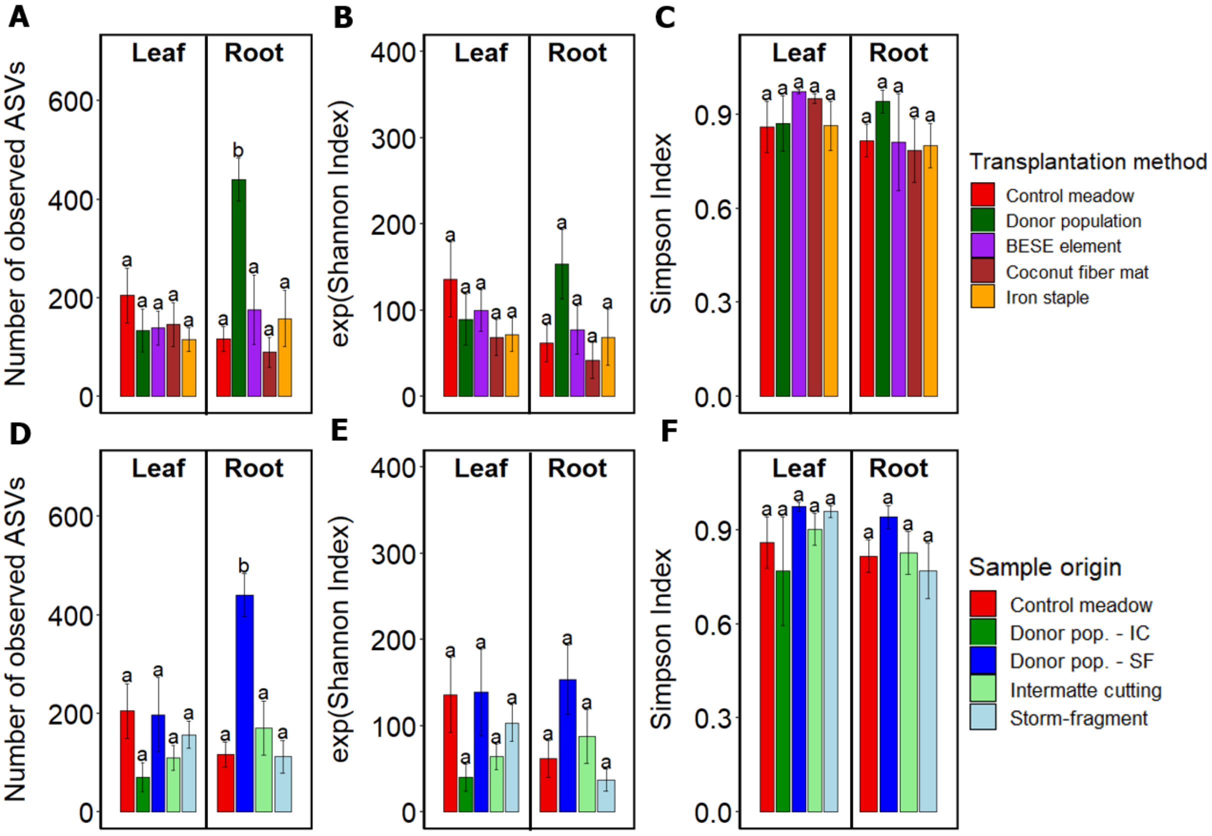


Figure 3.37. *Mean number of observed ASVs (**A**, **D**), exponentiated Shannon Index (**B**, **E**), and Simpson Index (**C**, **F**) of bacterial communities associated with the different tissues (leaf and root) of the transplanted P. oceanica seagrass cuttings and control meadows as a function of transplantation method (**A, B**, and **C**) and sample origin (**D**, **E**, and **F**). "Donor pop.-IC" - donor population of intermatte cuttings before transplantation, "Donor pop.- SF" donor population of storm-fragments before transplantation. Statistically significant differences (p<0.05) within tissues are represented by different lowercase letters (a, b).*

3.3 Bacterial community structure: beta diversity analysis

The variation in bacterial community structure (beta diversity) among sample types (leaf, root, sediment, water), transplantation methods, and sample origins was evaluated using CAP ordination, PERMANOVA, and Linear Discriminant Analysis (LDA) Effect Size (LEfSe). Bacterial community structure displayed a clear differentiation between unvegetated areas (i.e., dead matte) and vegetated areas (i.e., control meadow) for both water and sediment samples (Fig. 6; Table S16, S20). No differentiation was observed between sample tissues. Instead, clustering was primarily driven by transplantation method (Figure 3.38A) and sample origin (Figure 3.38B).

For the transplantation method, CAP ordination revealed that the control meadow samples were more similar to the transplants on iron staples (Figure 3.38A). Donor populations, transplants on BESE elements and coconut fiber mats formed a separate cluster (Figure 3.38A).

Pairwise PERMANOVA tests indicated that leaf communities did not differ significantly among transplantation methods or between transplants and control meadows (Table S3.23). In contrast, root communities of control meadows differed significantly from those transplanted on coconut fiber mats, BESE elements, and from donor populations (Table S3.23). No significant difference was detected between control meadow roots and those transplanted using iron staples (Table S3.23). Differential abundance analysis supported these results, highlighting several ASVs and bacterial orders enriched in control meadow roots compared to transplants on coconut fiber mats, BESE elements, and donor populations. The most notable were ASV23 (*Gammaproteobacteria*), ASV27 (*Candidatus* Thiodiazotropha), ASV79 (Desulfosarcinaceae), and the bacterial orders *Chromatiales*, *Desulfobacterales*, *Desulfobulbales*, and *Spirochaetales* (Figure 3.39; Table S3.28). Additional pairwise comparisons showed that the roots of donor populations before transplantation differed significantly from those transplanted on coconut fiber mats and iron staples, but not from those on BESE elements (Table S3.23).

For sample origins, CAP ordination showed three distinct clusters: one for control meadow samples, another grouping intermatte cuttings, storm-fragments, and donor populations of intermatte cuttings, and a third composed of donor populations of storm-fragments, which were the most dissimilar from control meadows (Figure 3.38B). Pairwise PERMANOVA tests showed no significant differences among leaf communities from different the different sample origins (Table S3.27). In contrast, root communities of control meadows differed significantly from those of storm-fragments and intermatte cuttings (Table S3.27). Differential abundance analysis revealed ASVs and bacterial orders driving these differences, including ASV79 (*Desulfosarcinaceae*) and ASV23 (*Gammaproteobacteria*), which were more abundant in control meadow roots compared to transplanted roots (Figure 3.40A; Table S3.29). ASV27 (*Candidatus*Thiodiazotropha) was also enriched in control roots relative to storm-fragments but showed slightly higher abundance in intermatte cuttings (Figure 3.40A; Table S3.29). Conversely, ASV19 (*Gammaproteobacteria*) was higher in storm-fragments compared to control meadows (Figure 3.40A; Table S3.29). At the order level, *Desulfobacterales*, *Chromatiales*, and *Desulfobulbales* dominated in control meadow roots compared to both intermatte cuttings and storm-fragments (Figure 3.40B; Table S3.29). Further pairwise tests showed that donor populations of storm-fragments before transplantation differed significantly from their transplanted counterparts two years later, as well as from intermatte cuttings and control meadows (Figure 3.38B; Table S3.29). No significant difference was detected between the two transplanted types (i.e., intermatte cuttings and storm-fragments) after two years (Figure 3.38B; Table S3.27).

All the *p*–values for the beta diversity statistical analysis are reported in Supplementary Tables S3.20–S3.27. The lowest taxonomical levels of the ASVs represented in Figure 3.39A and Figure 3.40A are reported in Supplementary Tables S3.28 and S3.29, respectively.

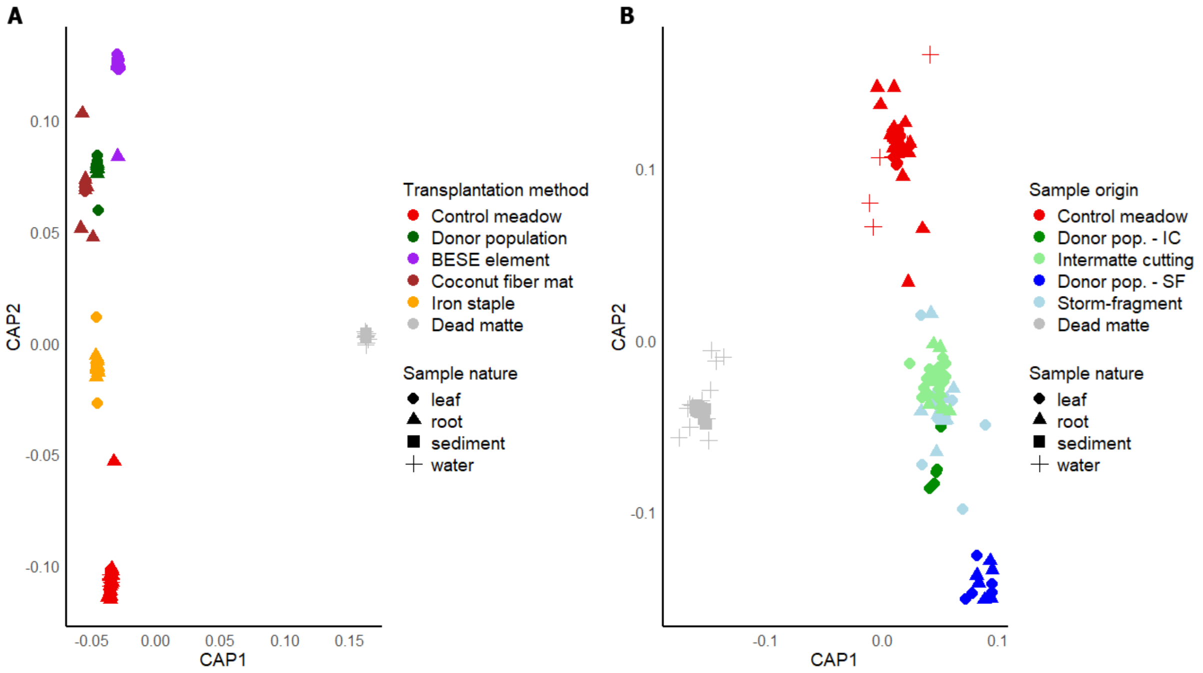


Figure 3.38. *Canonical analysis of principal coordinates (CAP) ordination plot based on Bray–Curtis dissimilarity matrix of square root transformed bacterial abundances showing canonical axes that best discriminate the bacterial communities associated with the different tissues (leaf and root) of the transplanted P. oceanica seagrass plants and control meadows, as well as sediment and seawater, as a function of transplantation method (**A**) and sample origin (**B**). "Donor pop.-IC" - donor population of intermatte cuttings before transplantation, "Donor pop.- SF" donor population of storm-fragments before transplantation.*

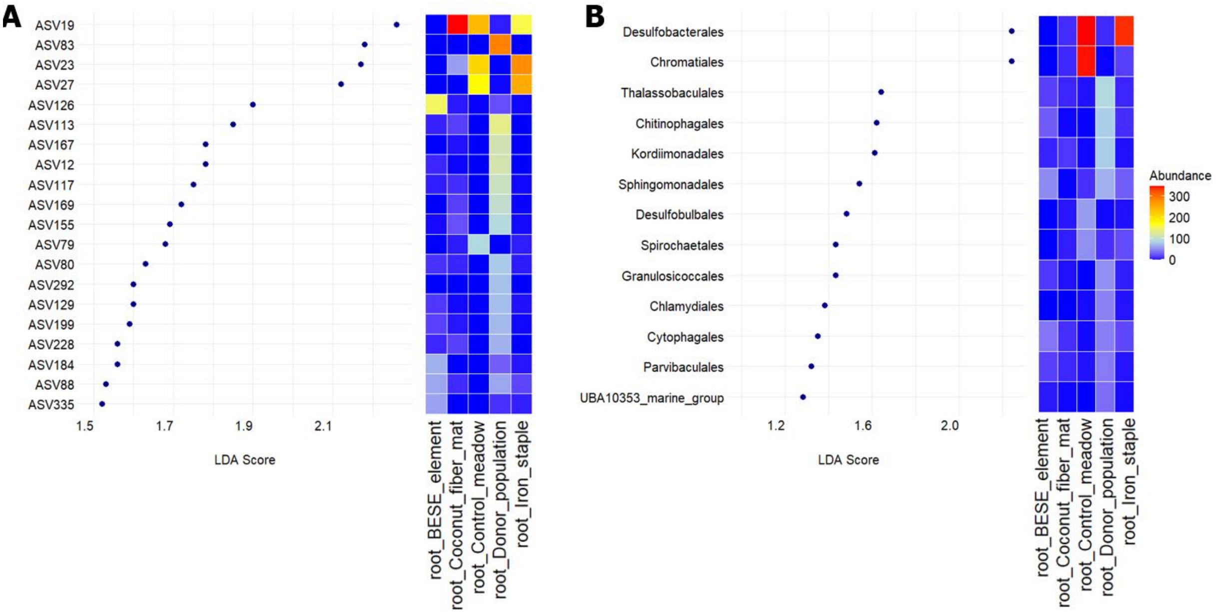


Figure 3.39. *Linear Discriminant Analysis (LDA) Effect Size (LEfSe) plots displaying the most differentially abundant (A) ASVs and (B) bacterial orders from P. oceanica seagrass roots according to the experimental factor 'transplantation method'. Differentially abundant features were determined using the Kruskal-Wallis rank test (adjusted p-value cut off = 0.05), with the Log LDA Score value adjusted to 1.0 and significant ASVs/taxa given in descending order from the highest to lowest LDA score.*

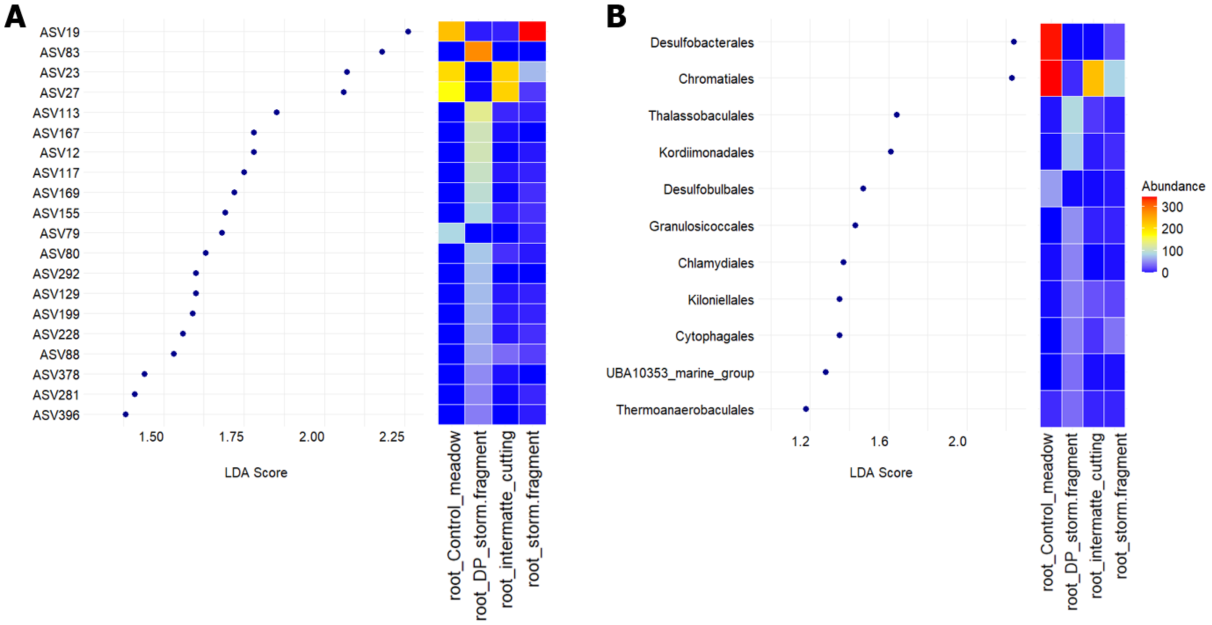


Figure 3.40. *Linear Discriminant Analysis (LDA) Effect Size (LEfSe) plots displaying the most differentially abundant (A) ASVs and (B) bacterial orders from P. oceanica seagrass roots according to the experimental factor 'sample origin'. Differentially abundant features were determined using the Kruskal-Wallis rank test (adjusted p-value cut off = 0.05), with the Log LDA Score value adjusted to 1.0 and significant ASVs/taxa given in descending order from the highest to lowest LDA score.*

4. Discussion

To our knowledge, this is the first study to investigate the influence of transplantation methods and donor origins on the bacterial communities associated with *P. oceanica* cuttings transplanted into dead matte areas. Overall, our findings show that while bacterial diversity remained broadly stable across treatments, the composition of root-associated microbiomes was strongly shaped by the transplantation method and to a lesser extent by the donor origin. Among the transplantation methods, iron staples promoted microbial assemblages most similar to control meadows, whereas coconut fiber mats and BESE elements led to more distinct communities. Moreover, donor origin influenced the abundance of specific bacterial taxa, such as *Candidatus* Thiodiazotropha, which was more abundant in intermatte cuttings compared to storm-fragments. These patterns suggest that both the physical characteristics of the transplantation method and the initial microbial pool associated with donor material play a critical role in shaping the microbial trajectory of seagrass roots after restoration.

4.1 Bacterial community dynamics in transplanted P. oceanica cuttings

The analysis of alpha diversity showed that the Shannon and Simpson indices remained similar across all treatments for both leaves and roots, indicating a consistent balance between species richness and evenness regardless of the transplantation method or donor origin. However, a notable pattern emerged for the roots of donor populations originating from storm-fragments, which exhibited significantly higher ASV richness compared to the roots of control meadows and transplanted cuttings. This elevated richness may reflect the presence of low-abundance taxa, which do not strongly affect diversity indices sensitive to dominant species. Such a pattern suggests that the roots of the donor populations experience opportunistic colonization by microbial taxa. The roots of the donor populations, originating from storm-fragments drifting on the seafloor without anchoring in sediment, likely encounter diverse microbial sources, enhancing their richness through exposure to a larger pool of water and sediment-associated bacteria. Indeed, surrounding sediment and seawater generally harbour a higher bacterial richness than seagrass tissues (Frasca et al., 2024; Martin et al., 2020).

Following transplantation, environmental conditions gradually stabilize, and this stabilization is mirrored in the bacterial communities, which progressively resemble those found in established control meadows (Martin et al., 2018; Wang et al., 2021). In these mature meadows, long-term interactions between roots and their environment promote the development of a more specialized and functionally optimized bacterial community. This results in a potentially reduced ASV richness and change in bacterial community structure, as the host plant selectively supports beneficial microbial taxa over time (Aires et al., 2016; Cúcio et al., 2016). Such specialized communities are shaped by plant-derived exudates and rhizosphere-specific gradients in oxygen and redox potential (Brodersen et al., 2024; Lebeis et al., 2015; Wang et al., 2021). Medium and long-term studies are needed to determine whether the roots of the transplants will eventually develop a bacterial structure similar to that of the control meadow, as root age plays a key role in microbial colonization in long-lived seagrasses such as *P. oceanica* (García-Martínez et al., 2009, 2024).

Furthermore, the roots of the control meadows were significantly enriched in the bacterial orders *Chromatiales*, *Desulfobacterales*, and *Desulfobulbales* compared to the roots of storm-fragments and intermatte cuttings two years after transplantation. *Chromatiales* have been identified as key bacterial groups dominating the rhizosphere of seagrasses (Cúcio et al., 2016, 2018) and salt marsh vegetation (Rolando et al., 2024; Thomas et al., 2014). *Chromatiales* are involved in sulfur oxidation processes, and it is thought that they are critical in mitigating sulfide toxicity within the root zones (Cúcio et al., 2016; Thomas et al., 2014). In addition, the most abundant genus among the *Chromatiales* was *Candidatus* Thiodiazotropha, which has been demonstrated as a key endosymbiont in the coastal cordgrass *S. alterniflora* (Rolando et al., 2024). Originally discovered in symbiosis with bivalves from the family *Lucinidae*, these endosymbionts fix carbon and provide both carbon and nitrogen to their host by harnessing energy from the oxidation of reduced sulfur compounds (König et al., 2016; Lim et al., 2019; Osvatic et al., 2021; Petersen et al., 2016). Coastal vegetated plants benefit from their symbiosis with members of the *Candidatus* Thiodiazotropha genus, as it helps mitigate sulfide toxicity (Martin et al., 2020) and links sulfide oxidation to carbon and nitrogen fixation. Although nitrogen is likely transferred to the plant host, the precise mechanism behind this transfer remains to be fully understood and warrants further investigation (Lehnen et al., 2016; Rolando et al., 2024). Secondly, *Desulfobacterales* and *Desulfobulbales* are sulfate-reducing bacteria (SRB) capable of nitrogen fixation, commonly found in high abundance within the root microbiome of seagrasses (Brodersen et al., 2024; Crump et al., 2018; Cúcio et al., 2016; Frasca et al., 2024; Rolando et al., 2024). Moreover, SRB can oxidize ethanol (Galushko & Rozanova, 1991) in the rhizosphere, potentially representing a mutually beneficial interaction between plants and bacteria. Indeed, despite producing hydrogen sulfide, these bacteria help detoxify the rhizosphere by metabolizing ethanol released by the plant roots (Cúcio et al., 2016). Furthermore, it could be hypothesized that a mutualistic relationship exists between *Desulfobacterales and Desulfobulables*, which produce sulfide, and *Chromatiales*, which uses the oxygen released by the seagrass roots as the terminal electron acceptor for sulfide oxidation (van der Heide et al., 2012). Finally, *Desulfobulbales* are not exclusively composed of SRB but also include genera known as cable bacteria (e.g., *Candidatus* Electrothrix), which can couple oxygen reduction with sulfide oxidation over centimeter-scale distances within the sediment (Brodersen et al., 2024; Malkin et al., 2021; Scholz et al., 2021). These bacteria may also enhance nitrogen availability for seagrasses by indirectly promoting dissimilatory nitrate reduction to ammonium (DNRA) through the dissolution of iron sulfides (Kessler et al., 2019) and/or by facilitating nitrogen fixation (Kjeldsen et al., 2019).

The essential functions provided by these bacterial orders strongly influence the health and productivity of seagrass meadows (Brodersen et al., 2024; Crump et al., 2018), particularly under stressful environmental conditions such as those induced by transplantation (Christiaen et al., 2013; Fuggle et al., 2023; Wang et al., 2021). Numerous studies have reported reduced morphological traits in transplanted *P. oceanica* compared to control meadows (Bacci et al., 2024; Boulenger et al., 2024; Pansini et al., 2024), yet no conclusive explanation has been established for this phenomenon. Further research is needed to determine whether the limited development of *P. oceanica* cuttings is directly linked to their associated bacterial communities.

4.2 Contribution of donor origins to bacterial communities associated with transplanted P. oceanica cuttings

Although the donor population of intermatte cuttings lacked initial roots at the time of transplantation, the intermatte cuttings successfully established microbial communities similar to those of the storm-fragments. This illustrates the ability of roots to recruit and stabilize functional microbial communities over time, even under disturbed conditions, by progressively shaping the microbial community as plants grow and modify the surrounding sediment (Brodersen et al., 2018, 2024; Fuggle et al., 2023; Wang et al., 2021).

Moreover, the diversity and overall structure of bacterial communities associated with storm-fragments and intermatte cuttings showed no significant differences two years after transplantation. From a microbiological perspective, this finding suggests that both donor origins are equally suitable for transplantation onto dead matte in a restoration context. However, the notably higher abundance of *Chromatiales*, particularly the genus *Candidatus* Thiodiazotropha, in the roots of intermatte cuttings raises intriguing questions about their potential functional advantages compared to storm-fragments. Given the critical role of this genus in sulfur oxidation and nitrogen fixation processes (Martin et al., 2020; Rolando et al., 2024), further research are needed to determine if the higher abundance of *Candidatus*

Thiodiazotropha could contribute to increased plant performance, such as higher nitrogen content in transplanted seagrass tissues.

Furthermore, mesocosm experiments involving the inoculation of specific strains from this bacterial genus, although these have yet to be isolated, could help clarify their direct contribution to nutrient cycling and plant health (Pugnaire et al., 2019; Zhou et al., 2024). In addition, ^{15}N-DNA stable isotope probing would provide valuable evidence of active nitrogen fixation by this genus within the roots of intermatte cuttings (Buckley et al., 2007; Morando et al., 2016; Reay et al., 2019). Such approaches could shed light on whether microbial differences, even when subtle, can influence the long-term success and resilience of transplanted *P. oceanica* cuttings.

4.3 Influence of transplantation methods in shaping bacterial communities associated with P. oceanica cuttings

As expected, our results showed that bacterial communities associated with *P. oceanica* roots are more affected by transplantation methods than those associated with leaves. Among the three tested transplantation methods, cuttings secured with the iron staples exhibited a bacterial community structure most similar to that of the control meadow. In contrast, marked dissimilarities were observed between bacterial community associated with the control meadow, and those associated with cuttings transplanted using coconut fiber mats, BESE elements, and even the donor populations.

The three transplantation methods differed in the material composition of anchoring structures used to attach the cuttings to the seafloor (i.e., iron, coconut fibers, or starch-derived polymers) and the level of structural complexity they provided. Coconut fiber mats and BESE elements offered greater structural complexity compared to the iron staples. Additionally, these methods varied in the distance maintained between the cuttings and the sediment surface.

4.3.1 Influence of transplantation material composition on root-associated bacterial communities

The three transplantation methods differ in the type of material used to anchor the cuttings to the seafloor. The composition of the coconut fiber mats and BESE elements could explain the differences in bacterial community structure compared to the control seagrass meadows. The coconut fiber mats consist of a natural coconut fiber woven mesh with a high lignin content and, therefore, an increased hydrophobicity and resistance to microbial degradation (Lekha et al., 2004; Nitsch et al., 2021; Rautenbach et al., 2024). However, high abundance of bacterial taxa specialized in lignin degradation was not observed in the root samples from the transplants growing on the coconut fiber mats. BESE elements are composed of biodegradable potato-waste-derived Solanyl C1104M (Rodenburg Biopolymers, Oosterhout, the Netherlands), which could likely serve as a carbon source for microbial colonization (Liu et al., 2018). The most differentiating bacterial taxon between the roots of the transplants on the BESE elements and the plants from the other groups was ASV126, which belongs to the order *Pseudomonadales*. *Pseudomonadales* abundance is influenced by nutrient availability, particularly ammonium and phosphate, and they thrive in environments rich in labile organic carbon (de Vogel et al., 2024). Laboratory experiments on BESE elements biodegradation have shown that this compound releases a significant amount of dissolved organic carbon, soluble reactive phosphorus, and nitrate (Liu et al., 2018), which might have favoured *Pseudomonadales*. Moreover, members of this order are key contributors to the degradation of different biodegradable polymers (Rubio-Portillo et al., 2021; Sun et al., 2023). Furthermore, *Pseudomonadales* have also been found to be highly abundant in *P. oceanica* 'banquettes' (Boudouresque et al., 1982), which consist of banks of dead leaf material on the beaches (Egan et al., 2013). These bacteria are common in copiotrophic communities as they possess polymer-degrading enzymes (Lin et al., 2019; Offret et al., 2016; Skovhus et al., 2004), as well as ligninolytic and chitinolytic activity (Lin et al., 2019; Paulsen et al., 2019) which makes them effective in seagrass leave decomposition (Trevathan-Tackett et al., 2020). Given the significant accumulation of dead *P. oceanica* leaves within the BESE elements (Figure S3.8), this deposition could also explain the higher presence of *Pseudomonadales* in the roots of transplants on BESE elements compared to the other transplantation methods tested in this study. Further studies are needed to compare the core microbiome of the bacterial biofilm developing on the surface of restoration substrates with the root microbiome of the transplants. This would help assess the extent to which the transplantation material leaves its bacterial signature on the root microbiome of the seagrass transplants.

4.3.2 Effects of transplantation material structure and sediment contact on root-associated bacterial communities

Besides material composition, the three transplantation methods also differed in the height of the cuttings relative to the sediment and the underlying dead matte. The rhizomes and roots of the cuttings attached with iron staples have direct contact with the dead matte. In contrast, the cuttings on the coconut fiber mats are separated from the dead mat by the 5 mm thickness of the coconut fiber mats. As for the cuttings on the BESE elements, these layers measure 6 cm in height, creating a gap between the roots of the cuttings and the dead matte. This variation in positioning could influence the degree of interaction between the roots and the sediment microbial pool, affecting the recruitment and establishment of bacterial communities. Indeed, it is well established that plants recruit their root-associated microbiome from a larger pool of soil microbes, and the initial structure of this microbial pool plays a critical role in shaping the structure of root microbial communities (Bonito et al., 2014; Cúcio et al., 2016; Haney et al., 2015; Hartman et al., 2018). Cuttings anchored closer to the sediment may have increased exposure to beneficial sediment-associated bacteria. In contrast, elevated cuttings could encounter different oxygen and nutrient gradients, potentially promoting the proliferation of distinct bacterial groups. This could explain the observed differences in community structure and the varying degrees of similarity to the control meadows' bacterial communities. The reduced abundance of *Chromatiales* and *Desulfobacterales* in transplants on coconut fiber mats and BESE elements may be linked to limited initial recruitment due to reduced exposure to the sediment microbial pool, delaying the establishment of beneficial plant-microbe interactions. The use of iron staples appears to promote a more rapid microbial recovery, likely due to the direct contact between the roots and the sediment microbial pool, which closely resembles the microbial community of control meadows, despite differences in the dead matte bacterial community structure.

Moreover, empirical observations of the sampled cuttings revealed significant differences in root length and complexity two years after transplantation, whereas there were no initial differences at the time of planting (Figure S3.9). The roots of transplants on coconut fiber mats and BESE elements were notably smaller than those of control meadows and transplants on iron staples (Figure S3.9). Plant exudates released by the roots into the sediment promote microbial colonization through chemotaxis and attract key microbial partners that enhance plant fitness within the seagrass rhizosphere (Crump et al., 2018; Sogin et al., 2022; Zhang et al., 2024). This interaction might be weaker or delayed in transplants on coconut fiber mats and BESE elements due to their limited initial contact with the sediment microbial pool. Further research is needed to determine the influence of bacterial communities on the root system development of *P. oceanica transplants*.

4.4 Perspectives for microbiome-driven seagrass restoration

The results discussed in this study highlight the effects of transplantation methods and donor origins on the bacterial communities associated to *P. oceanica* transplants and point to several promising research and application pathways. A key next step involves extended monitoring to evaluate the medium-term (5 years) and long-term (10 years) dynamics of microbial communities in transplants compared to control meadows. Such monitoring would clarify whether the observed differences in bacterial community composition between transplants and control meadows attenuate over time and whether distinct transplantation methods and donor origins ultimately converge toward similar bacterial assemblages. Moreover, the bacterial orders *Desulfobacterales* and *Chromatiales* emerged as key contributors to the dissimilarity between control meadows and transplants. Further research is now warranted to elucidate how these taxa influence the overall fitness of transplanted seagrasses.

Furthermore, managing or manipulating microbial functions and communities are widely recognized as established methods in the bioremediation of terrestrial and aquatic ecosystems (Trevathan-Tackett et al., 2019), and could be applied to marine ecosystem restoration. These methods leverage beneficial microbial interactions to optimize nutrient cycling, enhance plant stress tolerance, and accelerate ecosystem recovery (Trevathan-Tackett et al., 2019; Sun et al., 2024). For example, plant growth-promoting rhizobacteria have demonstrated their effectiveness in enhancing seagrass growth, improving biomass production, rhizome elongation, and nitrogen uptake while also mitigating sulfide toxicity through microbial shifts in sulfur and iron cycling (Sun et al., 2024; Zhou et al., 2024). Further research is needed to assess the effects of inoculating *Desulfobacterales* and *Chromatiales* strains into *P. oceanica* cuttings and to evaluate their potential influence on transplant morphology, growth, and overall development. Tailored pre- and probiotic treatments could help optimize microbial consortia, as demonstrated by their success in terrestrial and aquaculture systems (Fuggle et al., 2023; Trevathan-Tackette et al., 2019). Collectively, these approaches could not only enhance initial transplant success but also ensure the long-term stability and ecological functionality of restored meadows.

5. Conclusion

To our knowledge, the present study is the first to investigate the succession of bacterial communities associated with the leaves and roots of *P. oceanica* transplants in a restoration project using different transplantation methods and donor origins. Our results reveal that while the overall alpha diversity of bacterial communities remains relatively stable across treatments, the root-associated microbiome exhibits pronounced shifts in composition compared to control meadows, particularly in the abundance of key bacterial orders such as *Chromatiales* and *Desulfobacterales*. Among the tested approaches, cuttings anchored with iron staples developed bacterial communities most similar to those of natural meadows, highlighting the critical role of direct sediment contact in facilitating the recruitment of functionally beneficial microbial partners. Conversely, cuttings transplanted on coconut fiber mats and BESE elements displayed more distinct microbial assemblages, likely influenced by differences in material composition, structural complexity, and sediment interaction.

Furthermore, the study shows that donor origin (storm-fragments or intermatte cuttings) does not significantly affect the long-term structure of root microbiomes two years after transplantation. This suggests that both sources are microbiologically suitable for restoration, although subtle differences in specific taxa, such as the higher abundance of *Candidatus* Thiodiazotropha in intermatte cuttings, raise intriguing questions about potential functional advantages related to nutrient cycling and sulfide detoxification.

Despite initial differences in bacterial community structure, the transplants displayed progressive stabilization towards conditions like those of control meadows, indicating the potential for long-term success with appropriate management strategies. These results emphasize the need for long-term monitoring to assess the full recovery and resilience of bacterial communities over time. By combining optimized transplantation methods with microbiome-targeted interventions, future restoration efforts could accelerate ecosystem recovery and enhance the stability and functionality of restored seagrass meadows. Overall, this study provides a foundational understanding of how transplantation methods and donor origins influence microbiome dynamics, laying the groundwork for improved methodologies that leverage beneficial plant-bacteria interactions for the sustainable recovery of degraded seagrass ecosystems.

Physiological and biochemical performance indicators of *Posidonia oceanica* transplantation success

Arnaud Boulenger[a,b], Ana Lucía Azul Acuna[a], Stéphane Roberty[c], Gilles Lepoint[d] , Michel Marengo[b], Sylvie Gobert[a,b]

[a] Laboratory of Oceanology, MARE Centre, UR FOCUS, University of Liege, 11 allée du six août, 4000, Liege, Belgium

[b] STAtion de REcherche Sous-marines et Océanographiques (STARESO), 20260 Calvi, France

[c] Laboratory of Animal Physiology and Ecophysiology, University of Liege, 4 Chemin de la Vallée, 4000 Liege, Belgium

[d] Laboratory of Trophic and Isotope Ecology (LETIS), UR FOCUS, University of Liege, 15 allée du six août, 4000, Liege, Belgium

Ce chapitre a été publié dans Marine Pollution Bulletin (2026). Le chapitre présenté ici est la version avant publication tel que présenté lors de la défense de thèse le 20 novembre 2025. La version finale de l'article peut être trouvée dans ce lien : https://orbi.uliege.be/handle/2268/337850
La partie Material & Methods de ce chapitre a été complétée sur base de l'article 4 : Boulenger, A., Roberty, S., Lopez Velosa, M. M., Marengo, M., & Gobert, S. (2024). The use of photo-biological parameters to assess the establishment success of Posidonia oceanica cuttings after transplantation. Water, 16(12), 1702.

Abstract

Evaluating the success of *Posidonia oceanica* restoration is challenging due to the species' slow growth and delayed structural responses. This three-year study in Calvi Bay (Corsica, NW Mediterranean) examined how transplantation method (iron staples, coconut fiber mats, BESE elements), donor origin (intermatte cuttings vs. storm-fragments), and depth (20 m vs. 28 m) influence the physiological and biochemical performance of transplanted cuttings. Plant responses were assessed through photosynthetic activity, leaf elemental content (C, N, P, S), and rhizome carbohydrate reserves. Transplanting depth had limited effects on the measured characteristics. The transplanting method, influencing root development, suggests distinct strategies for resource acquisition without altering physiological performance. In contrast, donor origin emerged as the main driver of variability: intermatte cuttings consistently showed higher nitrogen and phosphorus contents, lower C:N ratios, and faster convergence towards natural meadow trait profiles than storm-fragments. Multivariate analyses revealed early convergence (12–24 months) between intermatte cuttings and natural meadows, whereas storm-fragments remained distinct; by 36 months, both donor types again diverged from reference conditions. These results demonstrate the value of trait-based approaches, particularly eco-physiological indicators, as sensitive, early measures of transplantation success, complementing traditional structural metrics. Full convergence with reference meadows appears to be a long-term process, emphasizing the importance of extended monitoring and careful donor selection to improve seagrass restoration outcomes.

Keywords

Seagrasss, Restoration, Nutrients, Sulfur intrusion, Physiology, Photosynthetic activity, Carbohydrates

ACKNOWLEDGMENTS

This work was supported by the University of Liege (grant FSR2021) and the Fonds National de la Recherche Scientifique — FNRS (grants ASP 40006932 and CDR J.0076.23). This study is part of the STARECAPMED (STAtion of Reference and rEsearch on Change of local and global Anthropogenic Pressures on Mediterranean Ecosystems Drifts) project funded by the Territorial Collectivity of Corsica and by the Rhone-Mediterranean and Corsican Water Agency. The authors are grateful to STARESO for facilities and field assistance. Authors are grateful to Cédric Delforge for field work and samples processing assistance, as well as ICP-MS analyses. The authors have no conflicts of interest to declare.

AUTHORS’ CONTRIBUTIONS

AB : Conceptualization, Field Sampling, Methodology, Formal Analysis, Investigation, Writing, Visualization; ALAA: Formal Analysis, Investigation, Writing; SR: Conceptualization, Methodology, Formal Analysis, Investigation, Writing, Visualization, Supervision; GL: Methodology, Formal Analysis, Investigation, Writing; MM: Conceptualization, Writing, Supervision, Funding; SG: Conceptualization, Methodology, Writing, Supervision, Funding.

1. Introduction

Seagrasses form underwater meadows in the photic zones of temperate and tropical coastlines and are widely recognized as foundational habitat-forming species (Den Hartog & Kuo 2006, 2006; Larkum et al., 2006). These highly diverse and productive ecosystems (Vieira et al., 2024) fulfil important services such as carbon sequestration (Fourqurean et al., 2012) and protection against coastal erosion (Ganthy et al., 2015). Seagrasses display considerable variability in morphology and life-history traits. Some species, such as Cymodoceaceae, produce short-lived shoots with rapid growth and decay cycles, whereas others, like Posidoniaceae, are slow-growing and long-lived (Larkum et al., 2006). Despite their polyphyletic origins and morphological diversity, all seagrasses share a suite of adaptations to the marine environment: (1) complete submersion of plant structures; (2) tolerance to saline conditions; (3) a robust anchoring system of roots and rhizomes capable of nutrient uptake and survival in anoxic sediments; and (4) hydrophilous pollination mechanisms (Lakrum et al., 2006; Jackson et al., 2009). Their photosynthetic machinery is highly plastic, enabling acclimation to variable light conditions, providing protection from photoinhibition in clear tropical waters and enhancing light capture in dimmer temperate environments (Cummings & Zimmerman, 2003; Ralph et al., 2002). However, seagrasses require light intensities 10-20 times higher than many marine autotrophs, making them particularly vulnerable to habitat disturbances, often driven by human activity (Duarte, 1991; Short & Wyllie-Echeverria, 1996). Over recent decades, seagrass meadows have faced extensive declines, with global loss rates averaging 7% per year in the late 20th century (Waycott et al., 2009). Key drivers include sediment and nutrient runoff, coastal development, marine heatwaves, dredging, trawling, anchoring, and disease (Turschwell et al., 2021). The degradation and loss of seagrass meadows undermine the vital ecosystem services they provide, prompting increasing global effort to conserve and restore them (Unsworth et al., 2022, 2024). Restoration is now widely implemented as a management tool to slow degradation, promote recovery of impacted meadows, and safeguard ecosystem functions and services (Rezek et al., 2019; van Katwijk et al., 2016).

Assessing restoration success, however, remains challenging. Monitoring has traditionally relied on simple metrics such as transplant survival, but this binary measure provides limited insight into plant health or functional recovery. Comparisons with reference meadows are rarely included, and changes in shoot condition are often overlooked (Pansini et al., 2022). Moreover structural indicators (e.g., shoot density, biomass) often fail to effectively monitor recovery processes after disturbances or restoration actions, especially for larger, slow-growing seagrass species like *Enhalus* or *Posidonia* spp. (Marbà & Duarte, 1998; Roca et al., 2016). In contrast, physiological and biochemical indicators can reveal stress responses and functional adjustments at earlier stages, offering sensitive tools for evaluating restoration outcomes (Cooke & Suski, 2008; Roca et al., 2016; Boulenger et al., 2024). These metrics capture the organism's regulatory capacity to cope with new environmental conditions, critical for both degradation and restoration contexts (Adolph, 1965; Horn et al., 2009), and can guide donor selection by identifying populations best suited for transplantation (Cooke & Suski, 2008).

Here, *Posidonia oceanica* (L.) Delile, a slow-growing, Mediterranean-endemic seagrass characterized by high morphological and physiological plasticity (Hemminga & Duarte, 2000), was used as a model species to evaluate the role of physiological and biochemical traits in transplantation methods and donor sources performance. The availability of donor material for transplanting is one of the main constraint in *P. oceanica* restoration endeavours. The use of germinated seedlings from floating or beach-stranded seeds is challenging (Mancini et al., 2024; Sutera et al., 2024), as episodes of mass flowering are unpredictable, irregular in space and time (Marín-Guirao et al., 2019; Montefalcone et al., 2013; Stipcich et al., 2024), and do not always result in fruit production (Balestri & Cinelli, 2003). Another option is using fragments of *P. oceanica* rhizomes either extracted from donor meadows, which is a destructive harvesting impacting natural meadows, or collected from naturally detached fragments of unknown origin. A significant amount of seagrass fragments (later on referred to as storm-fragments) are dislodged during storms and accumulate in natural storage areas (Boulenger et al., 2025a) characterized by sandy intermattes within *P. oceanica* meadows (Abadie et al., 2015; Gobert et al., 2016). Although storm-fragments provide a good opportunity as donor material for transplantation while minimizing the impact on the surrounding natural meadows, there remains uncertainties in the performance of those fragments compared to cuttings manually excised from healthy meadows. Indeed, as the origin and life-history of the storm-fragments are unknown, their ability to survive for extended periods of time is uncertain (Balestri et al., 2011). Moreover, shading and sediment deposition, as well as the lack of belowground nutrients absorption by the roots (Lepoint et al., 2004) could conversely affect their growth rates, metabolism and carbohydrates storage (Lai et al., 2020; Kraemer & Alberte, 1995).

In this study, transplantation trials were conducted at shallow and deep sites in meadows degraded by boat anchoring, testing multiple transplantation methods and donor sources. As recommended by Roca and colleagues (2016), a multi-trait approach combining indicators of photosynthetic activity (Fv/Fm, α, rETRmax, Ek), leaf elemental composition (C, N, S, P), and rhizome carbohydrates reserves (sucrose, starch, total carbohydrates) was applied. The study aimed to: (1) assess the influence of transplantation method, donor source, and transplantation depth on physiological and biochemical traits of *P. oceanica* cuttings; (2) determine whether certain donor sources develop trait profiles resembling those of reference meadows; and (3) evaluate the timescales over which such convergence occurs.

2. Material and Methods

2.1 Study area and seagrass transplantation

This study was carried out between May 2022 and May 2025 in Alga Bay, a sub-bay of Calvi Bay (8°43′52′′ E; 42°34′20′′ N) located in front of the STARESO oceanographic research station (Calvi, NW Corsica, France). The bay hosts an extensive *P. oceanica* meadow, covering approximately 0.78 km² and extending from 3 to 37 m in depth (Abadie et al., 2016). Intensive anchoring activity has caused significant seagrass decline (Fullgrabe et al., 2022) and limited natural recolonisation (Boulenger et al., 2025a), prompting restoration efforts (Boulenger et al., 2024). Seven anchoring patches (i.e. dead matte patches surrounded by meadows) were selected as experimental sites; three at 20 m and four at 28 m depth. In spring 2022, a total of 693 *P. oceanica* fragments (i.e., a living rhizome with a couple of foliar shoots; with 99 cuttings per site) were transplanted as part of a pilot restoration project designed to test transplantation methods prior to upscaling (see Boulenger et al., in review). Both storm-fragments and cuttings extracted from *P. oceanica* meadows were used as donor sources in this study to test their physiological and biochemical performance three years after transplantation. A total of 462 storm-fragments were collected by SCUBA divers near the STARESO at depths of 6-28 m, while 231 cuttings were manually harvested from the erosion edge of a natural sandy intermatte at 15 m depth (Gobert et al., 2016). Harvesting from eroding edges was chosen to minimize disturbance to intact donor meadows, as these zones naturally produce fragments when matte structure degrade (Gobert et al., 2016). Furthermore, shoots obtained from erosion edges exhibit similar photosynthetic efficiency, leaf surface area, and biomass to those from undisturbed meadows at the same depth (Abadie et al., 2017; Lapeyra et al., 2016). All harvested material was stored in outdoor flow-through seawater aquaria until biometric measurements were performed. Only cuttings with at least three shoots and a plagiotropic rhizome of at least 15 cm in length were retained, while those with severe leaf necrosis were discarded. After initial biometric measurements, selected cuttings were transplanted into the experimental sites using three different biodegradable materials: (i) iron staples, (ii) biodegradable mat in natural coconut fibre woven mesh (referred to as coconut fiber mat), and (iii) BESE elements (BESE Ecosystem Restoration Products, Culemborg, The Netherlands). BESE elements are biodegradable sheets made of potato-waste-derived Solanyl C1104M (Rodenburg Biopolymers, Oosterhout, the Netherlands) stacked together to form a 6-cm high 3D honeycomb-shaped matrix.

2.2 Sampling strategy

Seven field campaigns were conducted between May 2022 (initial transplantation) and May 2025. Six post-transplantation monitoring campaigns were carried out at 3, 12, 15, 24, 27, and 36 months. These included three spring surveys (12, 24, 36 months; April-June), and three fall surveys (2, 15, 27 months; September-October). Prior to transplantation, 20 *P. oceanica* fragments were collected from nearby reference meadows at 20 and 28 m depth, along with 20 cuttings, comprising both storm-fragments and cuttings from intermattes, set aside for physiological and biochemical analyses. At each monitoring campaign, *in situ* survival and shoot production were assessed following Boulenger et al., (in review). Foliar shoots were sampled twice annually using the Non-Destructive Shoot sampling Method (NDSM; Gobert et al., 2020). At each site (n=7), 12 foliar shoots were sampled, resulting in 84 sampled shoots per campaign, plus 10 control shoots from reference meadows at 20 m and 28 m depth. Whole cuttings (rhizome with roots and foliar shoots) were sampled annually during spring to assess rhizome carbohydrate storage. Six rhizomes per site were sampled, along with 10 complete fragments from reference meadows at 20 and 28 m.

2.3 Photosynthetic activity measurements

After sampling, leaves from both transplanted and control plants were transported to the laboratory under shaded conditions. Chlorophyll *a* fluorescence analysis was used to assess photo-physiological performance, as it provides sensitive indicators of plant stress and acclimation (Gera et al., 2012; Larkum et al., 2007; Madonia et al., 2021). Photosynthetic activity was measured with a Pulse-Amplitude-Modulated (PAM) chlorophyll fluorometer, widely applied tool for seagrass health assessment (Belshe et al., 2007; Gobert et al., 2015; Madonia et al., 2021). Photosynthetic activity was measured in the laboratory using a DIVING-PAM-I (Heinz Walz GmbH; hereafter referred to as a PAM device). PAM measurements were taken on the convex middle section of the second intermediate leaf, which showed the strongest correlation with the photosynthetic rate of the whole shoot (Buia et al., 1992; Lassauque, 2009). Visible epiphytic growth on this section was removed by rubbing the leaf with a finger. To ensure standardized measurements, leaf holder clips were utilized during PAM measurements to maintain a constant distance between the tip of the fiber optic and the leaf surface (Lassauque, 2009). Rapid light curves (Ralph & Gademann, 2005) were obtained by exposing the samples for 10 s to 9 sequential increasing light steps (0, 38, 117, 237, 377, 564, 775, 1139, and 1548 µmol photons m^{-2} s^{-1}). These RLCs were obtained with the following settings: GAIN = 5, DAMP = 2, MEAS-INT = 2, SAT-INT = 8, and SAT-WIDTH = 0.8. Four parameters were recorded: maximum photochemical quantum yield (Fv/Fm), maximum relative electron transport rate (rETRmax), photosynthetic efficiency (α), and saturation irradiance (Ek). Fv/Fm was measured at the beginning of each RLC, i.e., at a photosynthetic photon flux density (PPFD) of 0 μmol photons m^{-2} s^{-1}. The effective photochemical quantum yield of the Photosytem II (Y(II)) and relative electron transport rates (rETRs) were calculated at the end of each of light step as Y(II) = (Fm′–F)/Fm′ and $rETR_{PSII}$= Y(II) × PPFD. rETRmax, α and Ek were derived from the RLCs and plotted as the rETR versus the photosynthetic photon flux density (PPFD) (Figure 3.41). These parameters were derived from the equation introduced by Platt et al. (1980), considering photoinhibition. Data acquisition and modelling were carried out using WinControl-3 software version 3.33 (Heinz Walz GmbH, Effeltrich, Germany).

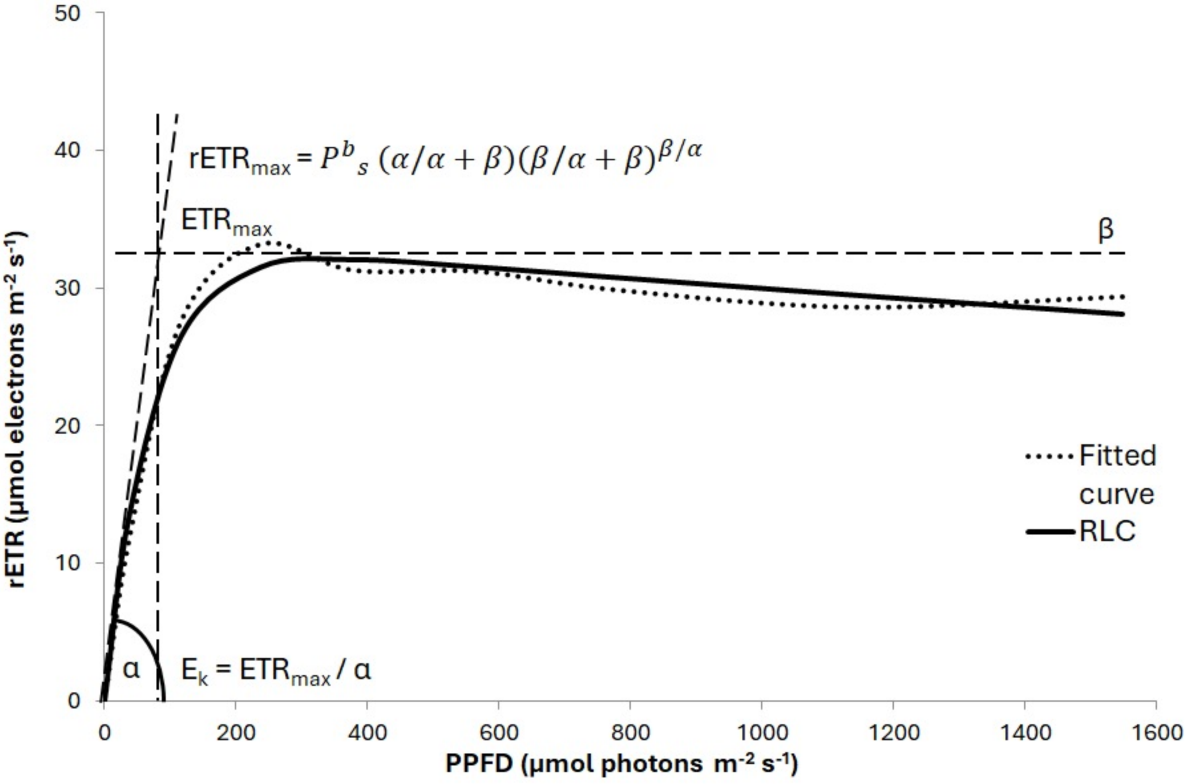


Figure 3.41. *Rapid light curve on which the relative electron transport rate is plotted against the PPFD. The fitted curve is plotted with a dotted line, and the rETRmax, Ek, and α are displayed.*

2.4 Leaves' elemental concentrations

Following photosynthetic activity measurements, epiphytes were scraped from all sampled leaves using a ceramic scalpel blade (Dauby & Poulicek, 1995). Leaves were weighed fresh, oven-dried at 60°C for 48 h and reweighed to determine dry biomass. For phosphorus analysis, dried leaves were shredded with ceramic scissors to facilitate homogenization during the mineralization process. Approximately 100 mg of dried powders per sample was digested in Teflon bombs using a closed microwave digestion system (Ethos D, Milestone Inc.) with nitric acid-hydrogen peroxide (HNO_3/H_2O_2; suprapur grade, Merck), following Richir & Gobert (2014). Ten analytical blanks were prepared to establish detection (LD) and quantification limits (LQ). The quantity of material placed in each bomb varied between 80 and 120 mg, depending on the quantity of available dried leaves powder for each sample. Phosphorus concentrations in the samples were determined using Inductively Coupled Plasma Mass Spectrometry with the Dynamic Reaction Cell technique (ICP-MS ELAN DRC II, Perkin Elmer), following the method described by Richir & Gobert (2014). Accuracy was verified using a Certified Reference Material (GBW 07603 bush branches and leaves). The calculated LDs and LQs were based on the measurement distribution over their respective blanks, following the recommendations of Currie (1999). Values consistently below LD were excluded from the dataset. Remaining coarse powders were ground to fine consistency and analysed for carbon, nitrogen and sulfur content with a C-N-S elemental analyser (Carlo Erba, Italy). Results are expressed in % of dry weight.

2.5 Carbohydrate storage in rhizomes

The rhizome were cleaned of scales, frozen at -20°C, and sent to MicroPolluants Technology SA (Saint Julien Les Metz, France) for the analysis of soluble carbohydrates and starch content. Each rhizome sample was placed in ethanol (*v*/*v*) and heated at 80 °C for 15 min to extract sucrose; the extract was then centrifuged to separate the solid part from the organic phase. The solvent was removed, and the extraction process was repeated twice (Zimmerman et al., 1989). The combined ethanol extract obtained was evaporated to dryness at room temperature, and the residue was dissolved in hot water. Starch was extracted from the sample pre-extracted from ethanol by incubation in sodium hydroxide solution for 24 h at room temperature (Gera et al., 2013) or by boiling it in sodium hydroxide for 30 min (Huber & Israel, 1982). After cooling, the pH was adjusted to 5.5 with acetic acid. The content of sucrose and starch was then determined by spectrophotometry after a reaction with anthrone (Yemn & Willis, 1954). Results are expressed as total carbohydrate reserves (TCR), sucrose and starch, with an accuracy of 1%.

2.6 Data analysis

2.6.1 Univariate

To assess the effects of the different experimental treatments on the physiological and biochemical traits of *P. oceanica transplants*, Generalized Linear Mixed Models (GLMMs) were used. Fixed factors included in the GLMMs were 'Transplantation method' (three levels: iron staple, coconut fiber mat, and BESE element), 'Donor source' (two levels: intermatte cutting and storm-fragment), and 'Bathymetry' (two levels: shallow and deep). The experimental site and time of sampling were selected as random factors. A Gamma distribution with a log link function was used for all traits. GLMs were built using the *glm* function in RStudio software version 4.3.2 (RStudio Inc., Boston, MA, USA). Model selection was guided by Akaike's Information Criterion (AIC), progressively removing non-significant terms based on statistical criteria until no further variables could be eliminated. Overdispersion was assessed by comparing the residual deviance to the residual degrees of freedom. To test the statistical significance of differences between treatments, estimated marginal means (EMMs) were computed using the *emmeans* function in RStudio, applying Bonferroni correction to adjust p-values for multiple comparisons.

In addition to the GLMM approach, univariate statistics were used to determine whether specific donor sources promote a temporal convergence of physiological and biochemical traits toward values similar to those observed in reference control meadows. Given that the data did not meet the assumptions required for parametric tests, two-way permutational analyses of variance (PERMANOVA; Anderson, 2001) were performed. The PERMANOVA design included the fixed factor 'Donor source' (three levels: intermatte cutting, storm-fragment, and control meadow), and 'Months post-transplanting' (six levels: 3, 12, 15, 24, 27, and 36 months. All main effects and interactions among these factors were tested. Prior to analysis, a resemblance matrix based on Euclidean distances was constructed using untransformed data. The influence of each factor on the response variables was assessed through permutation tests on the residuals of a reduced model, using Type III partial sums of squares. A total of 999 permutations were used, and Monte Carlo p-values were calculated when the number of unique permutations was less than 100 (Anderson et al., 2008).

2.6.2 Multivariate

Non-metric multidimensional scaling (nMDS) based on a Bray–Curtis dissimilarity matrix was used to visualize temporal changes in the combined physiological and biochemical traits of *P. oceanica* transplants and control meadows. The stress value of each ordination was used as a measure of the reliability of the two-dimensional representation. A PERMANOVA was performed on the Bray–Curtis dissimilarity matrix to test for the effects of donor source, months post-transplantation, and their interaction on the multivariate trait structure. Pairwise post-hoc tests were conducted when significant main effects were detected. Finally, a similarity percentage (SIMPER) analysis was used to identify the physiological and biochemical traits that contributed most to the observed dissimilarities among donor sources and between donor sources and control meadows at each time point. nMDS and SIMPER analysis were performed using Rstudio software.

All PERMANOVA analyses were carried out using PRIMER-E with PERMANOVA+ software (version 7.0.24; PRIMER-E, Auckland, New Zealand). Statistical significance was set at $p < 0.05$, and all reported values are presented as mean ± standard error.

3. Results

3.1 Effects of transplantation method, donor source and transplantation depth on the physiological and biochemical traits of P. oceanica transplants.

GLMMs were used to test the effects of transplantation method, donor source, and transplantation depth on the physiological and biochemical traits of *P. oceanica* transplants. Most traits (Fv/Fm, alpha, carbon content, sulfur content, total carbohydrate reserves, sucrose, and starch) were not significantly affected by any factor or their interactions. Among photosynthetic parameters, rETRmax was significantly influenced by donor source ($p = 0.002$; $F = 8.3346$), with intermatte cuttings showing significantly higher values than storm-fragments. Ek was significantly affected by the interaction between donor source and transplantation depth ($p = 0.011$; $F= 4.4767$). Post-hoc tests showed that, at shallow sites, intermatte cutting had higher Ek values than storm-fragments ($p = 0.001$), while no difference was observed at deep sites. For leaf elemental concentrations, both nitrogen and phosphorus contents were significantly influenced by donor source ($p < 0.001$; $F = 23.443$ for nitrogen; $p = 0.007$; $F = 6.5313$ for phosphorus). Intermatte cuttings showed significantly higher nitrogen and phosphorus contents compared to storm-fragments. The C/N ratio was significantly affected by both donor source ($p < 0.001$; $F = 24.2719$) and transplantation method ($p = 0.003$; $F = 5.0774$). Storm-fragments showed higher C/N ratios compared to intermatte cuttings. Post-hoc comparisons for transplantation method revealed that BESE elements had significantly higher C/N ratios than coconut fiber mats ($p = 0.012$) and iron staples ($p = 0.009$), while no significant difference was found between the latter two transplantation methods. Finally, the C/P and N/P ratios were significantly influenced by the interaction between transplantation method and transplantation depth ($p = 0.011$; $F = 3.9004$ for C/P; $p = 0.013$; $F = 3.4048$ for N/P). At shallow sites, BESE elements showed significantly higher C/P ratios than coconut fiber mats ($p < 0.001$) and iron staples ($p = 0.014$). For the N/P ratio, BESE elements had significantly higher values compared to coconut fiber mats ($p = 0.016$) , but not compared to iron staples. At deep sites, no significant differences in C/P or N/P ratios were detected among transplantation methods.

3.2 Temporal dynamics of individual physiological and biochemical traits in P. oceanica transplants and control meadows

3.2.1 Photosynthetic activity

Donor source, months post-transplantation, and their interaction significantly influenced photosynthetic parameters, with the exception of donor source for α (Table S3.30). For Fv/Fm, intermatte cuttings initially (0 month) had lower values than storm-fragments and the controls. This difference progressively disappeared, although control meadows generally maintained higher values than transplants throughout the study (Figure 3.42A; Table S3.31). No consistent patterns were found for α, rETRmax, and Ek with respect to the interaction between donor source and time (Figure 3.42B, C, D; Table S3.31). Differences between controls and transplants were sometimes observed (e.g., 12, 24 months) but did not persist, and by 36 months, no significant differences remained (Figure 3.42B, C, D; Table S3.31). At transplantation (0 month) intermatte cuttings displayed distinct behaviour. They had higher rETRmax and Ek values than both storm-fragments and controls, and α values higher than controls (Figure 3.42B–D; Table S3.31). These initial differences diminished over time, converging with the other groups.

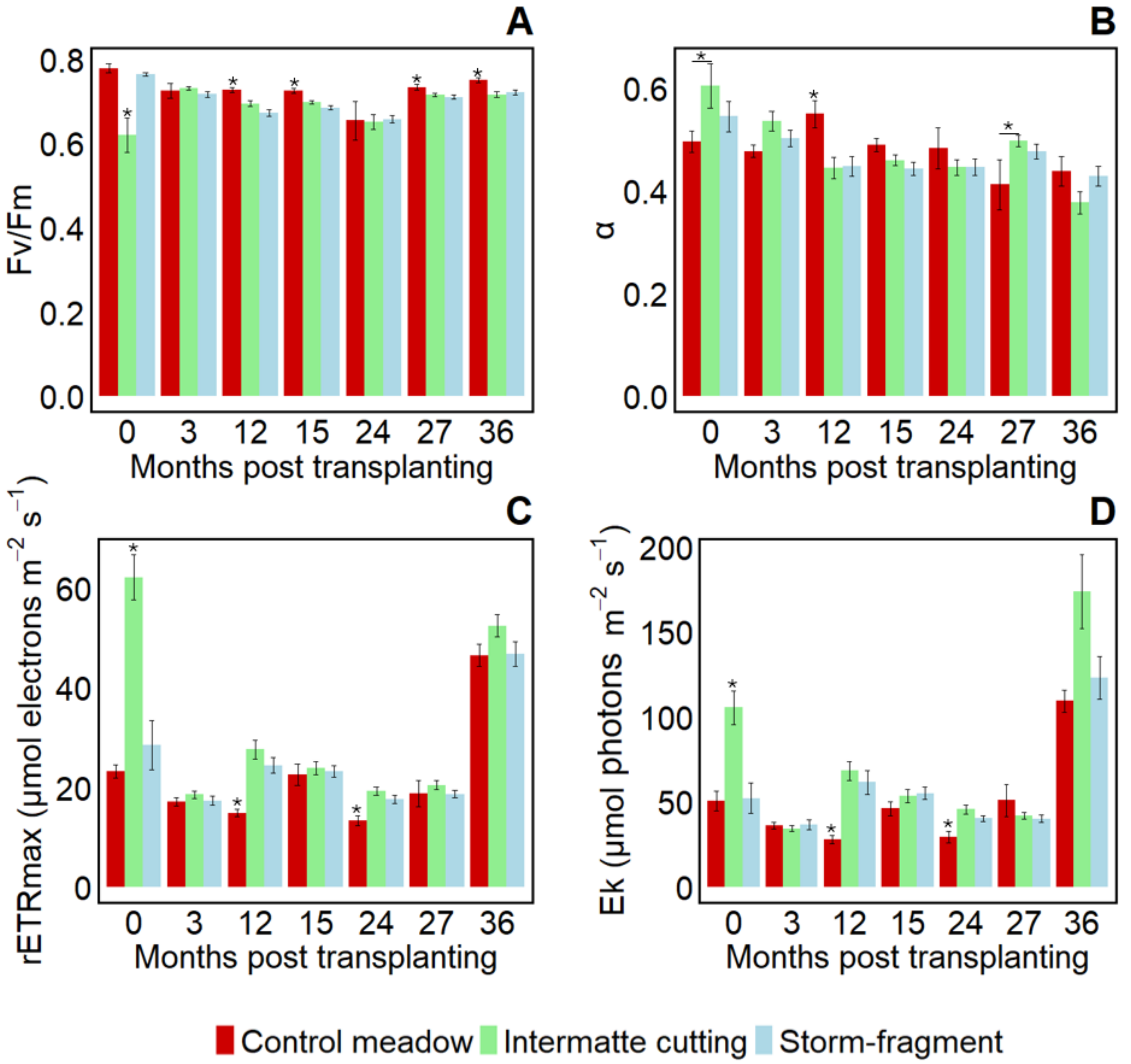


Figure 3.42. *Temporal dynamics of photosynthetic parameters in P. oceanica transplants (storm-fragments and intermatte cuttings) compared to control meadow : (A) maximum photochemical quantum yield (Fv/Fm), (B) photosynthetic efficiency (α), (C) maximum relative electron transport rate (rETRmax), and (D) saturating light intensity (Ek). Error bars indicate standard error. Significant differences ($p < 0.05$) are marked with an asterisk (*).*

3.2.2 Leaves' elemental concentrations

The elemental concentrations measured in the leaves of both transplant types and control meadows showed no consistent overall patterns (Figure 3.43). Carbon (C) content was significantly affected by donor source, months post-transplantation, and their interaction (Figure 3.43A; Table S3.30). Significant differences were observed between the control meadow and the transplants for up to two years, but diminished thereafter (Figure 3.43A; Table S3.31). No significant differences in C content were detected between storm fragments and intermatte cuttings after transplantation (i.e., beyond the initial measurements at 0 months following post-transplanting) (Figure 3.43A; Table S3.31). Nitrogen (N) content was also significantly influenced by donor source, months post-transplantation, and their interaction (Figure 3.43B; Table S3.30). From 12 to 24 months, storm fragments exhibited significantly lower N content than both control meadows and intermatte cuttings (Figure 3.43B; Table S3.31). At 27 months, storm-fragments and intermatte cuttings did not differ, although N content in storm-fragments remained significantly lower than in control meadows. By 36 months, control meadows still displayed significantly higher N content than both transplant types (Figure 3.43B; Table S3.31). Phosphorus (P) content was significantly affected by donor source and months post-transplantation, but not their interaction (Figure 3.43C; Table S3.31). Sulfur (S) content was not significantly affected by donor source but was significantly influenced by months post-transplantation, and its interaction with donor source (Figure 3.43D; Table S3.30). Marked temporal variability was observed throughout the study period (Figure 3.43D).

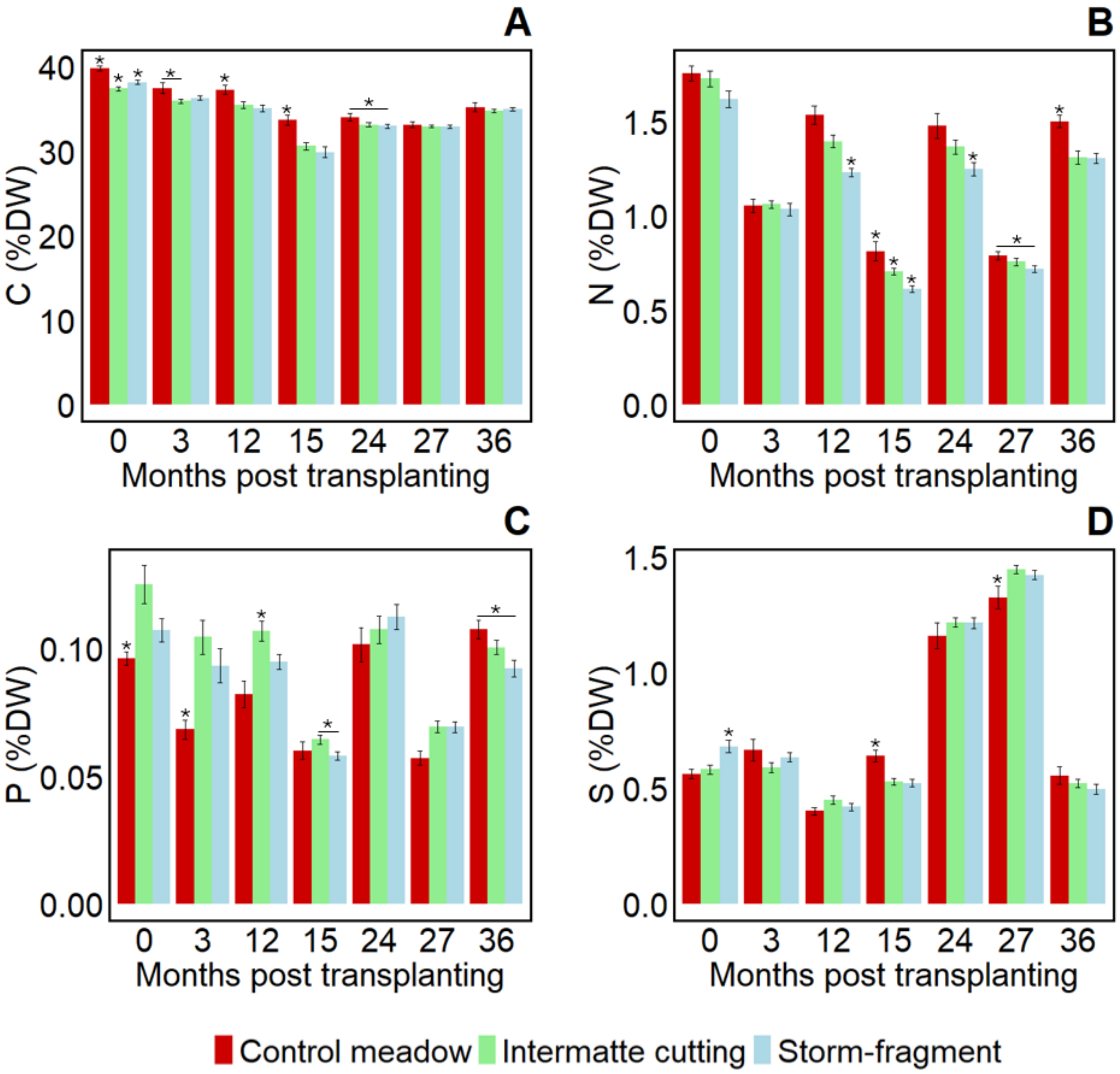


Figure 3.43. *Temporal dynamics of leaves' elemental content in P. oceanica transplants (storm-fragments and intermatte cuttings) compared to control meadow: (A) carbon content, (B), nitrogen content, (C), phosphorus content, and (D) sulfur content. Error bars indicate standard error. Significant differences ($p < 0.05$) are marked with an asterisk (*).*

The three elemental ratios C:N, C:P, and N:P were significantly affected by donor source and months post transplantation but not by their interaction (Figure 3.44; Table S3.30). C:N ratios were consistently higher in storm-fragments compared to both control meadows and intermatte cuttings (Figure 3.44A; Table S3.31). C:P ratios were highest in control meadows, followed by storm-fragments and then intermatte cuttings (Figure 3.44B; Table S3.31). Finally, N:P ratios were significantly higher in control meadows than in either transplant type (Figure 3.44C; Table S3.31).

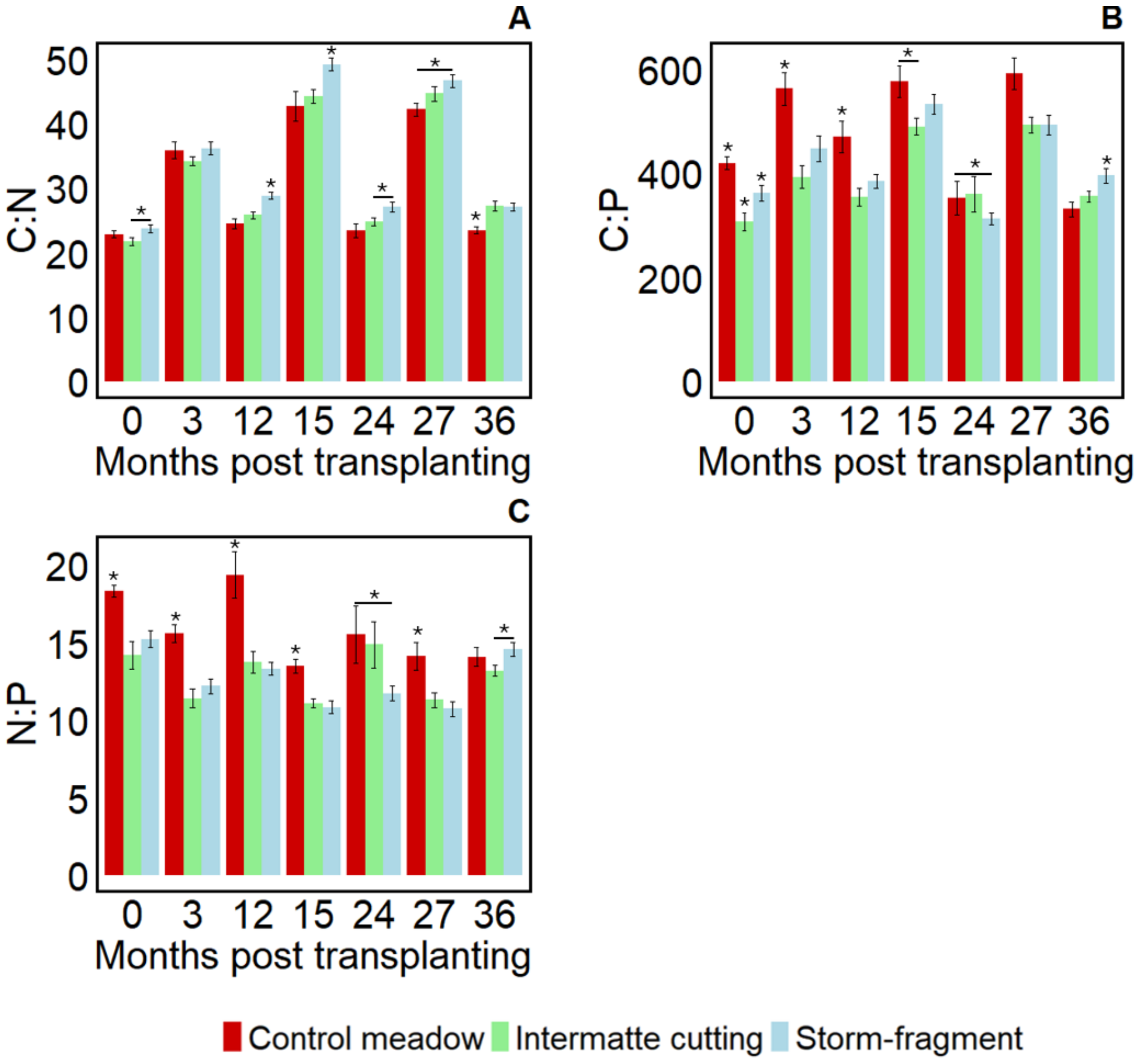


Figure 3.44. *Temporal dynamics of leaves' elemental ratios in P. oceanica transplants (storm-fragments and intermatte cuttings) compared to control meadow: (A) C/N, (B) C/P, and (C) N/P. Error bars indicate standard error. Significant differences (p < 0.05) are marked with an asterisk (*).*

3.2.3 Carbohydrate storage

Total carbohydrate reserves were significantly influenced by donor source, months post-transplantation, and their interaction (Figure 3.45A; Table S3.30). Intermatte cuttings initially showed significantly higher carbohydrate content than both control meadows and storm-fragments, but this difference diminished over time (Figure 3.45A; Table S3.31). At 12 and 24 months, no significant differences were detected between donor sources, whereas by 36 months, control meadows had significantly higher carbohydrate reserves than both transplants types (Figure 3.45A; Table S3.31).

Sucrose content was significantly affected by donor source and months post-transplantation, but not by their interaction (Table S3.30). Notable inter-annual variability was observed, including a marked decrease at 24 months in both control meadows and transplants (Figure 3.45B). At this time point, sucrose content was significantly higher in control meadows than in storm-fragments, and by 36 months, control meadows exceeded both transplant types (Figure 3.45B; Table S3.31).

Starch content was significantly influenced only by the donor source (Table S3.30). Its temporal dynamics (Figure 3.45C) closely mirrored those of total carbohydrate content (Figure 3.45A). Intermatte cuttings initially contained significantly more starch than control meadow and storm-fragments, but this difference disappeared over time (Figure 3.45C; Table S3.31). As with total carbohydrates, no differences were observed between donor sources at 12 and 24 months, while control meadows showed significantly higher starch content than transplants at 36 months (Figure 3.45C; Table S3.30).

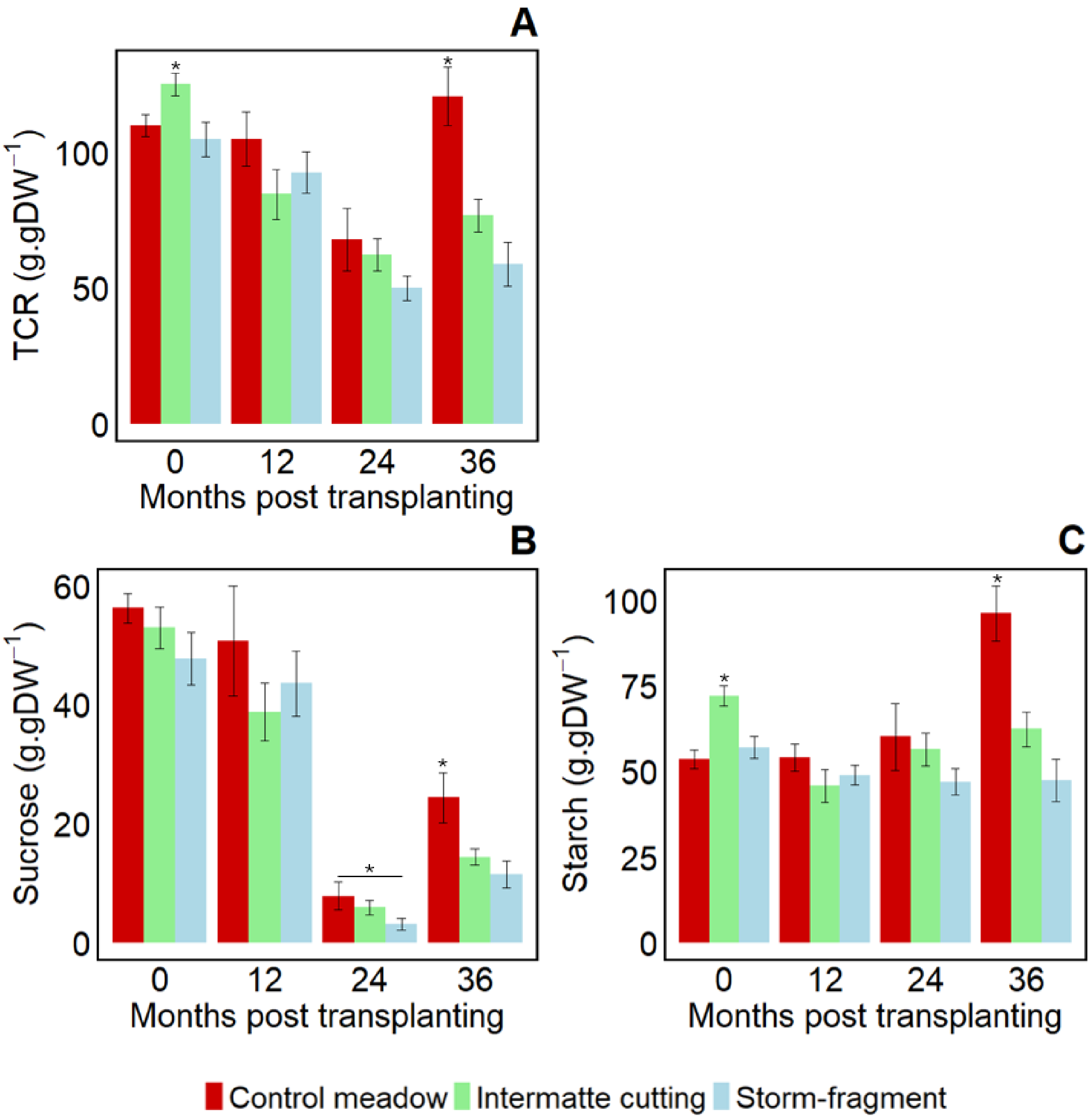


Figure 3.45. *Temporal dynamics of (A) total carbohydrate reserve (TCR), (B) sucrose, and (C) starch content in P. oceanica transplants (storm-fragments and intermatte cuttings) compared to control meadow. Error bars indicate standard error. Significant differences ($p < 0.05$) are marked with an asterisk (*).*

3.3 Temporal dynamics of combined physiological and biochemical traits in P. oceanica transplants and control meadows

The nMDS ordinations revealed strong dissimilarities among control meadows, storm-fragments, and intermatte cuttings prior transplantation, with the greatest separation observed between control meadows and intermatte cuttings (Figure 3.46). PERMANOVA confirmed that donor source, months post-transplantation, and their interaction significantly influenced the multivariate structure of physiological and biochemical traits throughout the study period. Post-hoc tests supported the nMDS results, showing significant differences among all three groups before transplantation (Table S3.32). At 12 months, dissimilarity between storm-fragments and intermatte cuttings largely disappeared, although both donor sources remained distinct from control meadows (Figure 3.46; Table S3.33). Across 0 and 12 months, six traits consistently explained more than 90% of group dissimilarities: C:P, TCR, starch, sucrose, Ek, and rETRmax (Table S3.34). By 24 months, intermatte cuttings and storm-fragments showed high similarity and no longer formed isolated clusters, while differences with control meadows had further decreased (Figure 3.46). Post-hoc tests detected no significant differences between intermatte cuttings and control meadows, though storm-fragments remained distinct (Table S3.33). SIMPER analysis identified C:P, TCR, starch, and Ek as the primary contributors (>85%) to dissimilarities, with C:N and N:P distinguishing storm-fragments and intermatte cuttings, and sucrose and N:P differentiating both donor sources from control meadows (Table S3.34). By 36 months, control meadows again formed a distinct cluster, while intermatte cuttings and storm-fragments overlapped partially but remained somewhat separated (Figure 3.46). Post-hoc tests confirmed significant differences among all three groups (Table S3.33). As at 0 and 12 months, the same six traits (C:P, TCR, starch, sucrose, Ek, rETRmax) explained more than 90% of observed dissimilarities (Table S3.34).

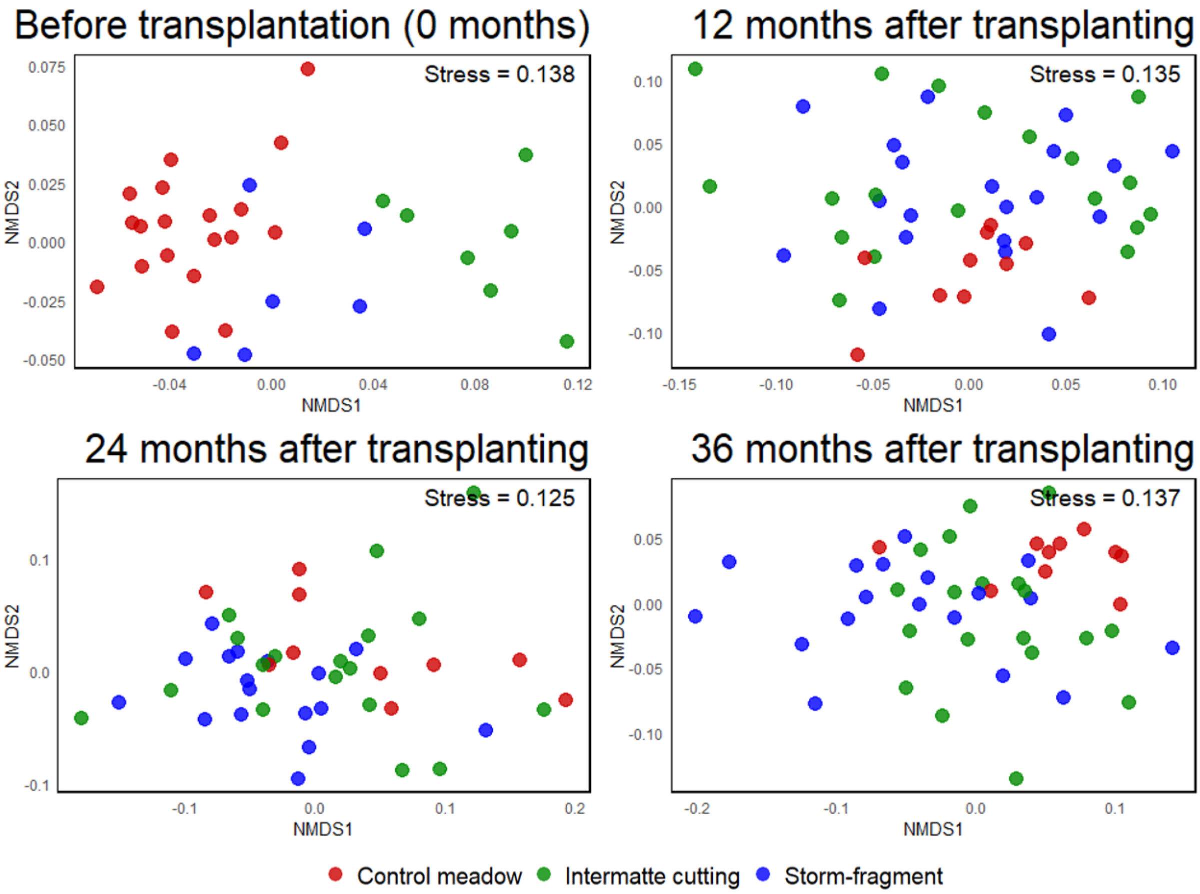


Figure 3.46. *Non metric multidimensional scaling (nMDS) ordination plots illustrating time series of multivariate physiological and biochemical traits' shifts as a function of donor source from pre-transplantation (0 months) to post-transplantation (12, 24 and 36 months).*

4. Discussion

Assessing the success of *P. oceanica* restoration efforts remains challenging due to the slow growth and delayed structural responses of this foundational seagrass species. Traditional metrics such as shoot density or coverage often fail to capture early signs of recovery, especially over the short timescales of most restoration projects (Cooke & Suski, 2008; Horn et al., 2009; Pansini et al., 2022). In this context, physiological and biochemical indicators offer a valuable alternative, as they can respond more rapidly and specifically to environmental conditions (Roca et al., 2016). Their integration into monitoring frameworks is therefore essential to better understand transplant performance and identify the underlying drivers influencing restoration outcomes (Pansini et al., 2022; Roca et al., 2016). This study aimed to evaluate the success of a pilot *P. oceanica* transplantation effort in Calvi Bay (Corsica) by combining physiological (photosynthetic activity) and biochemical indicators (elemental nutrient content and carbohydrate storage) to assess transplant performance. Over a three-year monitoring period, the effects of transplantation method, donor source, and transplantation depth on the physiological and biochemical traits of *P. oceanica* transplants were evaluated. The study specifically aimed to determine whether certain donor sources were more likely to develop trait profiles that progressively converged with those of natural meadows, thereby offering greater potential for long-term restoration success.

4.1 Influence of transplantation methods on P. oceanica transplants' physiological and biochemical traits

The three transplantation methods tested in this study (i.e. iron staples, coconut fiber mats, and BESE elements) had a very limited overall influence on the physiological and biochemical traits of *P. oceanica* transplants. Among the few significant differences, the C:N ratio in leaf tissues was notably affected by the transplantation method, with higher values observed in transplants using BESE elements compared to those using coconut fiber mats or iron staples. The C:N ratio is a complex indicator, as it can respond to both changes in nutrient availability and light limitation (Roca et al., 2016). However, since planting densities were standardized across all transplantation methods, the hypothesis of self-shading effects can be ruled out. Although no significant differences were observed in C or N content among transplantation methods, higher C:N ratios in BESE elements suggests reduced nitrogen availability or uptake, possibly due to limited root development (Boulenger et al., in review; de Boer, 2007; Lepoint et al., 2004; Udy & Dennison, 1997). Microenvironmental constraints associated with BESE elements' biodegradation (Nitsch et al., 2021), may also underlie this pattern; the release of dissolved organic carbon (DOC) from BESE elements (Nitsch et al., 2021) could further influence microbial or redox dynamics in the underlying sediment (Tu et al., 2025), indirectly affecting nitrogen cycling (Pedersen et al., 1999). However, the absence of significant differences in N content between transplantation methods does not suggest a pronounced nutrient limitation.

Furthermore, no significant differences were observed in elemental nutrient contents (C, N, S, P) or rhizomes' carbohydrate reserves (TCR, sucrose, starch) among the three transplantation methods, despite the markedly reduced root development in cuttings transplanted using BESE elements and coconut fiber mats compared to those fixed with iron staples three years post-transplantation (Boulenger et al., in review). This unexpected result may reflect the existence of distinct resource-use strategies among transplantation methods. Cuttings anchored with iron staples had direct access to the sediment (Boulenger et al., 2025c, in review), facilitating root development and enabling efficient nutrient uptake from interstitial porewater (Lepoint et al., 2002, 2004). This enhanced nutrient availability likely supported root growth, creating a positive feedback loop that reinforced both belowground development and nutrient assimilation. In contrast, cuttings transplanted using coconut fiber mats or BESE elements experienced limited or delayed contact with the sediment, likely restricting root system development and access to porewater-derived nutrients (Boulenger et al., 2025c, in review). As a result, while iron staple transplants may rely on active nutrient uptake for root system development, those on BESE elements and coconut fiber mats may adopt a more conservative survival strategy, characterized by reduced root development and a tighter regulation of internal resource use. Furthermore, Kraemer et al. (1997) hypothesized that the activity of leaf glutamine synthetase, the key enzyme responsible for converting inorganic nitrogen into organic forms, may be upregulated as a compensatory metabolic adjustment when root system is reduced. This enzymatic response reflects a form of metabolic plasticity that enables plants to maintain nitrogen assimilation and support the *de novo* synthesis of nitrogen-containing organic compounds, even in the absence of an efficient root system (Kraemer et al., 1997). Such contrasting strategies, root-supported nutrient uptake versus foliar metabolic compensation, may help explain why nutrient contents and carbohydrate reserves remained comparable across the three transplantation methods, despite underlying differences in root system development.

4.2 *Influence of transplantation depth on P. oceanica transplants' physiological and biochemical traits*

When *P. oceanica* cuttings are used for restoration purposes, accounting for light intensity is critical to avoid excessive mortality caused by levels falling outside the species' tolerance range (Stipcich et al., 2023). In the present study, as with transplantation method, the two transplantation depths tested (20 m vs. 28 m) had only a limited influence on the physiological and biochemical traits of *P. oceanica* transplants. It is important to note that most cuttings were transplanted to sites deeper than their original location. In particular, intermatte cuttings were harvested at 15 m depth, while storm-derived fragments originated from a broader depth range, spanning 6 m to 28 m. Previous studies have shown that transplanting cuttings deeper than their original depth can compromise survival, photosynthetic performance, and carbohydrate storage (Genot et al., 1994; Molenaar & Meinesz, 1992). However, the results of this study align with more recent studies suggesting that *P. oceanica* may acclimate to different depth-related light environments through physiological buffering and morphological plasticity (Boulenger et al., 2024; Dattolo et al., 2017; Ismael et al., 2023; Ruiz & Romero, 2003; Stipcich et al., 2023). Interestingly, studies reporting reduced survival and physiological performance typically used orthotropic rhizomes (Genot et al., 1994; Molenaar & Meinesz, 1992), while those observing minimal or no impact of transplantation depth relied on plagiotropic rhizomes (Boulenger et al., 2024; Dattolo et al., 2017; Mancini et al., 2022; Stipcich et al., 2023).

The limited physiological and biochemical traits' responses may result from insufficient irradiance contrast between shallow (20 m) and deep (28 m) sites to exceed a critical acclimation threshold (Ruiz & Romero, 2003). Ismael et al. (2023) showed that *P. oceanica* can maintain photosynthetic activity and carbon allocation even under low-light conditions in deep waters, partly through starch mobilization in rhizomes and possibly enhanced amylase activity. In contrast, shallow-water plants may produce more carbohydrates due to higher irradiance but also face greater stress (e.g., epiphytes, oxidative bursts), leading to increased carbohydrate consumption (Costa et al., 2015; Sureda et al., 2008). These compensatory mechanisms could explain the lack of significant differences in carbohydrate reserves between the two transplantation depths. Interestingly, while depth alone did not significantly alter photosynthetic activity parameters, significantly higher Ek (i.e. saturating irradiance) values were observed in intermatte cuttings compared to storm-fragments, but only at shallow sites. This result suggests that the light history of donor material plays a key role in shaping physiological responses after transplantation. Intermatte cuttings, originally located on the eroding edges of natural sandy intermattes (Gobert et al., 2016), are typically exposed to higher light levels than storm-fragments, which often lie directly on the sediment surface and may be partially shaded by *P. oceanica* litter. The significantly higher Ek values observed in intermatte cuttings suggest that they retained a greater capacity for light utilization in shallow conditions. In contrast, storm fragments appeared less acclimated to elevated irradiance, likely due to their more shaded origin, which resulted in lower Ek values. However, these differences in Ek did not translate into significant variations in other photosynthetic activity parameters, carbon content or carbohydrate reserves. Finally, these results indicate that, while transplanting cuttings at similar depths may facilitate acclimation through pre-existing physiological adaptations, this condition is not essential, especially when using plagiotropic rhizomes and when donor and transplantation sites share similar environmental conditions within the same coastal area.

4.3 Influence of donor origin on P. oceanica transplants' physiological and biochemical traits

P. oceanica restoration projects have typically used rhizome fragments from two main donor sources: either cuttings harvested directly from natural *P. oceanica* meadows (e.g., Bacci et al., 2024; Calvo et al., 2021; Pirrotta et al., 2015), or naturally detached storm-fragments (e.g., Castejón-Silvo & Terrados, 2021; Mancini et al., 2021; Piazzi et al., 2021). This study aimed to experimentally compare the physiological and biochemical traits of *P. oceanica* transplants derived from these two donor sources. Such comparisons are essential, as restored populations may develop altered physiological traits, the ecological consequences of which remain largely uncertain (Cooke & Suski, 2008). By identifying trait-specific differences related to donor origin, this study contributes to a better understanding of the biological performance and restoration potential of each donor source. Among these traits, rETRmax, N, and P contents were significantly higher in intermatte cuttings compared to storm-fragments. The C:N ratio was also significantly influenced by donor origin, with higher values observed in storm-fragments than in intermatte cuttings.

Temporal dynamics of these traits revealed that rETRmax differed significantly between intermatte cuttings and storm-fragments only prior to transplantation. This difference is likely attributable to varying light exposures in their original environments (Dattolo et al., 2014; Horn et al., 2009; Major et al., 2002), as discussed in the preceding section. Nitrogen content exhibited longer-lasting effects, with significantly higher N contents in intermatte cuttings compared to storm-fragments at 12, 15, and 24 months post-transplantation. Symbiotic N_2-fixing microorganisms play a critical role in nitrogen assimilation by eukaryotes in nitrogen-limited environments (Lilburn et al., 2001; Pool et al., 2018; Welsh et al., 2002). At 24 months post transplanting, the same samples in the study of Boulenger et al. (2025c) showed that the roots of intermatte cuttings had a notably higher abundance of the bacterial order *Chromatiales*, particularly the genus *Candidatus* Thiodiazotropha, than those of storm-fragments. *Candidatus* Thiodiazotropha has been identified as a key endosymbiont in the coastal cordgrass *Spartina alterniflora* (Rolando et al., 2024). Given the pivotal role of this genus in sulfur oxidation and nitrogen fixation processes (Martin et al., 2020; Rolando et al., 2024), further research is warranted to determine whether the higher abundance of *Candidatus* Thiodiazotropha in intermatte cuttings could contribute to improved plant performance (Boulenger et al., 2025c), for example through increased nitrogen content in transplanted seagrass tissues (Mohr et al., 2021; Zhou et al., 2024). Higher nitrogen content in the intermatte cuttings explains why the C:N ratio was significantly higher in the storm-fragments.

4.4 Temporal convergence and divergence of physiological and biochemical traits between natural meadows and transplants

Survival of transplanted *P. oceanica* cuttings can be influenced by a number of different factors: organic matter content of the sediment (Boulenger et al., 2025a; Cancemi et al., 2003), surrounding algal community (Pereda-Briones et al., 2018), nutrients' uptake (Lepoint et al., 2004; Vangeluwe et al., 2004), carbohydrate reserves (Genot et al., 1994). Assessing the nutrient content of transplanted cuttings in comparison with that of shoots from the surrounding natural meadows can provide insight into whether nutrient uptake in the transplants is sufficient to meet these requirements (Castejón-Silvo & Terrados, 2021). The three-year monitoring of nutrient contents in the leaves of natural *P. oceanica* meadows highlights a natural seasonal dynamic (Gobert et al., 2005; Lepoint et al., 2002). Previous transplantation experiments with *P. oceanica* cuttings in the Bay of Calvi have shown that cuttings are unable to meet their nutrient requirements for growth, exhibiting lower phosphorus (Gobert et al., 2005; Vangeluwe et al., 2004. Vangeluwe, 2006) and nitrogen contents (Gobert et al., 2005; Lepoint et al., 2004; Vangeluwe et al., 2004; Vangeluwe, 2006) in their shoots compared with those from natural meadows. Regarding phosphorus, substantial temporal variability has been observed, with P contents generally higher in transplants than in natural meadows, as noted by Castejón-Silvo & Terrados (2021). However, 36 months after transplantation, natural meadows display higher P content than transplants, in agreement with the results reported by Vangeluwe (2006).

Nitrogen acquisition by *P. oceanica* transplants has long been considered a critical factor for the long-term success of seagrass restoration projects (Lepoint et al., 2004; Pansini et al., 2024; Pergent-Martini et al., 2024). Previous studies have reported highly contrasting results, making it difficult to generalize this process in transplanted cuttings: pronounced temporal variability without a clear pattern (Pansini et al., 2024), higher N contents in transplants than in natural meadows (Castejón-Silvo & Terrados, 2021), and the opposite outcome, with higher concentrations in natural meadows than in transplants (Gobert et al., 2005; Lepoint et al., 2004; Vangeluwe et al., 2004; Vangeluwe, 2006). The higher nitrogen content in transplants compared to control meadows observed by Castejón-Silvo and Terrados (2021) may be explained by the storage of transplants in mesocosms for several months prior to transplantation. Indeed, their results show that the transplants had significantly higher nitrogen concentrations when maintained in mesocosms than before their storage. Moreover, the water circulating in their tanks exhibited strong variations in nitrate concentrations (0.66 ± 0.25 µM – 4.20 ± 0.38 µM), with very high values recorded in summer (4.20 ± 0.38 µM) (Castejón-Silvo & Terrados, 2021). In contrast, the water column in the Bay of Calvi is much more oligotrophic, with monthly mean nitrate concentrations never exceeding 1 µM and with higher values in winter than in summer (Fullgrabe et al., 2020; Lepoint et al., 2002).

The results of the present study are consistent with earlier work conducted in the Bay of Calvi (Gobert et al., 2005; Lepoint et al., 2004; Vangeluwe et al., 2004; Vangeluwe, 2006), showing overall higher nitrogen concentrations in natural meadows compared to transplants, with these differences persisting for up to three years after transplantation. The hypothesis proposed by Lepoint et al. (2004) suggests that transplants prioritize the development of the root system by reallocating part of the nitrogen stored in both leaves and rhizomes. Interestingly, in our study, foliar nitrogen content was similar across the three transplantation methods, whereas Boulenger et al. (in review) reported that cuttings placed on BESE elements and coconut fiber mats had developed almost no roots three years after transplantation. In contrast, cuttings attached with iron staples had developed a substantial root system over the same period, although still less developed than in natural meadows. Therefore, the similar foliar nitrogen content observed across transplantation methods may result from different nitrogen utilization strategies (Figure 3.47). In BESE elements and coconut fiber mats, the absence of roots may be linked to the lack of direct contact with the sediment (Boulenger et al., 2025c), which limits the release of root exudates into the sediment. Such exudates promote microbial colonization via chemotaxis and attract key microbial partners that enhance plant fitness within the seagrass rhizosphere (Boulenger et al., 2025c; Crump et al., 2018; Sogin et al., 2022; Zhang et al., 2024). Consequently, only foliar nutrient uptake and internal nitrogen recycling occur in these treatments (Figure 3.47) (Alcoverro et al., 2000; Lepoint et al., 2002; Vangeluwe et al., 2004), and nitrogen is not invested in root system construction. By contrast, cuttings attached with iron staples can also absorb nutrients from the sediment porewater through their roots (Lepoint et al., 2002). This sedimentary nitrogen may be directly allocated to continued root system development, which would explain why higher foliar nitrogen concentrations are not observed with this transplantation method (Figure 3.47). Finally, natural meadows exhibit higher foliar nitrogen content than transplants because their root systems are already fully developed. As a result, the retranslocation of nitrogen from leaves and rhizomes to the roots is no longer necessary (Figure 3.47) (Lepoint et al., 2004).

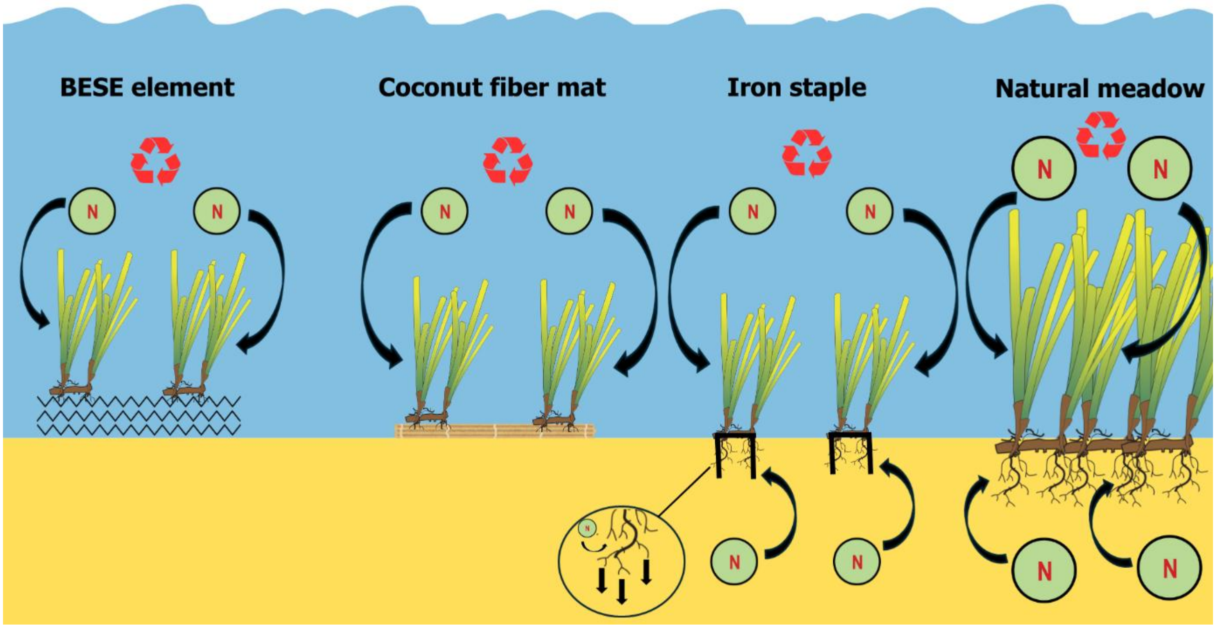


Figure 3.47. *Conceptual figure of nitrogen uptake and allocation strategies in P. oceanica cuttings under different transplantation methods.*

Another key parameter in assessing transplant health is the internal carbohydrate reserves, especially starch (Govers et al., 2015), stored in rhizomes as they can strongly influence biomass production (Alcoverro et al., 1995) and play a critical role in the overwintering capacity of seagrasses when photosynthetic activity is reduced (Alcoverro et al., 2001; Govers et al., 2015). Unlike natural meadows, transplanted cuttings are not physiologically integrated into an extensive rhizome network and therefore cannot translocate resources over long distances (Alcoverro et al., 2000; Marbà et al., 2002), which may limit their ability to buffer environmental stress and sustain growth (Castejón-Silvo & Terrados, 2021). Before transplantation, the higher TCR and starch content in intermatte cuttings compared to storm-fragments and control meadow can be explained by their greater light exposure on the eroding edges of intermattes (Genot et al., 1994; Gera et al., 2013). TCR and starch content remained relatively stable in both transplants and control meadows at 12 and 24 months after transplantation. However, a marked increase in starch content was observed in control meadows at 36 months, whereas transplants maintained similar values throughout the three-year monitoring period. The constant starch levels in transplants suggest that they are able to maintain and replenish their starch reserves, with no differences detected between donor origins.

The temporal analysis of multivariate trait structure revealed the recovery dynamics of physiological and biochemical traits in transplanted cuttings. Such indicators are known to be particularly effective in capturing recovery processes in large seagrass species (Roca et al., 2016). Nevertheless, because their responses are highly stress-specific, they should be interpreted in combination with other complementary indicators (Roca et al., 2016). Overall, the combined trait analysis indicated that transplants from both donor sources acclimated to their local environments, as evidenced by the convergence of their physiological and biochemical traits at 12 and 24 months post-transplantation. Over the same period, intermatte cuttings converged towards trait values comparable to those of natural meadows, whereas storm-fragment cuttings consistently displayed traits distinct from natural meadows. These results suggest that the intermatte cuttings show a faster convergence than the storm-fragments, potentially linked to their different life histories (Pergent-Martini et al., 2024). However, 36 months post-transplanting, divergences re-emerged between the two donor sources and between control meadows and transplants. These findings indicate that the time elapsed since intervention can strongly influence the assessment of restoration success (Pansini et al., 2024). Further long-term research is needed to determine when the cuttings reach a stable state, with complete convergence of traits between transplants and natural meadows, thereby reflecting the full recovery of physiological and biochemical traits in P*. oceanica* transplants.

5. Conclusion

This study demonstrates that the physiological and biochemical performance of *P. oceanica* transplants is influenced more by donor origin than by transplantation method or depth. While iron staples, coconut fiber mats, and BESE elements produced comparable nutrient contents and carbohydrate reserves, differences in root system development suggest distinct resource acquisition strategies. Depth-related light variation within the tested bathymetric range had minimal effects on transplant performance, indicating a capacity for physiological acclimation.

Donor origin emerged as a key driver of recovery trajectories, with intermatte cuttings exhibiting consistently higher nitrogen content and faster convergence towards natural meadow trait profiles than storm-fragments. These differences are likely linked to pre-transplant light history, nutrient assimilation capacity, and associated microbial partners. Despite early convergence of physiological and biochemical traits in both donor sources within the first two years, divergences reappeared by the third year, highlighting the importance of long-term monitoring to capture non-linear recovery patterns. Overall, this work emphasizes the importance of integrating physiological and biochemical indicators into restoration assessment frameworks, as they provide early, sensitive insights into transplant performance. Achieving full recovery in *P. oceanica* transplants is a long-term process, and future studies should aim to identify the time thresholds at which physiological and biochemical trait convergence with natural meadows becomes stable and sustained. Such knowledge will refine restoration strategies, optimize donor material selection, and improve the long-term success of seagrass restoration projects.

Chapitre IV


©STARESO/Arnaud Abadie

Discussion générale et perspectives

1. Vers une stratégie intégrée de restauration des herbiers de *P. oceanica*

En février 2024, le Parlement européen a adopté la *Loi sur la restauration de la nature*. Dans le cadre de cette législation, les États membres de l'Union européenne sont tenus de restaurer au moins 30 % des habitats terrestres et marins en mauvais état d'ici 2030, 60 % d'ici 2040 et 90 % d'ici 2050. Au cours des siècles passés, de nombreuses activités humaines ont largement contribué à la dégradation des habitats marins côtiers européens (Airoldi & Beck, 2007). Cette situation concerne particulièrement les vastes herbiers de *P. oceanica*, qui ont connu une régression bien documentée dans l'ensemble du bassin méditerranéen (Boudouresque et al., 2009 ; de los Santos et al., 2019 ; Telesca et al., 2015). Pour répondre aux objectifs de la nouvelle législation européenne, il est primordial de commencer par identifier les sites prioritaires à restaurer. Pour la restauration des herbiers de *P. oceanica*, l'identification de ces sites doit s'inscrire dans une stratégie de gestion intégrée à l'échelle régionale en tenant compte des conditions écologiques et socio-économiques locales et ne peut plus se limiter à des projets isolés de transplantation. De plus, il devient nécessaire d'intégrer aux démarches de planification d'actions de restauration, les projections climatiques futures ainsi que les évolutions socio-économiques (population, éducation, urbanisation, PIB). Les différents scénarios SSP ('*Shared Socio-economic Pathways*') décrivent ces trajectoires alternatives d'évolution des sociétés futures (IPCC, 2021). Parmi ces scénarios socio-économiques, le Groupe d'experts intergouvernemental sur l'évolution du climat (GIEC) en a sélectionné cinq principaux qui couvrent l'ensemble des développements futurs possibles des facteurs anthropiques responsables du changement climatique. Ainsi, on retrouve deux scénarios avec des émissions de gaz à effet de serre (GES) très élevées (SSP5-8.5) et élevées (SSP3-7.0), un scénario avec des émissions de GES intermédiaires (SSP2-4.5), et deux scénarios avec des émissions de GES faibles (SSP1-2.6) et très faibles (SSP1-1.9) (IPCC, 2023).

Cependant, les approches méthodologiques visant à identifier les sites prioritaires de restauration, en intégrant les dimensions socio-économiques et environnementales à l'échelle régionale, ont jusqu'à présent reçu relativement peu d'attention dans la littérature scientifique consacrée à la restauration marine, alors même qu'elles jouent un rôle déterminant pour garantir des interventions écologiquement pertinentes (Boudouresque et al., 2021; Fabbrizzi et al., 2023; Lester et al., 2020). Dans le cadre de cette thèse, des analyses préliminaires limitées à l'échelle de la Corse ont déjà été réalisées afin d'identifier les sites prioritaires pour la restauration des herbiers de *P. oceanica* (Bucher, 2025). En effet, les grandes îles méditerranéennes méritent une attention particulière, car la fixation de carbone par les herbiers de *P. oceanica* y est d'autant plus importante que la densité de population humaine est relativement faible et que ces territoires abritent de vastes surfaces d'herbiers. À titre d'exemple, la fixation annuelle de carbone par ces écosystèmes atteint près de 14,4 % pour la Corse (Pergent-Martini et al., 2021). La méthodologie utilisée pour arriver à cette sélection de sites prioritaires s'est basée sur une approche en deux phases. Dans un premier temps, les modèles de distribution d'espèces ('*Species Distribution Models*', SDMs) utilisant des données de distribution de *P. oceanica* actuelles et des variables environnementales ont permis d'estimer les zones favorables à *P. oceanica* dans les conditions climatiques présentes (2000-2020) et futures (2030-20250), selon deux scénarios contrastés (SSP1-1.9 et SSP5-8.5). Dans un second temps, les SDMs présents et futurs ont été combinés dans une analyse multicritère spatialisée ('*Multi-Criteria Decision Analysis*', MCDA) au sein d'un environnement SIG, afin d'identifier les sites propices à la restauration de *P. oceanica* autour de la Corse. Cette analyse a intégré les conditions géomorphologiques (bathymétrie, nature du substrat), les pressions anthropiques (ancrage, chalutage), ainsi que la présence d'aires de gestion (parc marin, réserves naturelles). La classification des sites selon différents degrés d'aptitude à la restauration (i.e., très favorable, modérément favorable, et peu favorable) a été établie au moyen du processus d'analyse hiérarchique ('*Analytical Hierarchy Process*', AHP), une méthode structurée d'aide à la décision multicritères permettant d'intégrer divers facteurs écologiques, environnementaux et socio-économiques en un score unique d'aptitude (Saaty, 2008). Dans cette étude, l'AHP a été utilisé pour définir les pondérations de l'ensemble des variables influençant le potentiel de restauration de *P. oceanica*. Les résultats montrent que 88 hectares sont hautement appropriés pour des actions de restauration, tandis que 1 256 hectares apparaissent comme modérément favorables (Figure 4.48) (Bucher, 2025). Ce type d'approche, combinant modèles de distribution d'espèces et analyse multicritère spatialisée, constitue un outil essentiel pour élaborer une stratégie adaptée aux besoins de gestion actuels et futurs. Les résultats qui en découlent représentent une base stratégique majeure pour orienter les futures actions de restauration des herbiers de *P. oceanica* en Corse.

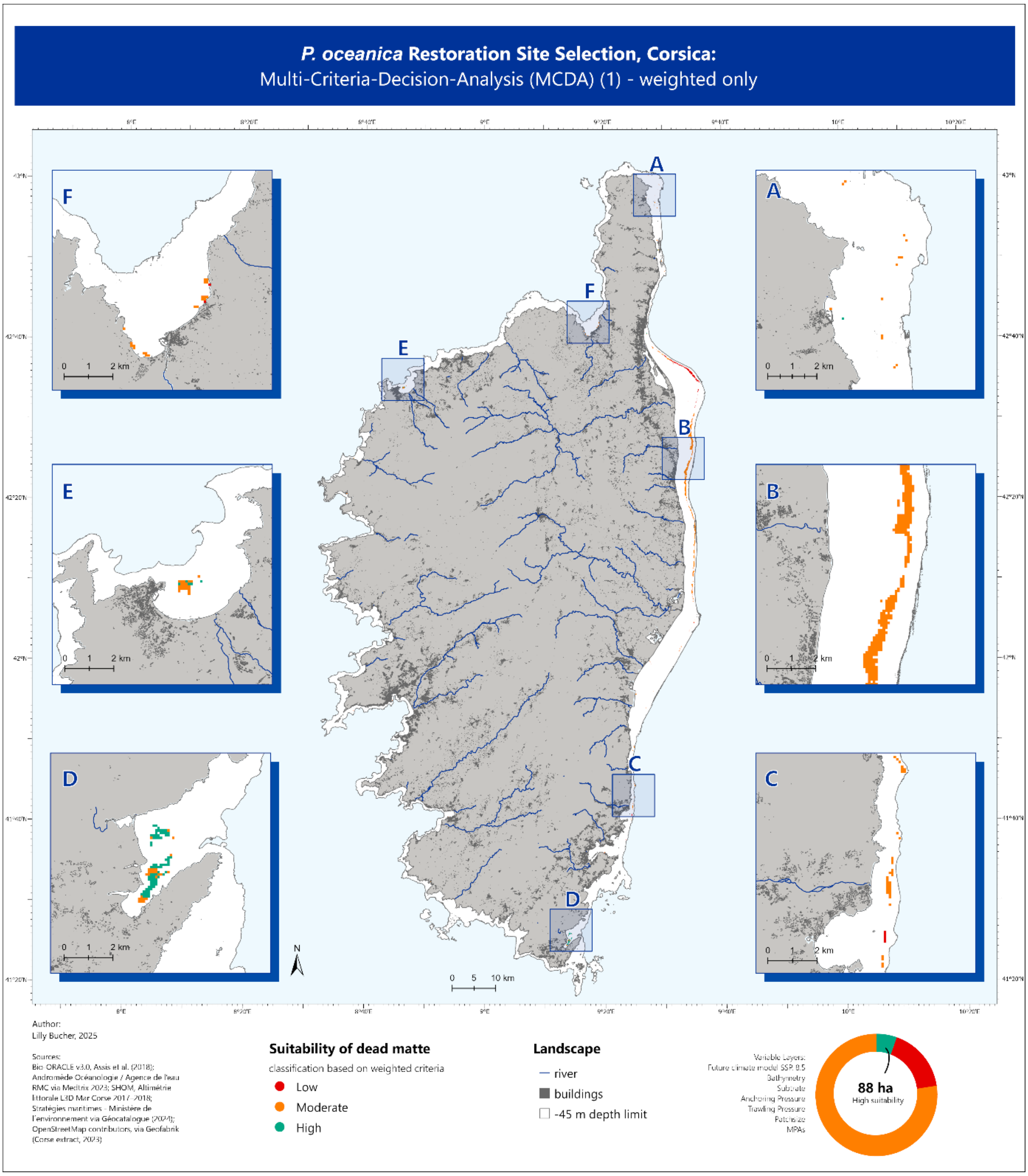


Figure 4.48. *Identification des sites prioritaires à la restauration des herbiers de P. oceanica en Corse.*

La restauration des herbiers de *P. oceanica* vise à stabiliser les zones dégradées afin d'enrayer leur érosion dans le temps, tout en renforçant la connectivité entre herbiers fragmentés afin de favoriser la résilience de l'ensemble de cet écosystème (Barcelona et al., 2021 ; Pergent-Martini et al., 2024). Ces résultats montrent la présence de surfaces non négligeables (88 hectares hautement favorables à la restauration en Corse) qui nécessiteraient la mise en œuvre d'actions de restauration actives ou passives (i.e., réduction des pressions), selon la dynamique locale de recolonisation naturelle. Celle-ci peut s'opérer par la croissance des rhizomes plagiotropes en bordure d'herbiers, par l'établissement de boutures-épaves (Boudouresque et al., 2021), ou encore par la dispersion et l'établissement de nouveaux patchs issus de graines germées (Balestri et al., 2017 ; Boudouresque et al., 2021).

La régression d'environ 416 400 hectares d'herbiers de *P. oceanica* à l'échelle de l'entièreté du bassin méditerranéen mise en évidence sur les cinquante dernières années (Telesca et al., 2015) appelle à la nécessité de la mise en place de projets de restauration alliés à des stratégies coordonnées de protection par les États membres européens méditerranéens dans les années à venir. Cependant, la plupart des actions de restauration d'herbiers de *P. oceanica* recensées jusqu'à aujourd'hui sont expérimentales et de petite échelle, mobilisant une grande diversité de techniques et de contextes d'intervention, ce qui reflète le caractère encore émergent de la restauration des herbiers de *P. oceanica* (Pansini et al., 2022). Jusqu'à présent, les quelques rares projets de restauration d'herbiers de *P. oceanica* menés à large échelle, ou dans un contexte industriel et non de recherche académique, étaient des mesures visant à compenser les destructions occasionnées par des projets de construction d'infrastructures maritimes (Bacci et al., 2024; Boudouresque et al., 2021; Descamp et al., 2025). Au vu de l'étendue des zones dégradées, le développement d'une véritable stratégie de restauration des herbiers marins apparaît ainsi comme une condition essentielle pour permettre la mise en place de projets de restauration d'herbiers de *P. oceanica* sur de vastes étendues en dehors du contexte de la recherche académique et des mesures compensatoires. Des exemples de restauration issus d'autres écosystèmes marins montrent qu'une telle stratégie est réalisable. Aux États-Unis, la restauration des récifs d'huîtres (*Crassostrea virginica* et *Ostrea lurida*) est devenue une priorité afin de restaurer les services écosystémiques apportés par cet habitat. La demande croissante pour la restauration des récifs d'huîtres a conduit à l'émergence d'une économie de leur restauration, évaluée entre 70 et 90 millions de dollars de dépenses annuelles, soutenant environ 1 500 emplois et générant 210 millions de dollars de retombées économiques (Hall & DeAngelis, 2024).

Pour les écosystèmes terrestres, le succès de la restauration est positivement corrélé au niveau d'investissement et les techniques sont désormais suffisamment abouties pour que de fortes dépenses soient directement récompensées (Ruiz-Jaen & Aide, 2005). Concernant les écosystèmes marins, et en particulier les herbiers marins, les techniques de restauration en sont encore à un stade bien moins avancé (Bayraktarov et al., 2015 ; Duarte et al., 2020 ; Ruiz-Jaen & Aide, 2005). Néanmoins, à mesure que la science de la restauration des habitats marins progresse, des investissements plus conséquents devraient permettre d'obtenir des gains relativement plus importants (Bayraktarov et al., 2016). Ainsi, un besoin d'investissement est nécessaire aussi bien dans la restauration à grande échelle que dans l'amélioration des pratiques de restauration (Bayraktarov et al., 2016).

Avant toute action de restauration d'un herbier marin, il est essentiel de caractériser les conditions abiotiques du site à restaurer (Laegdsgaard, 2006 ; Bayraktarov et al., 2016). Le succès des méthodes varie selon les sites, principalement en raison de facteurs environnementaux (e.g., hydrodynamisme, granulométrie, nutriments dans le sédiment) dont l'importance peut changer à petite échelle spatiale (Unsworth et al., 2024). De précédents travaux avaient montré que la matte morte résultant de l'action de l'ancrage présentait des teneurs élevées en H_2S, ce qui pouvait induire une difficulté de recolonisation naturelle de cet habitat (Abadie et al., 2016) à cause de la toxicité de ce composé chimique pour *P. oceanica* (Calleja et al., 2007 ; Holmer et al., 2003 ; Marbà et al., 2006). Les résultats de ces travaux de thèse sont venus compléter cette première caractérisation et ont également démontré que la matte morte présente une granulométrie similaire à l'herbier naturel avec un contenu en matière organique réduit, ainsi qu'une colonisation importante par des macroalgues comme *C. cylindracea* et *C. prolifera* (Chapitre 2.1). De plus, des différences importantes dans la structure des communautés bactériennes entre la matte morte et le sédiment sous l'herbier de *P. oceanica* ont également été démontrées (Chapitre 3.1). Enfin, les résultats de cette thèse ont également mis en évidence une dynamique d'érosion induite par un hydrodynamisme modifié suite à la perte de la canopée foliaire de l'herbier de *P. oceanica*, ces derniers résultats venant corroborer ceux d'Abadie et al. (2019). Les résultats de cette thèse viennent donc démontrer que la matte morte présente des caractéristiques sédimentaires drastiquement modifiées comparées à la matte sous un herbier de *P. oceanica*, ce qui explique la difficulté et la lenteur de la recolonisation observée dans ces travaux de thèse (Chapitre 2.1), ainsi que dans d'autres études antérieures (e.g., Abadie et al., 2016, 2019 ; Gonzalez-Correa et al., 2005 ; Kiparissis et al., 2011).

La profondeur des sites ressort également comme facteur influençant la dynamique de recolonisation avec des mécanismes différents. La recolonisation naturelle a plus faible profondeur présente un dualisme entre processus de recolonisation et d'érosion, mais bénéficie également d'un apport de boutures-épaves qui peuvent générer l'établissement de nouveaux patchs de *P. oceanica*. En revanche, la recolonisation naturelle dans les tranches bathymétriques plus profondes est moins soumise aux processus d'érosion (Bonamano et al., 2010; Vacchi et al., 2017; Uhrin & Turner, 2018) mais bénéficie d'un apport moindre de boutures-épaves. Ces résultats sont essentiels afin de sélectionner des méthodes de restauration adaptées aux spécificités environnementales locales. De plus, la lenteur de la recolonisation mais également la présence d'une dynamique progressive justifient la restauration afin d'accélérer les processus de recolonisation naturelle observés via la transplantation de nouveaux patchs d'herbiers qui, à terme, pourront fusionner entre eux et avec les patchs naturels.

L'étude des différentes méthodes de transplantation utilisées dans ces travaux de thèse avait pour but de déterminer si l'établissement sur le long-terme des boutures de *P. oceanica* est facilité par la rigidité/structure tri-dimensionnelle du matériel de transplantation, et quelle(s) méthode(s) de transplantation est la plus performante parmi les trois testées dans ces travaux de thèse. Ainsi, les différentes méthodes se sont avérées avoir des performances variables en fonction de la profondeur de transplantation, reflétant les différentes dynamiques de recolonisation naturelle (érosion/disponibilité en boutures-épaves) aux deux profondeurs étudiées ici (Chapitre 2.1). La transplantation à l'aide d'agrafes métalliques n'est pas recommandée dans des zones à hydrodynamisme important, car les transplants sont fréquemment décrochés lors des premières années de transplantation alors que le système racinaire n'est pas encore complètement fonctionnel. En revanche, cette méthode s'est révélée être la plus performante à faible hydrodynamisme (Figure 4.52) et témoigne du rapport cout-bénéfice moyen le plus avantageux (9,8 €/m²) en comparaison du géotextile en fibre de noix de coco (20,5 €/m²) et des éléments BESE (62,25 €/m²). Le géotextile en fibre de noix de coco est une méthode présentant des taux de survie et un rapport cout-bénéfice peu favorables comparé aux deux autres méthodes, et n'est donc pas recommandée pour des projets de transplantation à plus grande échelle dans des conditions environnementales similaires à ces travaux. La structure formée par les éléments BESE, imitant la matrice formée par l'enchevêtrement des racines et des rhizomes, présente des taux de survie des transplants similaires dans les deux profondeurs de transplantation. Ceci suggère que, malgré le coût important de ce matériel, cela pourrait être une méthode à privilégier dans des zones à hydrodynamisme important (Figure 4.52). De plus, cette méthode pourrait également servir dans le cas de matte morte fortement dégradée physiquement ou sur des substrats à prédominance sableuse ou graveleux (Figure 4.52). Ces résultats démontrent ainsi qu'il n'existe pas une seule technique fonctionnelle, mais qu'il est nécessaire de développer un panel de méthodes, à l'efficacité démontrée, et adaptées aux objectifs du projet de restauration, aux limitations financières et aux conditions environnementales locales (Figure 4.52).

2. Fonctionnement écologique et performances des méthodes de transplantation des herbiers de *P. oceanica*

Un des défis essentiels associés à la restauration des herbiers de *P. oceanica* réside dans la nomenclature de cet écosystème. En effet, pour parler de l'écosystème formé par cette magnoliophyte marine, on utilise le terme 'herbier' ou 'prairie', en référence à la terminologie anglo-saxonne de '*seagrass meadow*'. Cependant, bien que cette terminologie soit relativement bien adaptée pour certaines magnoliophytes marines à croissance rapide et à une canopée de petite taille (e.g., *Zostera noltii*, *Halophila stipulacea, Thalassia hemprichii*), elle ne correspond pas pour décrire correctement l'écosystème formé par *P. oceanica*. Une analogie plus correcte avec le milieu terrestre pour décrire l'écosystème formé par *P. oceanica* serait d'utiliser le terme 'forêt' (MEDTRIX, 2025). En effet, les forêts terrestres et les forêts de *P. oceanica* présentent de grandes similitudes d'un point de vue structurel et fonctionnel (Figure 4.49). Ces deux écosystèmes sont tous deux formés par des magnoliophytes structurant l'écosystème et générant un écosystème pérenne avec des fonctions de nurseries, de production d'oxygène, de cycles biogéochimiques, de stabilisation des sols et de piégeage de carbone (MEDTRIX, 2025).

Figure 4.49. *Illustration des similitudes entre une forêt terrestre et une forêt sous-marine formée par P. oceanica (photo du bas :©STARESO/Arnaud Abadie).*

De plus, les herbiers de *P. oceanica* forment des communautés climaciques, ou ‘*climax*’ en anglais, des fonds côtiers méditerranéens (Figure 4.50). En effet, ce sont des communautés stables et pérennes qui constituent l’aboutissement de la succession écologique primaire dans un climat déterminé (Shugart, 2024). L’établissement de communautés climaciques dépend d’espèces pionnières à croissance rapide qui vont, à terme, permettre l’installation et la dominance des espèces climaciques (Figure 4.50) (Pandolfi, 2008). En milieu marin, les espèces pionnières telles que certaines macroalgues (*C. prolifera*) et magnoliophytes marines (*C. nodosa*) modifient le sédiment et facilitent l’installation de *P. oceanica* (Molinier & Picard, 1952). Cette communauté climacique se maintient jusqu’à ce qu’une perturbation externe, naturelle (e.g., tempêtes, incendies) ou anthropique (e.g., déforestation, ancrage), réinitialise le cycle de succession (Figure 4.50).

La transplantation de boutures de *P. oceanica* sur des zones de matte morte à la suite de l'action de l'ancrage, correspond aux premiers stades de succession secondaire (Figure 4.50), avec la présence de macroalgues telles que *C. cylindracea* et *C. prolifera* (Figure 4.50), et une physico-chimie du sédiment fortement modifiée. Pour évaluer le succès des projets de transplantation, la zone transplantée est comparée à l'herbier naturel et l'objectif de la restauration vise à atteindre des caractéristiques structurelles similaires sans tenir compte du temps nécessaire pour atteindre le stade climacique. Deux problématiques majeures émergent face à cet objectif complexe. La première, c'est que l'on va transplanter une espèce climacique sur un substrat qui est profondément modifié voire altéré, et que l'on s'attend à atteindre des caractéristiques structurelles similaires à un herbier naturel dans un laps de temps relativement court (Montefalcone, 2024). La seconde problématique, c'est que la majorité des suivis de projets de transplantation de boutures de *P. oceanica* se focalise sur la mesure du taux de survie et de la morphologie foliaire (Pansini et al., 2024), mais omettent d'autres descripteurs clés du rétablissement de l'écosystème. Parmi ceux-ci figurent, d'une part, les descripteurs physiologiques et biochimiques, qui renseignent à la fois sur l'état des boutures transplantées et sur les facteurs limitant leur développement, et d'autre part, le rétablissement de la biodiversité associée (microbienne, épiphyte, vagile), des caractéristiques physico-chimiques du sédiment ainsi que de la structure tridimensionnelle, qui sous-tend l'effet d'« écosystème ingénieur » (sensu Wright & Jones, 2006).

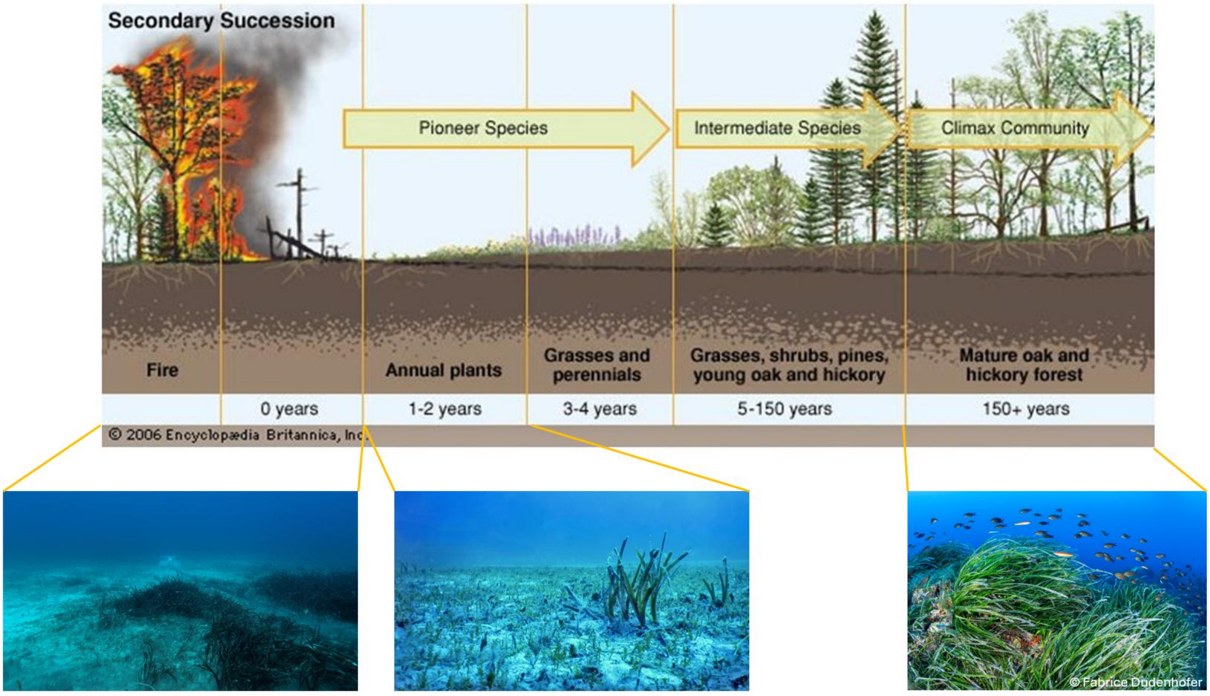


Figure 4.50. *Comparaison de la succession écologique secondaire dans une forêt terrestre (haut) et dans une forêt sous-marine formée par P. oceanica (bas).*

Un des objectifs majeurs de cette thèse était de déterminer si les traits des boutures transplantées, via différentes méthodes et populations donneuses, présentaient des traits similaires aux herbiers naturels trois ans après transplantation. Les résultats ont mis en avant que les transplants ont des traits morphologiques foliaires (e.g., surface foliaire, biomasse,…) réduits en comparaison des herbiers naturels, ce qui pourrait être dû à la présence de conditions environnementales sous-optimales et/ou de la réallocation de ressources internes vers la croissance des rhizomes et racines (Gobert et al., 2005 ; Lepoint et al., 2004). Les traits morphologiques racinaires (e.g., longueur totale, biomasse,…) des transplants sont également réduits en comparaison des herbiers naturels. Ces résultats suggèrent que le succès de la restauration (i.e., rétablissement des fonctions écologiques), en ce qui concerne le rétablissement des caractéristiques structurelles de l'herbier, n'est pas atteint trois ans après transplantation (Chapitre 2.2).

En revanche, la méthode de transplantation influence fortement les traits morphologiques racinaires, contrairement aux traits morphologiques foliaires. Alors que les transplants sur les éléments BESE et le géotextile en fibre de noix de coco ont un système racinaire extrêmement réduit trois ans après transplantation, les boutures transplantées à l'aide des agrafes métalliques ont développé un important système racinaire, bien qu'il ne soit pas encore équivalent à celui présent dans les herbiers naturels (Figure 4.51). Ces résultats démontrent que le choix de la méthode de transplantation influence le succès de la transplantation, mais également le succès de la restauration. Il est probable que ce développement racinaire plus important soit favorisé par le contact direct avec le sédiment, ce qui n'est pas le cas avec les deux autres méthodes de transplantation qui induisent une séparation physique entre le sédiment et les boutures de *P. oceanica* (Chapitre 2.2). Un contact direct avec le sédiment permet d'une part un accès aux nutriments contenus dans l'eau interstitielle du sédiment, mais également le recrutement et l'établissement de communautés bactériennes favorables associées aux racines. L'étude du microbiome racinaire (Chapitre 3.1) a montré que les boutures transplantées à l'aide d'agrafes métalliques présentaient une structure des communautés bactériennes la plus similaire à celle des herbiers naturels. Néanmoins, une différence dans la structure des communautés persiste, notamment avec des abondances plus faibles en *Chromatiales* chez les boutures sur les agrafes métalliques que dans les racines des herbiers naturels (Figure 4). Les bactéries de l'ordre des *Chromatiales*, en particulier le genre *Candidatus* Thiodiazotropha, sont impliquées dans la détoxification du H_2S sédimentaire et la fixation d'azote (Cúcio et al., 2016 ; Martin et al., 2020 ; Petersen et al., 2016 ; Rolando et al., 2024), ce qui pourrait potentiellement influencer positivement la croissance et le développement de *P. oceanica*.

Cependant, malgré des différences contrastées en termes de système racinaire et de structure de communautés bactériennes, aucune différence dans les traits physiologiques et biochimiques n'a été observée entre les méthodes de transplantation (Chapitre 3.2). Cette contradiction pourrait refléter des stratégies contrastées d'utilisation des ressources : les boutures fixées par agrafes métalliques, en contact direct avec le sédiment, privilégieraient une absorption racinaire active des nutriments qui seraient directement utilisés pour la croissance du système racinaire. En revanche, les boutures sur les éléments BESE et le géotextile en fibre de noix de coco adopteraient une stratégie plus conservative, limitant la croissance des racines et régulant plus strictement leurs ressources internes. Une compensation métabolique, par exemple via une augmentation enzymatique, pourrait en outre permettre de maintenir l'assimilation des nutriments tels que l'azote, expliquant la similarité observée des contenus nutritifs foliaires entre les différentes méthodes de transplantation. Enfin, l'ensemble de ces résultats suggère que les boutures transplantées à l'aide d'agrafes métalliques, dans des conditions environnementales appropriées à l'utilisation de cette méthode, permettent d'obtenir un succès de transplantation et un début de rétablissement des fonctionnalités écologiques de l'herbier de *P. oceanica*. Ces résultats démontrent qu'il est essentiel de poursuivre ce suivi sur le long terme afin d'évaluer la restauration des fonctions écosystémiques chez les herbiers transplantés (Figure 4.51).

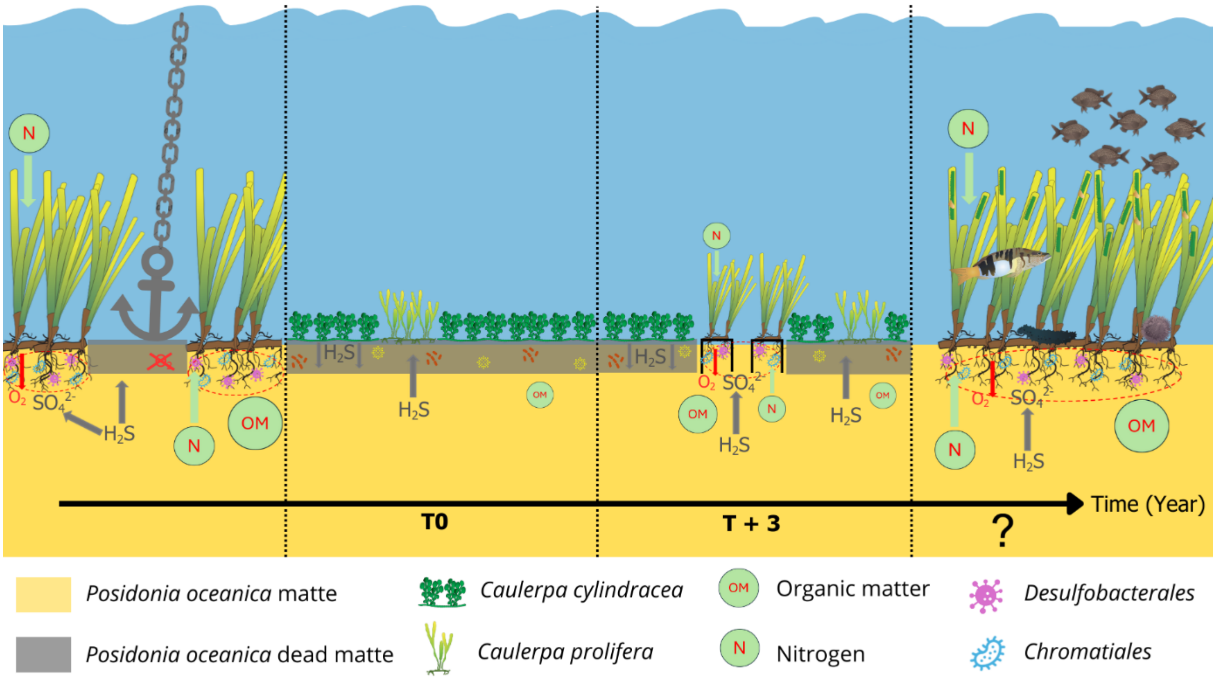


Figure 4.51. *Schéma conceptuel de la restauration des fonctions écosystémiques de l'herbier de P. oceanica après transplantation de boutures à l'aide d'agrafes métalliques.*

3. Quelles boutures pour la transplantation ?

Enfin, le dernier objectif de cette thèse était de déterminer si les boutures-épaves présentaient des performances similaires aux boutures prélevées dans un herbier naturel dans le cadre de leur utilisation comme matériel donneur pour la transplantation (Figure 4.52). Les boutures prélevées dans l'herbier ont toutes été prélevées sur les tombants d'érosion des intermattes naturelles (Gobert et al., 2016) afin de limiter l'impact sur les herbiers donneurs. Tout d'abord, aucune différence en termes de taux de survie et de morphologies foliaire et racinaire entre les deux types de boutures (épaves et intermattes) n'a été observé au terme des trois ans de suivi post-transplantation (Chapitre 2.2). Ensuite, l'étude du microbiome (Chapitre 3.1) a montré que la diversité et la structure des communautés microbiennes associées aux racines des deux types de boutures ne présentaient pas non plus de différences significatives deux ans après transplantation. Ces résultats viennent appuyer les mesures de taux de survie et de morphologie, en suggérant aussi que les boutures-épaves présentent une performance similaire aux boutures d'intermatte. Cependant, on constate une abondance plus importante de bactéries de l'ordre des *Chromatiales*, en particulier le genre *Candidatus* Thiodiazotropha, dans les racines des boutures d'intermatte en comparaison des boutures-épaves. Ces différences d'abondance amènent à un questionnement sur une potentielle performance accrue sur le long-terme des boutures d'intermatte, compte tenu du rôle important joué par ce genre bactérien dans les processus d'oxydation des sulfures et de fixation d'azote (Martin et al., 2020 ; Rolando et al., 2024). De plus, des teneurs en azote foliaires inférieures dans les boutures-épaves en comparaison des boutures d'intermatte ont été observées jusqu'à deux ans après transplantation (Chapitre 3.2), ce qui soutient cette hypothèse. Cependant, les différences de teneurs en azote foliaires entre les deux types de boutures s'estompent trois ans après transplantation. Le suivi des communautés bactériennes n'ayant été effectué que deux ans après transplantation, il n'est pas possible de corréler ces teneurs en azote foliaire similaires aux abondances de *Chromatiales* dans les deux types de boutures. L'étude de l'ensemble des traits physiologiques (activité photosynthétique) et biochimiques (contenus nutritifs foliaires et réserves en carbohydrates) montre que les deux types de boutures convergent vers des traits similaires aux herbiers naturels après transplantation, mais que cette convergence est plus marquée chez les boutures d'intermatte que chez les boutures-épaves (Chapitre 3.2). Cependant, au terme de trois ans après transplantation, les boutures d'intermatte ne présentent pas de traits physiologiques et biochimiques davantage similaires aux herbiers naturels que les boutures-épaves. L'ensemble de ces résultats indique que les boutures d'intermatte présentent une meilleure performance biologique que les boutures-épaves durant les deux premières années de transplantation, mais que ces différences semblent s'estomper au cours de la troisième année. Ces similarités de traits morphologiques, physiologiques et biochimiques au terme de trois ans après transplantation suggèrent donc que les boutures-épaves sont tout à fait adaptées comme matériel biologique viable pour des projets de restauration d'herbiers de *P. oceanica* par transplantation (Figure 4.52). Cependant, afin de maximiser les chances de survie durant les deux premières années après transplantation, ces travaux amènent à la recommandation de réaliser des transplantations mixtes de boutures-épaves et de boutures prélevées dans l'herbier.

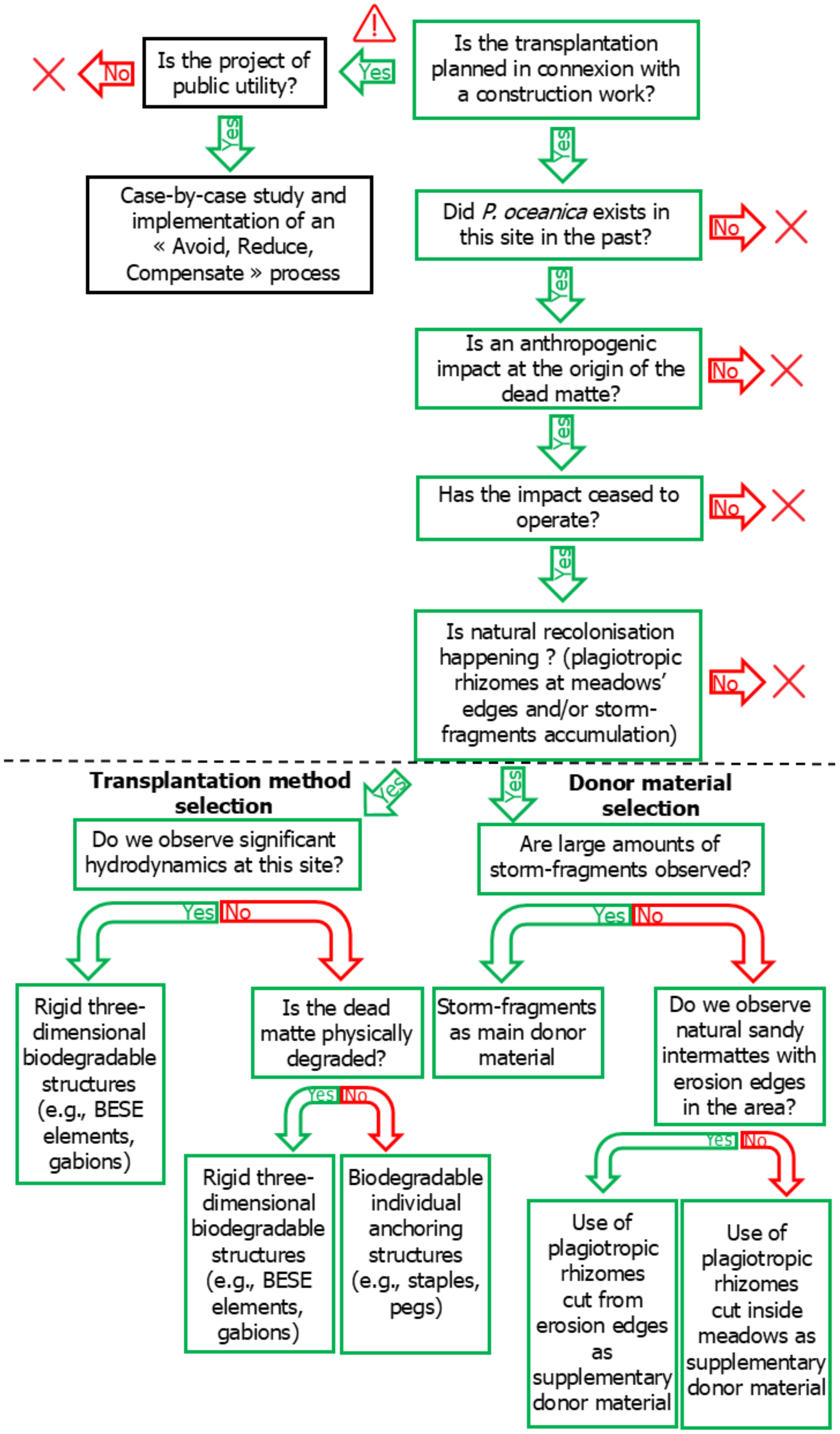


Figure 4.52. *Stratégie de prise de décision pour la transplantation de P. oceanica. La partie au-dessus de la ligne pointillée a été modifiée et adaptée de Boudouresque et al. (2021) et Pergent-Martini et al. (2024). La section au-dessous des pointillés vient compléter la stratégie de prise de décision sur la base des travaux de cette thèse.*

4. Perspectives

Ces travaux de thèse apportent de nombreux éléments de réponse, mis en évidence comme axes majeurs de recherche et développement (R&D) dans le récent guide sur la restauration des herbiers de *P. oceanica* de Pergent-Martini et al. (2024) (Tableau 4.4). Ainsi, ces résultats pourront bénéficier à l'ensemble des praticiens de la restauration des herbiers de *P. oceanica* sur le pourtour du bassin méditerranéen. Ces travaux amènent également de nombreuses nouvelles perspectives de recherche afin de continuer à optimiser les méthodes de restauration des herbiers de *P. oceanica*.

Tableau 4.4. *Connaissances apportées par ces travaux de thèse répondant aux axes de R&D dans le guide sur la restauration de l'herbier de P. oceanica de Pergent-Martini et al. (2024).*

Axes de R&D suggérés par Pergent-Martini et al. (2024)	Nouvelles connaissances apportées dans cette thèse
Améliorer la fixation des transplants par l'utilisation de matériaux biodégradables.	Les méthodes de fixation individuelles (e.g., agrafes, piquets) présentent un très bon rapport cout-bénéfice mais sont adaptées uniquement dans des zones à faible hydrodynamisme et sur de la matte morte peu altérée physiquement. Les structures tri-dimensionnelles rigides, telles que les éléments BESE, sont couteuses mais permettent d'obtenir de bons taux de survie dans des zones à hydrodynamisme important. Leur utilisation sur des fonds à granulométrie grossière et/ou sur des mattes fortement dégradées est également recommandée. Les méthodes qui permettent un contact direct entre les transplants et le sédiment/la matte morte favorisent le développement du système racinaire.
Caractériser le microbiome racinaire de *P. oceanica* et évaluer son influence sur la croissance des herbiers naturels et des transplants.	Dominance des genres bactériens *Desulfobacterales* et *Chromatiales* dans les racines des herbiers de *P. oceanica*. Les méthodes de transplantation qui permettent un contact direct entre les transplants et le sédiment/la matte morte favorisent l'établissement d'une structure des communautés bactériennes associées aux racines des transplants similaire à celle des herbiers naturels. Les boutures d'intermatte ont une plus grande abondance de *Chromatiales*, en particulier le genre *Candidatus* Thiodiazotropha, comparées aux boutures-épaves. Le rôle de ce genre bactérien dans les processus d'oxydation des sulfures et de

	fixation d'azote pourrait conduire à une meilleure croissance et au développement des boutures issues de l'herbier à long-terme.
Evaluer les performances des boutures-épaves par rapport aux boutures issues de l'herbier.	Les boutures-épaves présentent des performances similaires aux boutures issues de l'herbier en termes de taux de survie et de morphologie foliaire et racinaire. Les boutures d'intermatte ont une structure du microbiome racinaire, ainsi que des traits physiologiques et biochimiques plus similaires aux herbiers naturels que les boutures-épaves deux ans après transplantation. Les différences physiologiques et biochimiques entre les transplants s'estompent trois ans après la transplantation. Ces résultats suggèrent une meilleure performance des boutures issues de l'herbier durant les deux premières années qui suivent la transplantation.

Dans l'optique de développer un panel de méthodes de transplantation appropriées à différentes contraintes environnementales et socio-économiques, une quatrième méthode est actuellement en cours d'évaluation dans le cadre du projet REPAIR 2.0. Des gabions (structures métalliques remplies de granite concassé) bouturés ont été installés en juin 2024 dans la baie de l'Alga (i.e., même site que l'ensemble de ces travaux de thèse) afin de déterminer l'efficacité de cette méthode de transplantation (Figure 4.53). Les résultats après un an de suivi montrent un taux de survie global de 85,9 % des transplants. Au terme d'un suivi de plusieurs années, si cette méthode est validée, elle pourra venir compléter le portfolio des méthodes de transplantation (Figure 4.52). En effet, contrairement aux agrafes métalliques et aux éléments BESE, cette méthode pourrait être appropriée pour la restauration d'herbiers de *P. oceanica* sur fonds rocheux. Les gabions pourraient également servir dans le cadre de Zones de Mouillage et d'Equipement Légers (ZMEL), afin de concevoir des corps-morts éco-conçus sur lesquels le bouturage serait possible et permettrait d'amorcer la recolonisation naturelle. De plus, l'utilisation de gabions bouturés pour stabiliser les chaines reliant les corps morts au sein des ZMEL pourrait également être envisagée. Cela permettrait d'une part de limiter l'action de dragage des chaines, mais également de favoriser la recolonisation naturelle dans la zone via la transplantation de boutures de *P. oceanica* sur les gabions.

Figure 4.53. *Boutures de P. oceanica transplantées sur des gabions posés au sein d'une zone de matte morte dans le cadre du projet REPAIR 2.0 (© Fabrice Dudenhofer).*

Comme détaillé précédemment, l'herbier de *P. oceanica* présente un fonctionnement écologique davantage similaire à une forêt terrestre qu'à une prairie. Lors de la restauration de l'herbier de *P. oceanica*, il est dès lors essentiel de ne pas se cantonner à restaurer uniquement la structure de l'herbier en transplantant des boutures, mais bien de restaurer l'écosystème de *P. oceanica* et l'intégrité de son fonctionnement écologique. La restauration doit viser à restaurer la forêt et pas uniquement l'arbre. Le projet pilote de restauration par transplantation à petite échelle réalisé dans ces travaux de thèse a permis de mieux comprendre les conditions nécessaires à la survie et au développement des boutures. L'étape suivante consiste à déployer ces actions à une plus grande échelle afin d'aller vers une véritable restauration de la forêt et pas simplement de l'arbre, et d'évaluer, sur le long terme, la capacité des herbiers de *P. oceanica* transplantés à restaurer l'ensemble de leurs fonctions écosystémiques.

La restauration des forêts terrestres est en cours depuis plus d'un siècle (Rodrigues et al., 2009), ce qui a conduit à l'élaboration de méthodologies sylvicoles largement éprouvées (Lindenmayer & Hobbs, 2004 ; Brockerhoff et al., 2008 ; Chazdon, 2008 ; Rodrigues et al., 2011). En raison des fortes similarités entre les herbiers de *P. oceanica* et les écosystèmes forestiers terrestres, ainsi qu'entre leurs approches de restauration, les enseignements tirés de la sylviculture pourraient considérablement faire progresser les pratiques de restauration des herbiers de *P. oceanica*. En restauration forestière, les plantations d'arbres permettent de remodeler les conditions physico-chimiques des sites dégradés, favorisant ainsi le retour progressif des communautés forestières originelles. Dans ce contexte, les plantations d'arbres constituent un outil d'ingénierie écologique visant à recréer des conditions propices au rétablissement des communautés natives par des processus naturels (Lugo, 1997 ; Lamb et al., 2005 ; Chazdon, 2008). Ces plantations peuvent être constituées de monocultures ou de peuplements mixtes, associant des espèces indigènes, voire parfois exotiques, choisies en fonction de critères d'ingénierie écologique et de leur capacité d'adaptation aux conditions du site dégradé qui doit être restauré (Kelty, 2006). Après transplantation, la canopée modifie les conditions environnementales du sous-bois (e.g., intensité lumineuse, température, humidité). Les arbres stabilisent également le sol, l'enrichissant en matière organique et en nutriments grâce à l'accumulation de litière (Brockerhoff et al., 2008), et réduisent la probabilité d'invasion par des plantes opportunistes (Chazdon, 2003). Ainsi, les principes issus de la sylviculture indiquent que la restauration écosystémique de *P. oceanica* ne peut se limiter à une approche monospécifique. Elle doit prendre en compte l'ensemble des interactions interspécifiques, en particulier avec les bactéries, les champignons, les macroalgues et les autres magnoliophytes marines (Valdez et al., 2020).

Ainsi, un axe de recherche majeur à développer vise à poursuivre les travaux entamés sur le microbiome de *P. oceanica* (Chapitre 3.1), en approfondissant les connaissances sur l'influence du microbiome sur le développement des boutures de *P. oceanica*. D'une part, les travaux sur l'étude du microbiome réalisés dans cette thèse se sont basés sur des boutures-épaves et des boutures d'intermatte, toutes deux ayant un système racinaire quasiment absent lors de la transplantation. Il serait intéressant de tester différentes configurations de transplantation (i.e., nombres de boutures, arrangements, etc) combinant des boutures-épaves, des boutures-épaves avec des boutures d'intermatte (comme dans ces travaux), et des boutures-épaves avec des boutures prélevées au sein d'herbiers naturels avec un système racinaire bien développé et conservé lors de la transplantation. Ce genre de design permettrait de déterminer si le microbiome racinaire des boutures prélevées dans l'herbier peut être transmis aux racines en cours de développement chez les boutures-épaves transplantées, et permettre ainsi une meilleure survie et développement pendant les premières années qui suivent la transplantation. Par exemple, est ce que la co-transplantation de boutures-épaves et de boutures prélevées dans l'herbier amènerait à des abondances plus importantes de

Chromatiales dans les racines des boutures-épaves ? D'autre part, la manipulation de la structure des communautés microbiennes (e.g., inoculation de bactéries possédant des fonctions métaboliques recherchées) est déjà largement utilisée en bioremédiation des écosystèmes terrestres et aquatiques (e.g., dépollution de marées noires ; Trevathan-Tackett et al., 2019), et pourrait être transposée à la restauration des écosystèmes marins. Ces méthodes reposent sur l'exploitation des interactions microbiennes bénéfiques afin d'optimiser le cycle des nutriments et d'améliorer la tolérance des plantes aux stress environnementaux (Trevathan-Tackett et al., 2019 ; Sun et al., 2024). Par exemple, certaines bactéries présentes dans la rhizosphère ont montré une grande efficacité pour stimuler la croissance des magnoliophytes marines, en augmentant la production de biomasse foliaire, l'élongation des rhizomes et l'absorption d'azote, ainsi que la détoxification des sulfures (Sun et al., 2024 ; Zhou et al., 2024). Ainsi, des recherches supplémentaires sont nécessaires pour tester l'inoculation de souches de *Desulfobacterales* et de *Chromatiales* dans les boutures de *P. oceanica*, et évaluer leur influence potentielle sur la morphologie, la croissance et le développement des transplants.

Le second axe majeur nécessitant des besoins de recherche et développement concerne les interactions avec les autres magnoliophytes marines. L'intégration d'interactions positives entre magnoliophytes marines apparaît comme un levier clé pour améliorer le succès de restauration des herbiers marins (Valdez et al., 2020). Il a été démontré qu'une plus grande richesse spécifique peut accroître le succès des transplantations grâce à la complémentarité des niches et des stratégies de croissance différentes (Williams et al., 2017 ; Duffy, 2006). Les travaux novateurs de Hensel et al. (2024) ont montré que la transplantation simultanée de *Zostera marina* avec une espèce pionnière généraliste (*Ruppia maritima* dans leur étude), tolérante à de larges gammes de conditions environnementales, à croissance rapide et avec un haut taux de reproduction, permettait d'augmenter la surface totale de la zone restaurée et la biodiversité associée, en comparaison de la transplantation de *Z. marina* seule. En mer Méditerranée, la magnoliophyte marine *C. nodosa* (Figure 4.54) est une espèce rudérale (Grime, 1974) à la croissance rapide et qui montre une grande tolérance à des conditions environnementales dégradées (Montefalcone, 2024). En tant qu'espèce pionnière dans le processus de succession écologique primaire avant la colonisation par *P. oceanica*, *C. nodosa* colonise les fonds sableux et modifie les caractéristiques physico-chimiques du sédiment, créant ainsi des conditions environnementales favorables à l'établissement des herbiers de *P. oceanica* (Molinier & Picard, 1952). De plus, l'étude de Cúcio et al. (2016) sur le microbiome de la rhizosphère de *Z. marina*, *Z. noltii* et *C. nodosa* a montré qu'il y avait peu de différences entre espèces au sein d'une même région. Le rhizobiome est dominé par des bactéries du cycle du soufre, telles que les ordres *Desulfobacterales* et *Desulfobulbales* aussi observés dans ces travaux de thèse sur le microbiome racinaire de *P. oceanica*, soulignant l'importance de la réduction des sulfates dans les herbiers marins. Il est donc probable que les deux espèces natives de Méditerranée *C. nodosa* et *P. oceanica* présentent un microbiome similaire, et que *C. nodosa* joue également un rôle de pionnière en modifiant le microbiome du sédiment de manière favorable pour le développement de *P. oceanica*.

Figure 4.54. *Recolonisation naturelle de P. oceanica entourée d'un herbier de C. nodosa sur de la matte morte de P. oceanica.*

Une stratégie de restauration innovante pourrait être de transplanter simultanément *P. oceanica* et *C. nodosa*, cette dernière permettant une amélioration des conditions environnementales locales et facilitant ainsi le développement et la recolonisation des boutures de *P. oceanica* transplantées (Montefalcone, 2024). De plus, cette hypothèse d'interactions facilitatrices est étayée par les travaux de Molenaar & Meinesz (1995), qui ont montré un haut taux de survie (85 %) des boutures de *P. oceanica* transplantées sur un fond sableux avec la présence d'un herbier de *C. nodosa*. Ce design de transplantations mixtes pourrait intégrer simultanément la transplantation de boutures et de graines de *P. oceanica*, ainsi que de boutures et de graines de *C. nodosa*. La transplantation de boutures de *P. oceanica* permet d'augmenter la survie et le développement des graines de *P. oceanica* (Mancini et al., 2024), tandis que ces dernières augmentent la diversité génétique de l'herbier transplanté (Terrados et al., 2013; Procaccini et al., 2023) et permettent de réduire la relativement faible disponibilité de boutures-épaves et les prélèvements destructeurs de boutures au sein des herbiers naturels. Enfin, la transplantation de graines de *C. nodosa* permet d'avoir un matériel biologique abondant et une logistique de transplantation simplifiée. En revanche, la transplantation de boutures de *C. nodosa* assure une action directe des plants transplantés sur la modification des conditions environnementales locales pour faciliter la persistance et le développement des boutures et graines de *P. oceanica* transplantées. La mise en place de ces transplantations mixtes permettrait de dépasser les visions actuelles centrées sur une restauration monospécifique, en allant vers une approche globale à l'échelle de l'écosystème prenant en compte les interactions intra et interspécifiques.

Annexes

Annexes au Chapitre II

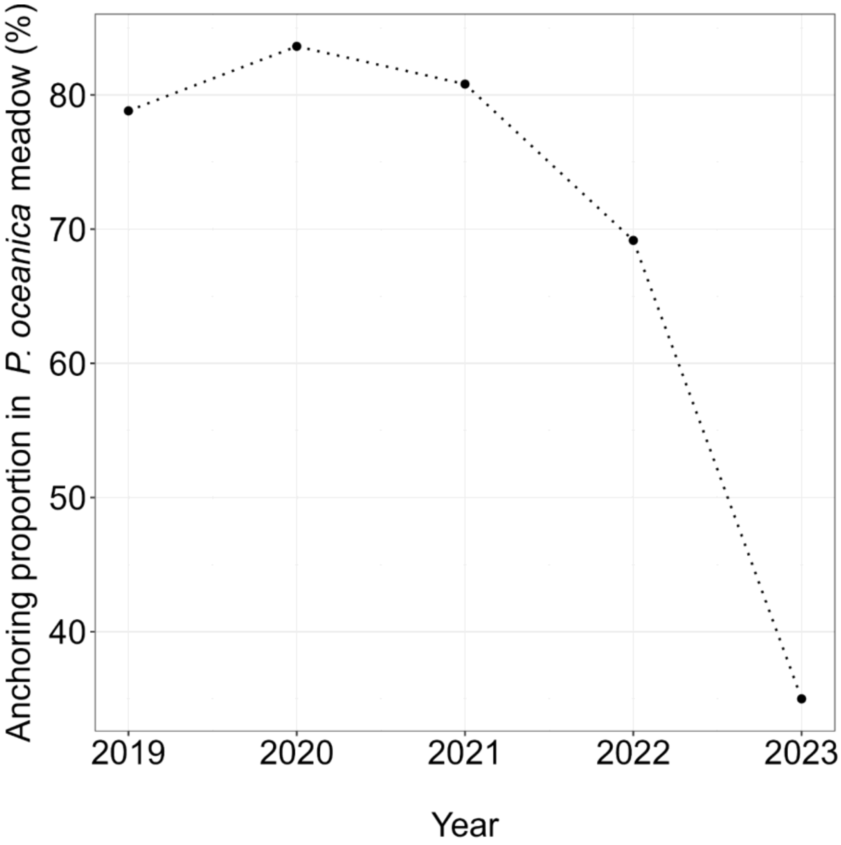


Figure S2.1. *Annual changes in the percentages of anchored boats over 20m long in P. oceanica meadows in Calvi Bay.*

Table S2.1. *Settings used for the different steps of the photogrammetric workflow in Agisoft Metashape (v1.8.4; Agisoft LLC).*

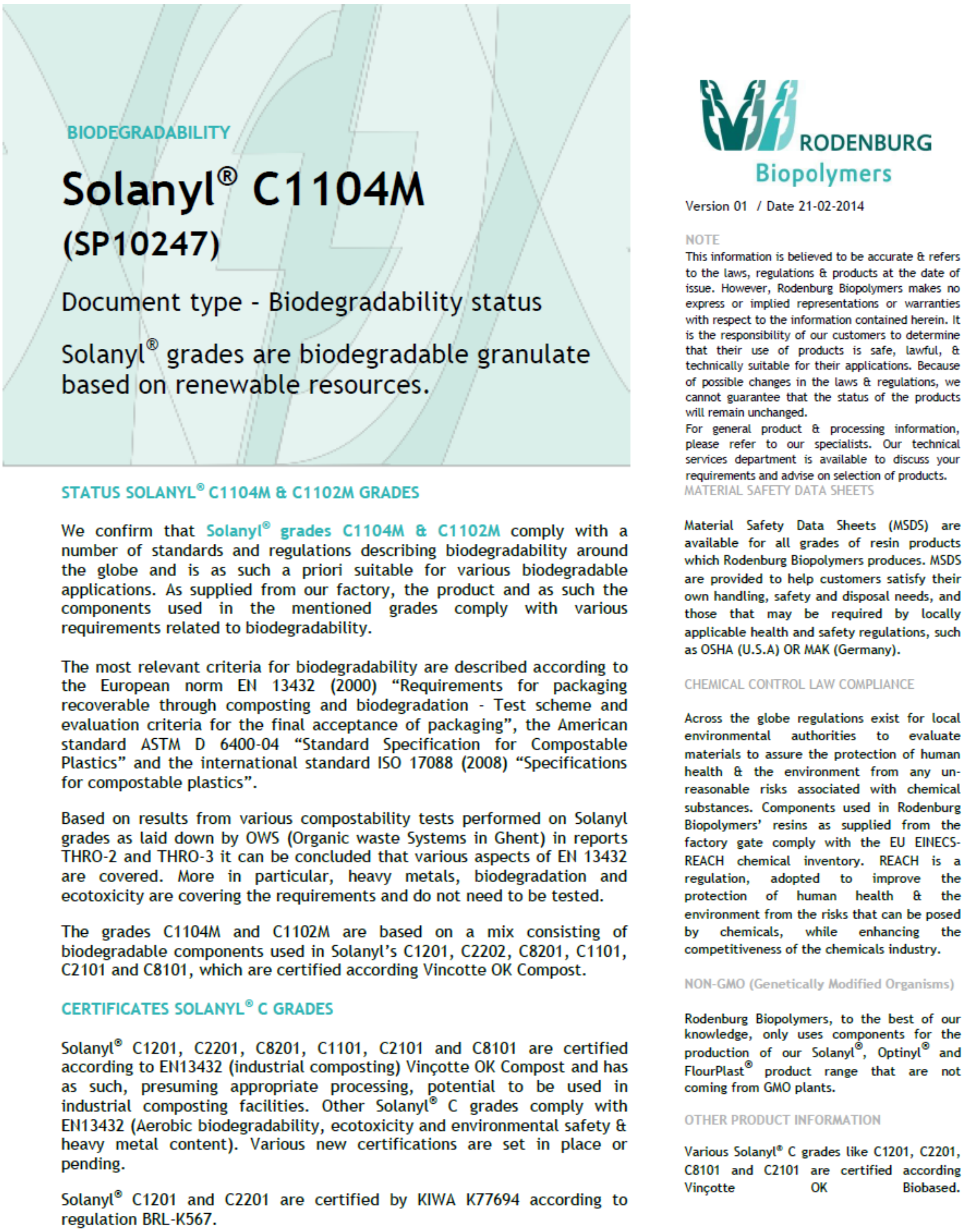
Photogrammetric step	**Parameter choices**
Bundle adjustment	accuracy = high; key point limit = 40000; tie point limit = 4000; generic preselection enabled
Optimization	f (focal length), cx – cy (principal point offset), b1 – b2 (affinity and non-orthogonality coefficients), k1 – k2 – k3 – k4 (radial distortion coefficients) and p1-p2 (tangential distortion coefficients)
Dense cloud	medium quality; mild depth filtering; reuse depth maps enabled
Mesh	source data = depth maps; medium quality; medium face counts
Texture	texture type = diffuse map; source data = images; mapping mode = orthophoto; blending mode = mosaic; hole filling and ghosting filter enabled
Orthomosaic	planar projection; surface = mesh; blending mode = mosaic; hole filling enabled

Table S2.2 *Annual recolonisation and erosion rates (m²/year) measured in each quadrat for the time intervals May 2022-May 2023 and May 2023-May 2024 for the six study sites. Recolonisation rates are positive values while erosion rates are negative values.*

Site	Bathymetry	Quadrat	Recolonisation/Erosion rate between May 2022 – May 2023 (m²/year)	Recolonisation/Erosion rate between May 2023 – May 2024 (m²/year)
AP1	Shallow	Q1	0,112	-0,028
AP1	Shallow	Q2	0,004	0,0213
AP1	Shallow	Q3	-0,173	-0,046
AP1	Shallow	Q4	-0,017	0,133
AP2	Shallow	Q1	-0,056	0,0995
AP2	Shallow	Q2	0,053	-0,013
AP2	Shallow	Q3	0,027	0,0281
AP2	Shallow	Q4	-0,255	0,0212
AP3	Shallow	Q1	0,205	0,4269
AP3	Shallow	Q2	0,183	0,593
AP3	Shallow	Q3	0,112	-0,149
AP3	Shallow	Q4	0,287	0,3201
AP3	Deep	Q1	0,107	0,0447
AP4	Deep	Q2	0,108	0,041
AP4	Deep	Q3	0,815	0,1081
AP4	Deep	Q4	-0,06	0
AP5	Deep	Q1	0,37	0,2613
AP5	Deep	Q2	0,196	0,4497
AP5	Deep	Q3	-0,078	0,3694
AP5	Deep	Q4	0,028	0,0064
AP6	Deep	Q1	0,638	-0,137
AP6	Deep	Q2	-0,044	0,0538
AP6	Deep	Q3	0,043	-0,082
AP6	Deep	Q4	0,009	-0,045

Table S2.3. *Number of storm-fragments accumulated within each of the 4 quadrats on each experimental site over the two years of monitoring.*

Site	Bathymetry	Quadrat	Number of storm-fragments in May 2022	Number of storm-fragments in May 2023	Number of storm-fragments in May 2024
AP1	Shallow	Q1	2	0	0
AP1	Shallow	Q2	1	1	0
AP1	Shallow	Q3	0	6	2
AP1	Shallow	Q4	1	4	0
AP2	Shallow	Q1	0	6	2
AP2	Shallow	Q2	1	3	0
AP2	Shallow	Q3	1	2	0
AP2	Shallow	Q4	0	1	3
AP3	Shallow	Q1	1	9	3
AP3	Shallow	Q2	0	0	1
AP3	Shallow	Q3	2	0	6
AP3	Shallow	Q4	3	25	25
AP3	Deep	Q1	0	0	0
AP4	Deep	Q2	0	2	0
AP4	Deep	Q3	0	3	1
AP4	Deep	Q4	0	0	0
AP5	Deep	Q1	0	0	0
AP5	Deep	Q2	1	1	0
AP5	Deep	Q3	1	0	0
AP5	Deep	Q4	0	0	0
AP6	Deep	Q1	1	0	2
AP6	Deep	Q2	0	1	1
AP6	Deep	Q3	0	0	2
AP6	Deep	Q4	0	0	0



BIODEGRADABILITY

Solanyl® C1104M

(SP10247)

Document type - Biodegradability status

Solanyl® grades are biodegradable granulate based on renewable resources.

STATUS SOLANYL® C1104M & C1102M GRADES

We confirm that **Solanyl® grades C1104M & C1102M** comply with a number of standards and regulations describing biodegradability around the globe and is as such a priori suitable for various biodegradable applications. As supplied from our factory, the product and as such the components used in the mentioned grades comply with various requirements related to biodegradability.

The most relevant criteria for biodegradability are described according to the European norm EN 13432 (2000) "Requirements for packaging recoverable through composting and biodegradation - Test scheme and evaluation criteria for the final acceptance of packaging", the American standard ASTM D 6400-04 "Standard Specification for Compostable Plastics" and the international standard ISO 17088 (2008) "Specifications for compostable plastics".

Based on results from various compostability tests performed on Solanyl grades as laid down by OWS (Organic waste Systems in Ghent) in reports THRO-2 and THRO-3 it can be concluded that various aspects of EN 13432 are covered. More in particular, heavy metals, biodegradation and ecotoxicity are covering the requirements and do not need to be tested.

The grades C1104M and C1102M are based on a mix consisting of biodegradable components used in Solanyl's C1201, C2202, C8201, C1101, C2101 and C8101, which are certified according Vincotte OK Compost.

CERTIFICATES SOLANYL® C GRADES

Solanyl® C1201, C2201, C8201, C1101, C2101 and C8101 are certified according to EN13432 (industrial composting) Vinçotte OK Compost and has as such, presuming appropriate processing, potential to be used in industrial composting facilities. Other Solanyl® C grades comply with EN13432 (Aerobic biodegradability, ecotoxicity and environmental safety & heavy metal content). Various new certifications are set in place or pending.

Solanyl® C1201 and C2201 are certified by KIWA K77694 according to regulation BRL-K567.

RODENBURG
Biopolymers

Version 01 / Date 21-02-2014

NOTE

This information is believed to be accurate & refers to the laws, regulations & products at the date of issue. However, Rodenburg Biopolymers makes no express or implied representations or warranties with respect to the information contained herein. It is the responsibility of our customers to determine that their use of products is safe, lawful, & technically suitable for their applications. Because of possible changes in the laws & regulations, we cannot guarantee that the status of the products will remain unchanged.

For general product & processing information, please refer to our specialists. Our technical services department is available to discuss your requirements and advise on selection of products.

MATERIAL SAFETY DATA SHEETS

Material Safety Data Sheets (MSDS) are available for all grades of resin products which Rodenburg Biopolymers produces. MSDS are provided to help customers satisfy their own handling, safety and disposal needs, and those that may be required by locally applicable health and safety regulations, such as OSHA (U.S.A) OR MAK (Germany).

CHEMICAL CONTROL LAW COMPLIANCE

Across the globe regulations exist for local environmental authorities to evaluate materials to assure the protection of human health & the environment from any unreasonable risks associated with chemical substances. Components used in Rodenburg Biopolymers' resins as supplied from the factory gate comply with the EU EINECS-REACH chemical inventory. REACH is a regulation, adopted to improve the protection of human health & the environment from the risks that can be posed by chemicals, while enhancing the competitiveness of the chemicals industry.

NON-GMO (Genetically Modified Organisms)

Rodenburg Biopolymers, to the best of our knowledge, only uses components for the production of our Solanyl®, Optinyl® and FlourPlast® product range that are not coming from GMO plants.

OTHER PRODUCT INFORMATION

Various Solanyl® C grades like C1201, C2201, C8101 and C2101 are certified according Vinçotte OK Biobased.

Figure S2.2 *Solanyl® C1104M biodegradability document status.*

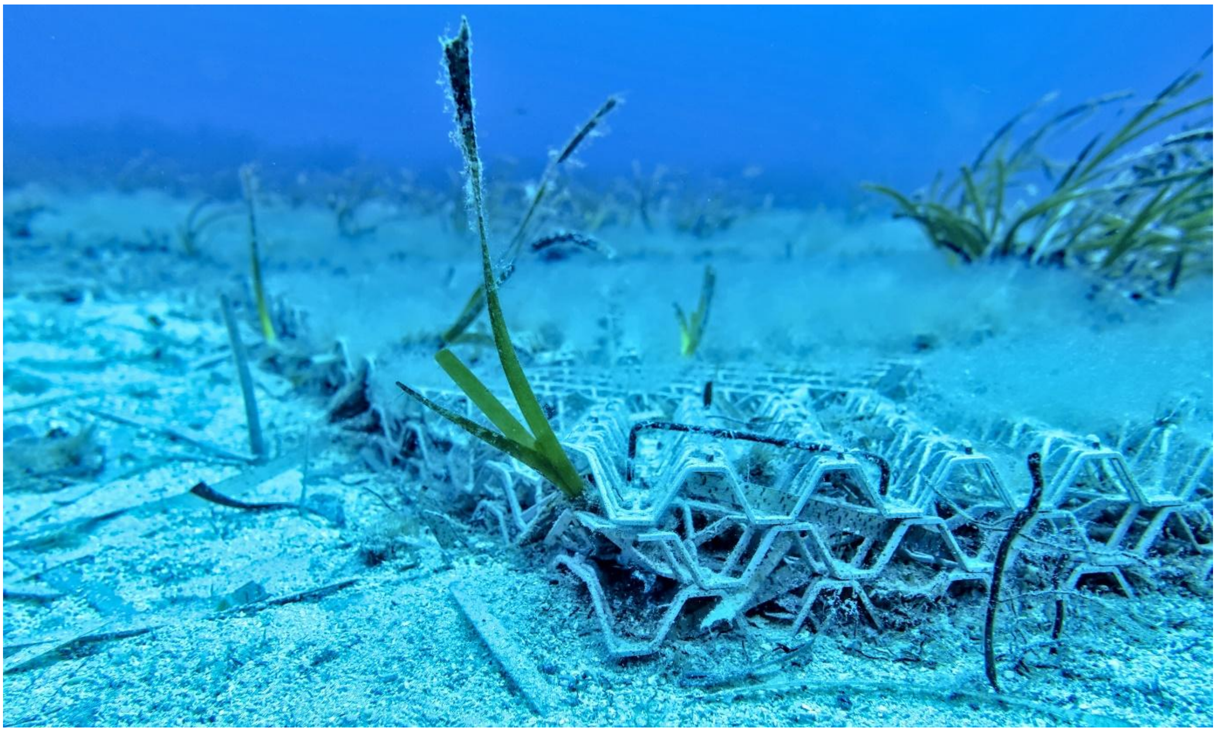


Figure S2.3 *P. oceanica drifting fragment trapped in the BESE elements lattices.*

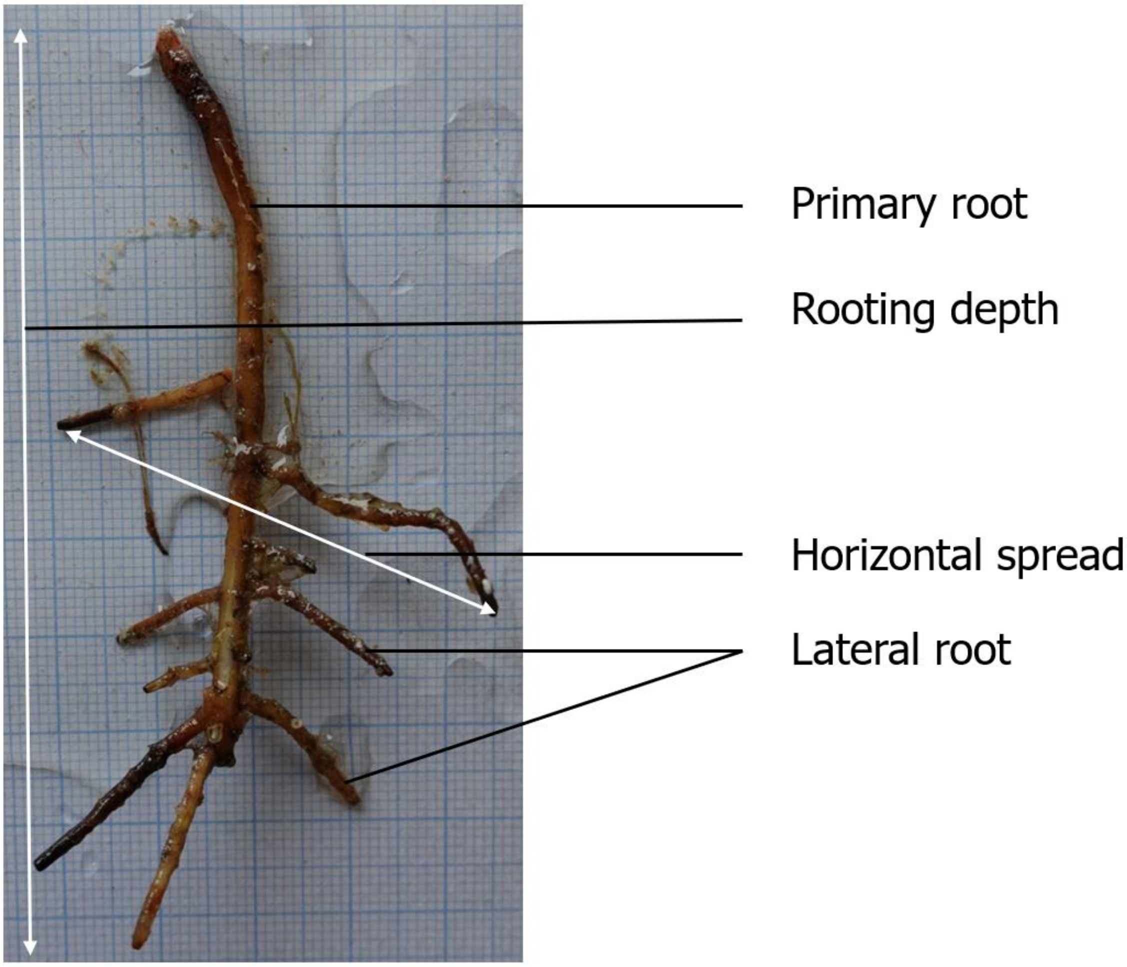


Figure S2.4 *P. oceanica root morphological traits.*

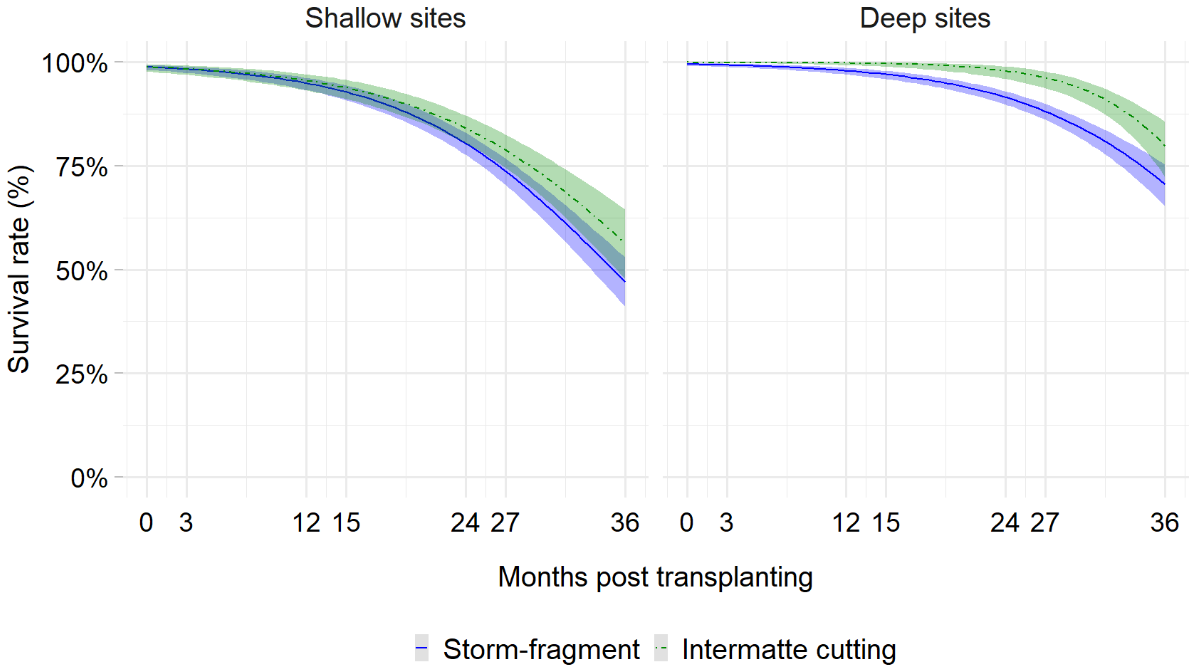


Figure S2.5 *Temporal dynamics of transplanted cuttings' survival rates according to donor source at shallow and deep sites. Shaded areas around the curves represent 95% confidence intervals.*

Table S2.4 *PERMANOVA results on the effect of transplantation effect (transplant vs control meadows) and months post transplanting (0, 3, 12, 15, 24, 27, 36) on P. oceanica leaf morphological traits (number of leaves, maximum leaf length, leaf surface area and biomass). Significant values ($P_{(perm)}$ < 0.05) are highlighted in bold.*

Factor	df	Variable	pseudo-F	$P_{(perm)}$
Transplantation effect	1	Number of leaves	15.469	**0.001**
Months	6		4.6916	**0.001**
Transplantation effect *Months	6		5.796	**0.001**
Transplantation effect	1	Max. leaf length	187.68	**0.001**
Months	6		5.5439	**0.004**
Transplantation effect *Months	6		4.5952	**0.01**
Transplantation effect	1	Leaf surface area	333.5	**0.001**
Months	6		17.129	**0.001**
Transplantation effect *Months	6		12.776	**0.001**
Transplantation effect	1	Biomass	203.7	**0.001**
Months	6		5.6274	**0.001**
Transplantation effect *Months	6		7.8526	**0.001**

Table S2.5 *Comparison of means (pair-wise tests) of P. oceanica leaf morphological traits for the significant interactions of transplantation effect (t = transplant vs c = control meadows) and months post transplanting (0, 3, 12, 15, 24, 27, 36).*

	Pair-wise tests of transplantation effect * months post transplanting						
	0	3	12	15	24	27	36
Number of leaves	t > c	t = c	t = c	c > t	c > t	c > t	c > t
Max. leaf length	c > t	c > t	c > t	c > t	c > t	c > t	c > t
Leaf surface area	c = t	c > t	c > t	c > t	c > t	c > t	c > t
Biomass	c > t	c > t	c = t	c > t	c > t	c > t	c > t

Table S2.6 *PERMANOVA results on the effect of transplantation method + effect (Trans.; BESE element vs coconut fiber mat vs iron staple vs control meadows) and bathymetry (Bath., shallow vs deep sites) on P. oceanica root morphological traits (number of primary roots, number of lateral roots, root biomass, total root length, maximum horizontal spread, maximum rooting depth) thirty-six months after transplantation. Significant values ($P_{(perm)} < 0.05$) are highlighted in bold.*

Factor	df	Variable	Pseudo-F	$P_{(perm)}$
Trans.	3	Number of primary roots	71.188	**0.001**
Bath.	1		4.1375	**0.045**
Trans.*Bath.	3		1.112	0.349
Trans.	3	Number of lateral roots	51.302	**0.001**
Bath.	1		0.48901	0.498
Trans.*Bath.	3		12.016	**0.001**
Trans.	3	Root biomass	50.151	**0.001**
Bath.	1		1.1993	0.286
Trans.*Bath.	3		1.5065	0.225
Trans.	3	Total root length	47.106	**0.001**
Bath.	1		0.47325	0.487
Trans.*Bath.	3		2.2725	0.089
Trans.	3	Maximum horizontal spread	77.839	**0.001**
Bath.	1		0.29372	0.573
Trans.*Bath.	3		5.8242	0.074
Trans.	3	Maximum rooting depth	29.831	**0.001**
Bath.	1		3.5112	0.058
Trans.*Bath.	3		0.87921	0.434

Table S2.7 *Comparison of means (pair-wise tests) of P. oceanica root morphological traits for the significant interactions of transplantation method + effect (b = BESE element, f = coconut fiber mat, s = iron staple, c = control meadow) and bathymetry (shallow vs deep).*

Variable	Pair-wise tests of transplantation method + effect	
Number of primary roots	b = f < s < c	
Number of lateral roots	b = f < s < c	
Root biomass	b = f < s < c	
Total root length	b = f < s < c	
Maximum horizontal spread	b = f < s < c	
Maximum rooting depth	b = f < s < c	
	Pair-wise tests of transplantation method + effect * bathymetry	
	Shallow	Deep
Number of lateral roots	b = f < s < c	b = f < s = c

Annexes au Chapitre III

Figure S3.6 *Erosion side of a natural sandy intermatte (©STARESO/ Arnaud Abadie). The P. oceanica fragments hanging against the matte often lack a well-developed root system.*

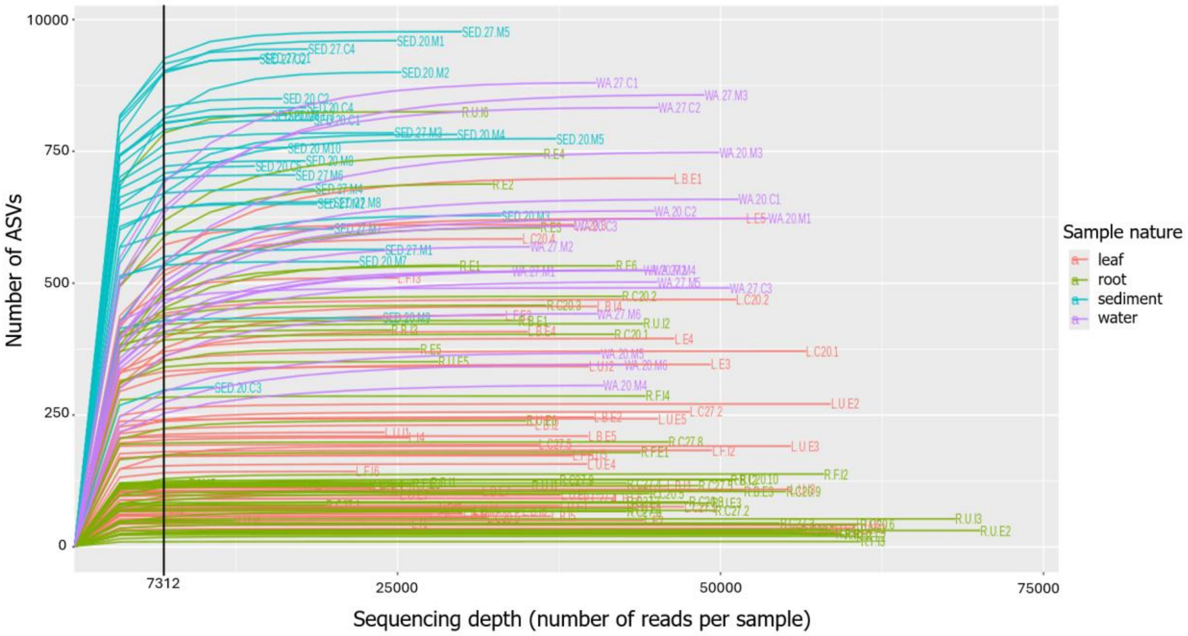


Figure S3.7 *Rarefaction curves depicting the number of ASVs against sequencing depth of the sequenced samples (comprising seawater, sediment, seagrass leaf and root samples). The cut off line of 7312 sequences used for samples normalization by rarefaction is represented by the black line.*

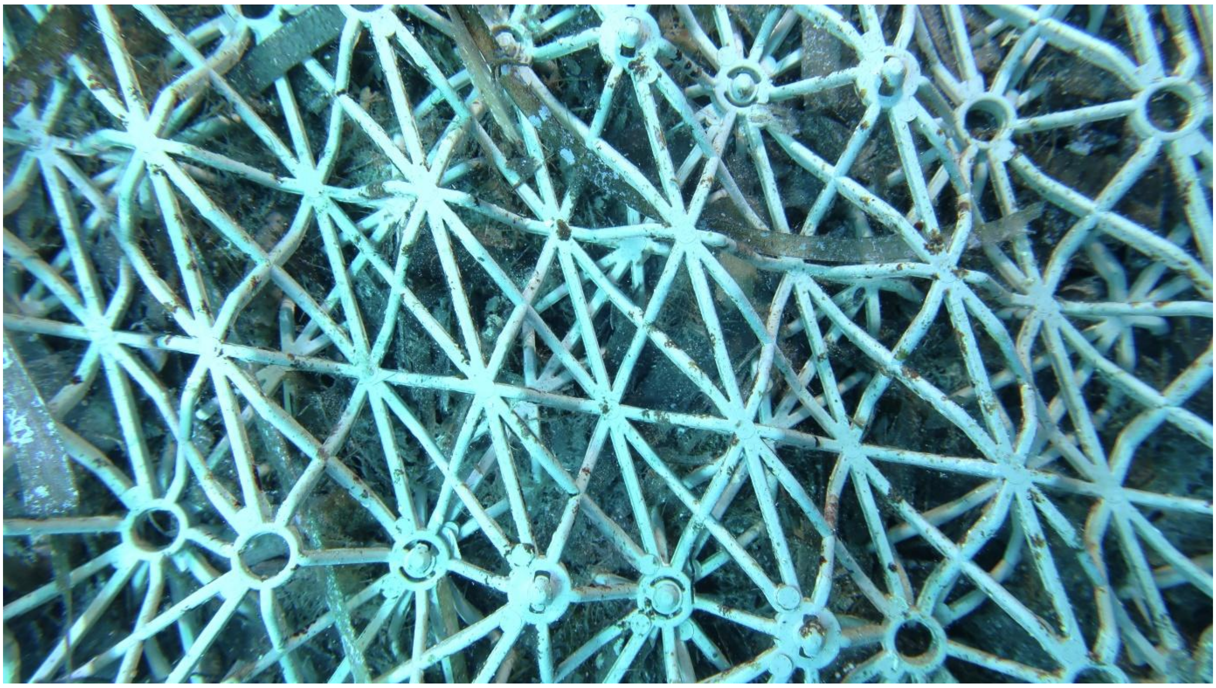


Figure S3.8 *Accumulation of dead P. oceanica leaves within the layers of the BESE elements.*

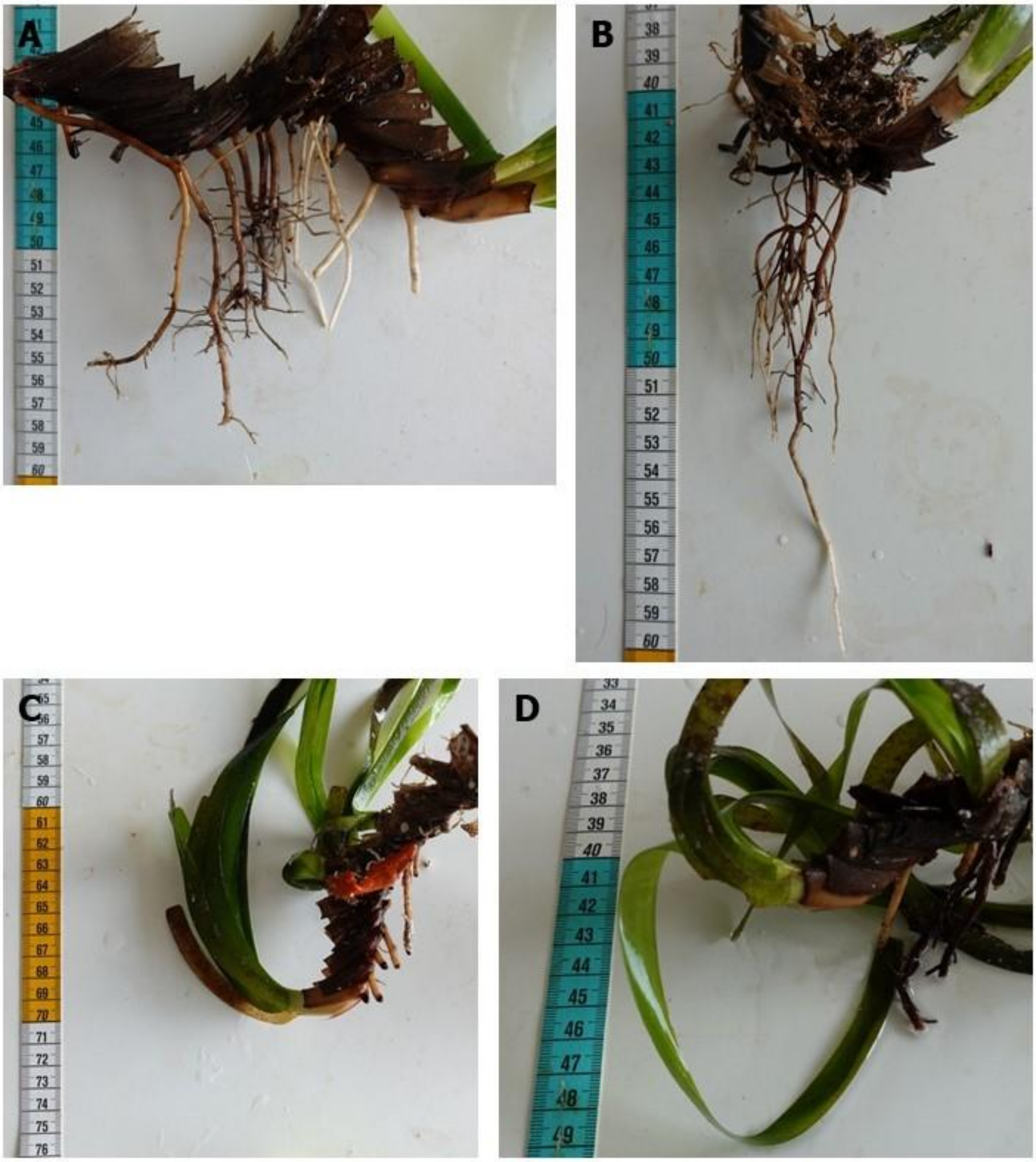


Figure S3.9 *Differences in roots' length and complexity between (A) P. oceanica control meadows and cuttings attached to (B) iron staples, (C) coconut fiber mats, and (D) BESE elements two years after transplantation on dead matte.*

Table S3.8. List of the top 20 most abundant bacterial orders for the interaction "Sample tissueXTransplantation method" . Abundance of the bacterial orders are reported in relative abundance (%). The top 5 most abundant are given in bold, excluding the categories "Not_assigned" and "Others".

Transplantation_Method	Tissue	Order	Relative abundance (%)
Iron staple	Leaf	**Rhizobiales**	**12.0423132880698**
Iron staple	Leaf	**Peptostreptococcales_Tissierellales**	**11.849842386033**
Iron staple	Leaf	**Bacillales**	**5.53922162948594**
Iron staple	Leaf	**Burkholderiales**	**5.37251454898157**
Iron staple	Leaf	**Micrococcales**	**4.66173617846751**
Iron staple	Leaf	**Frankiales**	3.76606450048497
Iron staple	Leaf	Corynebacteriales	3.54328322017459
Iron staple	Leaf	Enterobacterales	3.03255334626576
Iron staple	Leaf	Solirubrobacterales	3.02194471387003
Iron staple	Leaf	Gaiellales	2.40209747817653
Iron staple	Leaf	Lactobacillales	2.20811105722599
Iron staple	Leaf	Bacteroidales	1.69738118331717
Iron staple	Leaf	Pseudomonadales	1.59584141610087
Iron staple	Leaf	Lachnospirales	1.43368089233754
Iron staple	Leaf	Flavobacteriales	1.1578564500485
Iron staple	Leaf	Microtrichales	0.789585354025218
Iron staple	Leaf	Rhodobacterales	0.318258971871969
Iron staple	Leaf	Not_Assigned	4.35711687681862
Iron staple	Leaf	Others	31.1818016488846
Coconut fiber mat	Leaf	**Bacillales**	**9.8111281265952**
Coconut fiber mat	Leaf	**Peptostreptococcales_Tissierellales**	**8.44648630253531**
Coconut fiber mat	Leaf	**Micrococcales**	**8.38523055980943**
Coconut fiber mat	Leaf	**Corynebacteriales**	**7.35409222392377**
Coconut fiber mat	Leaf	**Lactobacillales**	**6.68708524757529**
Coconut fiber mat	Leaf	Enterobacterales	5.68657478305258
Coconut fiber mat	Leaf	Burkholderiales	4.65203335034882
Coconut fiber mat	Leaf	Pseudomonadales	3.77063127445976
Coconut fiber mat	Leaf	Rhizobiales	2.84839203675345
Coconut fiber mat	Leaf	Lachnospirales	2.72588055130168
Coconut fiber mat	Leaf	Bacteroidales	1.81725370086779
Coconut fiber mat	Leaf	Microtrichales	1.80023821677727
Coconut fiber mat	Leaf	Flavobacteriales	1.32720775906075
Coconut fiber mat	Leaf	Solirubrobacterales	1.13663433724689
Coconut fiber mat	Leaf	Rhodobacterales	1.11961885315637
Coconut fiber mat	Leaf	Frankiales	0.527480006806194
Coconut fiber mat	Leaf	Gaiellales	0.207588905904373
Coconut fiber mat	Leaf	Not_Assigned	7.6841926152799
Coconut fiber mat	Leaf	Others	23.9339799217288
BESE element	Leaf	**Burkholderiales**	**16.108367809956**
BESE element	Leaf	**Rhizobiales**	**11.7910049379421**
BESE element	Leaf	**Bacillales**	**5.9538902976111**
BESE element	Leaf	**Frankiales**	**4.95462431602829**
BESE element	Leaf	**Corynebacteriales**	**4.01541438676098**
BESE element	Leaf	Micrococcales	3.82523688776191
BESE element	Leaf	Enterobacterales	3.4515547844655
BESE element	Leaf	Solirubrobacterales	3.35646603496597

BESE element	Leaf	Lactobacillales	2.54570932870679
BESE element	Leaf	Gaiellales	2.4856532763913
BESE element	Leaf	Lachnospirales	1.92346189777125
BESE element	Leaf	Bacteroidales	1.72494328039504
BESE element	Leaf	Flavobacteriales	1.59315361003603
BESE element	Leaf	Pseudomonadales	1.29787801948485
BESE element	Leaf	Rhodobacterales	1.23281729614307
BESE element	Leaf	Microtrichales	0.667289470172161
BESE element	Leaf	Peptostreptococcales_Tissierellales	0.20685973575337
BESE element	Leaf	Not_Assigned	2.65080742025891
BESE element	Leaf	Others	30.1881756305885
Donor population	Leaf	**Bacillales**	**19.1206716342426**
Donor population	Leaf	**Enterobacterales**	**7.07036649920766**
Donor population	Leaf	**Lachnospirales**	**5.99480764691999**
Donor population	Leaf	**Burkholderiales**	**4.27526214639738**
Donor population	Leaf	**Flavobacteriales**	**3.14912842644728**
Donor population	Leaf	Rhizobiales	3.02100542836913
Donor population	Leaf	Bacteroidales	2.91985569304427
Donor population	Leaf	Pseudomonadales	2.84567922047271
Donor population	Leaf	Micrococcales	2.80859098418692
Donor population	Leaf	Lactobacillales	2.57257493509559
Donor population	Leaf	Peptostreptococcales_Tissierellales	2.17134765164031
Donor population	Leaf	Rhodobacterales	1.83418186722411
Donor population	Leaf	Frankiales	1.77686368387336
Donor population	Leaf	Microtrichales	1.54084763478202
Donor population	Leaf	Corynebacteriales	1.46667116221046
Donor population	Leaf	Solirubrobacterales	1.1261337199501
Donor population	Leaf	Gaiellales	0.849657776728818
Donor population	Leaf	Not_Assigned	8.93489328702923
Donor population	Leaf	Others	26.5214606021781
Control meadow	Leaf	**Rhizobiales**	**10.9133952088298**
Control meadow	Leaf	**Burkholderiales**	**9.84572933651072**
Control meadow	Leaf	**Bacillales**	**8.53505581337012**
Control meadow	Leaf	**Frankiales**	**5.33989715289101**
Control meadow	Leaf	**Solirubrobacterales**	**5.16273673648564**
Control meadow	Leaf	Flavobacteriales	3.2907939295121
Control meadow	Leaf	Enterobacterales	3.25316693841716
Control meadow	Leaf	Gaiellales	2.87376144487646
Control meadow	Leaf	Micrococcales	2.84397341025963
Control meadow	Leaf	Lactobacillales	2.10554371002132
Control meadow	Leaf	Lachnospirales	2.02401856264894
Control meadow	Leaf	Corynebacteriales	1.9660102847109
Control meadow	Leaf	Bacteroidales	1.66185877336009
Control meadow	Leaf	Pseudomonadales	1.3247836448012
Control meadow	Leaf	Microtrichales	1.25423303649818
Control meadow	Leaf	Rhodobacterales	0.512667753668632
Control meadow	Leaf	Peptostreptococcales_Tissierellales	0.08622852125925
Control meadow	Leaf	Not_Assigned	4.56070487896651
Control meadow	Leaf	Others	32.3670512981312

Iron staple	Root	**Microtrichales**	**14.4729995558094**
Iron staple	Root	**Chromatiales**	**12.2685449584364**
Iron staple	Root	**Pseudomonadales**	**4.77187638809569**
Iron staple	Root	**Rhizobiales**	**4.56881781838949**
Iron staple	Root	**Bacteroidales**	**2.87454787740339**
Iron staple	Root	Burkholderiales	2.71337013769909
Iron staple	Root	Micrococcales	1.71711403007805
Iron staple	Root	Corynebacteriales	1.71076844977473
Iron staple	Root	Peptostreptococcales_Tissierellales	1.64985087886287
Iron staple	Root	Bacillales	1.54832159400977
Iron staple	Root	Enterobacterales	1.36303064915287
Iron staple	Root	Rhodobacterales	1.35033948854623
Iron staple	Root	Flavobacteriales	1.15108826702202
Iron staple	Root	Desulfobacterales	0.843962180341392
Iron staple	Root	Solirubrobacterales	0.807157814582144
Iron staple	Root	Lactobacillales	0.789390189732852
Iron staple	Root	Lachnospirales	0.657402119423821
Iron staple	Root	Not_Assigned	15.8893330795101
Iron staple	Root	Others	27.8621739958119
Coconut fiber mat	Root	**Rhizobiales**	**17.7676388403772**
Coconut fiber mat	Root	**Bacillales**	**9.19053440447083**
Coconut fiber mat	Root	**Corynebacteriales**	**7.37862382116661**
Coconut fiber mat	Root	**Frankiales**	**4.28964373035278**
Coconut fiber mat	Root	**Flavobacteriales**	**4.03422982885086**
Coconut fiber mat	Root	Pseudomonadales	3.93381068808942
Coconut fiber mat	Root	Microtrichales	3.48847362906043
Coconut fiber mat	Root	Enterobacterales	2.13063220398184
Coconut fiber mat	Root	Solirubrobacterales	2.12844917918268
Coconut fiber mat	Root	Gaiellales	2.12626615438351
Coconut fiber mat	Root	Burkholderiales	2.0018337408313
Coconut fiber mat	Root	Micrococcales	1.92106182326231
Coconut fiber mat	Root	Bacteroidales	1.85993712888578
Coconut fiber mat	Root	Lactobacillales	1.41460006985679
Coconut fiber mat	Root	Rhodobacterales	0.855745721271394
Coconut fiber mat	Root	Chromatiales	0.331819769472581
Coconut fiber mat	Root	Desulfobacterales	0.146262661543835
Coconut fiber mat	Root	Not_Assigned	6.92237163814181
Coconut fiber mat	Root	Others	28.0562347188264
BESE element	Root	**Pseudomonadales**	**12.2622065338419**
BESE element	Root	**Flavobacteriales**	**6.0766151003293**
BESE element	Root	**Rhizobiales**	**5.10530430456516**
BESE element	Root	**Microtrichales**	**4.91814929757646**
BESE element	Root	**Rhodobacterales**	**4.35905332733174**
BESE element	Root	Lachnospirales	3.78337400203738
BESE element	Root	Frankiales	2.60832484423492
BESE element	Root	Bacillales	2.09660988841771
BESE element	Root	Burkholderiales	1.79574044680297
BESE element	Root	Micrococcales	1.67728791073417
BESE element	Root	Solirubrobacterales	1.36694226623392
BESE element	Root	Enterobacterales	1.26507308521475

BESE element	Root	Corynebacteriales	1.18689441140935
BESE element	Root	Lactobacillales	1.13951339698183
BESE element	Root	Gaiellales	1.07081092606193
BESE element	Root	Bacteroidales	0.604107933950866
BESE element	Root	Peptostreptococcales_Tissierellales	0.371940963256023
BESE element	Root	Not_Assigned	13.7570775390301
BESE element	Root	Others	34.5549738219895
Donor population	Root	**Enterobacterales**	**37.5085684908032**
Donor population	Root	**Flavobacteriales**	**16.9270535816292**
Donor population	Root	**Rhodobacterales**	**4.69838912372901**
Donor population	Root	**Rhizobiales**	**3.99577287787044**
Donor population	Root	**Microtrichales**	**3.08894093453673**
Donor population	Root	Pseudomonadales	2.90186221866789
Donor population	Root	Bacteroidales	1.73511938763852
Donor population	Root	Burkholderiales	0.756883354278532
Donor population	Root	Micrococcales	0.552667656803382
Donor population	Root	Chromatiales	0.458414257968696
Donor population	Root	Lachnospirales	0.458414257968696
Donor population	Root	Bacillales	0.401290985947675
Donor population	Root	Lactobacillales	0.274191705700903
Donor population	Root	Solirubrobacterales	0.169941734262539
Donor population	Root	Gaiellales	0.139952016451502
Donor population	Root	Corynebacteriales	0.0728321718268022
Donor population	Root	Frankiales	0.0556951902204958
Donor population	Root	Not_Assigned	5.08254312807038
Donor population	Root	Others	20.6900491260139
Control meadow	Root	**Chromatiales**	**16.691528817958**
Control meadow	Root	**Corynebacteriales**	**10.2408222901107**
Control meadow	Root	**Desulfobacterales**	**9.44540373749654**
Control meadow	Root	**Rhizobiales**	**6.95186619394522**
Control meadow	Root	**Pseudomonadales**	**3.83966977613847**
Control meadow	Root	Microtrichales	3.52446537860288
Control meadow	Root	Bacteroidales	2.50222226763254
Control meadow	Root	Burkholderiales	1.60258702604395
Control meadow	Root	Bacillales	1.56478293298458
Control meadow	Root	Solirubrobacterales	0.857744219549825
Control meadow	Root	Frankiales	0.853146424448009
Control meadow	Root	Gaiellales	0.762212254656545
Control meadow	Root	Flavobacteriales	0.757614459554729
Control meadow	Root	Micrococcales	0.715212571393541
Control meadow	Root	Enterobacterales	0.615082811398445
Control meadow	Root	Rhodobacterales	0.491453209771847
Control meadow	Root	Lachnospirales	0.393366914266447
Control meadow	Root	Not_Assigned	16.9975376252899
Control meadow	Root	Others	20.7759034667375

Table S3.9. List of the top 20 most abundant bacterial orders for the interaction "Sample tissueXSample origin" . Abundance of the bacterial orders are reported in relative abundance (%). The top 5 most abundant are given in bold, not taking into account the categories "Not_assigned" and "Others".

Sample_Origin	Tissue	Order	Relative abundance (%)
Storm-fragment	Leaf	**Rhizobiales**	**15.2928691720304**
Storm-fragment	Leaf	**Bacillales**	**6.68789074215055**
Storm-fragment	Leaf	**Burkholderiales**	**5.60823125014419**
Storm-fragment	Leaf	**Corynebacteriales**	**4.87115602002445**
Storm-fragment	Leaf	**Frankiales**	**4.71889634807484**
Storm-fragment	Leaf	Micrococcales	4.43860013380395
Storm-fragment	Leaf	Solirubrobacterales	3.60924631462385
Storm-fragment	Leaf	Gaiellales	3.48467021939235
Storm-fragment	Leaf	Lactobacillales	3.20898793457448
Storm-fragment	Leaf	Enterobacterales	3.15938819295453
Storm-fragment	Leaf	Peptostreptococcales_Tissierellales	2.83525964887997
Storm-fragment	Leaf	Bacteroidales	1.78905114540799
Storm-fragment	Leaf	Lachnospirales	1.64255888527464
Storm-fragment	Leaf	Pseudomonadales	1.37841142408933
Storm-fragment	Leaf	Flavobacteriales	1.25844925831084
Storm-fragment	Leaf	Rhodobacterales	0.867418737167509
Storm-fragment	Leaf	Microtrichales	0.709391653401619
Storm-fragment	Leaf	Not_Assigned	4.04180220084435
Storm-fragment	Leaf	Others	30.3873393775809
Intermatte cutting	Leaf	**Burkholderiales**	**14.1447703988691**
Intermatte cutting	Leaf	**Peptostreptococcales_Tissierellales**	**11.6104868913858**
Intermatte cutting	Leaf	**Bacillales**	**6.27960185954328**
Intermatte cutting	Leaf	**Micrococcales**	**5.80742942916685**
Intermatte cutting	Leaf	**Enterobacterales**	**4.37488159256183**
Intermatte cutting	Leaf	Corynebacteriales	3.90999577376528
Intermatte cutting	Leaf	Rhizobiales	3.77883676532739
Intermatte cutting	Leaf	Lactobacillales	3.15656013640537
Intermatte cutting	Leaf	Pseudomonadales	2.54157011906323
Intermatte cutting	Leaf	Frankiales	2.21367259796849
Intermatte cutting	Leaf	Lachnospirales	2.1510077383815
Intermatte cutting	Leaf	Solirubrobacterales	1.76481732464769
Intermatte cutting	Leaf	Bacteroidales	1.65697547326542
Intermatte cutting	Leaf	Flavobacteriales	1.4835541176642
Intermatte cutting	Leaf	Microtrichales	1.21686413384048
Intermatte cutting	Leaf	Rhodobacterales	0.766551538203703
Intermatte cutting	Leaf	Gaiellales	0.167592066337312
Intermatte cutting	Leaf	Not_Assigned	4.68966321281278
Intermatte cutting	Leaf	Others	28.2137600373075
Donor pop. - SF	Leaf	**Burkholderiales**	**7.94609988191985**
Donor pop. - SF	Leaf	**Lachnospirales**	**7.34875321247482**
Donor pop. - SF	Leaf	**Enterobacterales**	**7.25845662290755**
Donor pop. - SF	Leaf	**Bacillales**	**7.01535042022644**
Donor pop. - SF	Leaf	**Bacteroidales**	**5.60533444467597**
Donor pop. - SF	Leaf	Peptostreptococcales_Tissierellales	4.4731541293325
Donor pop. - SF	Leaf	Micrococcales	4.20226436063069
Donor pop. - SF	Leaf	Lactobacillales	3.63270125720636

Donor pop. - SF	Leaf	Frankiales	3.41043272904077
Donor pop. - SF	Leaf	Rhizobiales	3.02146280475099
Donor pop. - SF	Leaf	Pseudomonadales	2.75751892755435
Donor pop. - SF	Leaf	Solirubrobacterales	2.29909008821282
Donor pop. - SF	Leaf	Corynebacteriales	1.66006806973675
Donor pop. - SF	Leaf	Gaiellales	1.62533861221088
Donor pop. - SF	Leaf	Rhodobacterales	1.44474543307634
Donor pop. - SF	Leaf	Flavobacteriales	1.0418837257762
Donor pop. - SF	Leaf	Microtrichales	0.555671320413975
Donor pop. - SF	Leaf	Not_Assigned	5.09133847329305
Donor pop. - SF	Leaf	Others	29.6103354865597

Donor pop. - IC	**Leaf**	**Bacillales**	**30.5399030271262**
Donor pop. - IC	**Leaf**	**Enterobacterales**	**6.89293670554318**
Donor pop. - IC	**Leaf**	**Flavobacteriales**	**5.13694142314245**
Donor pop. - IC	**Leaf**	**Lachnospirales**	**4.71759926615123**
Donor pop. - IC	**Leaf**	**Rhizobiales**	**3.02057397457738**
Donor pop. - IC	Leaf	Pseudomonadales	2.92884287773555
Donor pop. - IC	Leaf	Microtrichales	2.47018739352641
Donor pop. - IC	Leaf	Rhodobacterales	2.20154632420391
Donor pop. - IC	Leaf	Lactobacillales	1.57253308871707
Donor pop. - IC	Leaf	Micrococcales	1.49390643428122
Donor pop. - IC	Leaf	Corynebacteriales	1.28423535578561
Donor pop. - IC	Leaf	Burkholderiales	0.812475429170489
Donor pop. - IC	Leaf	Bacteroidales	0.386581050976281
Donor pop. - IC	Leaf	Frankiales	0.235879963307561
Donor pop. - IC	Leaf	Gaiellales	0.117939981653781
Donor pop. - IC	Leaf	Solirubrobacterales	0.0196566636089634
Donor pop. - IC	Leaf	Chromatiales	0
Donor pop. - IC	Leaf	Not_Assigned	12.5606080461276
Donor pop. - IC	Leaf	Others	23.6076529943651

Control meadow	Leaf	**Rhizobiales**	**10.9133952088298**
Control meadow	Leaf	**Burkholderiales**	**9.84572933651072**
Control meadow	Leaf	**Bacillales**	**8.53505581337012**
Control meadow	Leaf	**Frankiales**	**5.33989715289101**
Control meadow	Leaf	**Solirubrobacterales**	**5.16273673648564**
Control meadow	Leaf	Flavobacteriales	3.2907939295121
Control meadow	Leaf	Enterobacterales	3.25316693841716
Control meadow	Leaf	Gaiellales	2.87376144487646
Control meadow	Leaf	Micrococcales	2.84397341025963
Control meadow	Leaf	Lactobacillales	2.10554371002132
Control meadow	Leaf	Lachnospirales	2.02401856264894
Control meadow	Leaf	Corynebacteriales	1.9660102847109
Control meadow	Leaf	Bacteroidales	1.66185877336009
Control meadow	Leaf	Pseudomonadales	1.3247836448012
Control meadow	Leaf	Microtrichales	1.25423303649818
Control meadow	Leaf	Rhodobacterales	0.512667753668632
Control meadow	Leaf	Peptostreptococcales_Tissierellales	0.08622852125925
Control meadow	Leaf	Not_Assigned	4.56070487896651
Control meadow	Leaf	Others	32.3670512981312

Storm-fragment	Root	**Microtrichales**	**14.7369076530295**
Storm-fragment	Root	**Pseudomonadales**	**5.8135624883243**
Storm-fragment	Root	**Chromatiales**	**5.75253751790273**
Storm-fragment	Root	**Rhizobiales**	**4.9741577931378**
Storm-fragment	Root	**Flavobacteriales**	**4.06625568217199**
Storm-fragment	Root	Corynebacteriales	3.85204558191668
Storm-fragment	Root	Burkholderiales	2.92670776511613
Storm-fragment	Root	Bacillales	2.90553583660253
Storm-fragment	Root	Rhodobacterales	2.04745002802167
Storm-fragment	Root	Peptostreptococcales_Tissierellales	1.53932374369512
Storm-fragment	Root	Micrococcales	1.27529734105486
Storm-fragment	Root	Enterobacterales	1.24167133694502
Storm-fragment	Root	Lactobacillales	1.13207547169811
Storm-fragment	Root	Bacteroidales	1.08724079955165
Storm-fragment	Root	Desulfobacterales	0.890466405131079
Storm-fragment	Root	Frankiales	0.490690578491811
Storm-fragment	Root	Solirubrobacterales	0.424683977831745
Storm-fragment	Root	Not_Assigned	15.7842954106731
Storm-fragment	Root	Others	28.4388816240115

Intermatte cutting	Root	**Rhizobiales**	**11.4425733075972**
Intermatte cutting	Root	**Pseudomonadales**	**7.01579999768837**
Intermatte cutting	Root	**Chromatiales**	**6.01024052520256**
Intermatte cutting	Root	**Bacillales**	**4.60245726372242**
Intermatte cutting	Root	**Microtrichales**	**3.75062125082352**
Intermatte cutting	Root	Frankiales	3.49749765947364
Intermatte cutting	Root	Bacteroidales	2.88838289855407
Intermatte cutting	Root	Corynebacteriales	2.46882187727551
Intermatte cutting	Root	Flavobacteriales	2.37520082294063
Intermatte cutting	Root	Micrococcales	2.21569828592563
Intermatte cutting	Root	Lachnospirales	2.14288191033183
Intermatte cutting	Root	Solirubrobacterales	2.13479120193252
Intermatte cutting	Root	Rhodobacterales	1.90940718223743
Intermatte cutting	Root	Gaiellales	1.8782001641258
Intermatte cutting	Root	Enterobacterales	1.8342791756724
Intermatte cutting	Root	Burkholderiales	1.69095805545603
Intermatte cutting	Root	Lactobacillales	0.973196638888568
Intermatte cutting	Root	Not_Assigned	10.1989158450745
Intermatte cutting	Root	Others	30.6949918515008

Donor pop. - SF	Root	**Enterobacterales**	**37.5085684908032**
Donor pop. - SF	Root	**Flavobacteriales**	**16.9270535816292**
Donor pop. - SF	Root	**Rhodobacterales**	**4.69838912372901**
Donor pop. - SF	Root	**Rhizobiales**	**3.99577287787044**
Donor pop. - SF	Root	**Microtrichales**	**3.08894093453673**
Donor pop. - SF	Root	Pseudomonadales	2.90186221866789
Donor pop. - SF	Root	Bacteroidales	1.73511938763852
Donor pop. - SF	Root	Burkholderiales	0.756883354278532
Donor pop. - SF	Root	Micrococcales	0.552667656803382
Donor pop. - SF	Root	Chromatiales	0.458414257968696
Donor pop. - SF	Root	Lachnospirales	0.458414257968696
Donor pop. - SF	Root	Bacillales	0.401290985947675

Donor pop. - SF	Root	Lactobacillales	0.274191705700903
Donor pop. - SF	Root	Solirubrobacterales	0.169941734262539
Donor pop. - SF	Root	Gaiellales	0.139952016451502
Donor pop. - SF	Root	Corynebacteriales	0.0728321718268022
Donor pop. - SF	Root	Frankiales	0.0556951902204958
Donor pop. - SF	Root	Not_Assigned	5.08254312807038
Donor pop. - SF	Root	Others	20.6900491260139
Control meadow	Root	**Chromatiales**	**16.691528817958**
Control meadow	Root	**Corynebacteriales**	**10.2408222901107**
Control meadow	Root	**Desulfobacterales**	**9.44540373749654**
Control meadow	Root	**Rhizobiales**	**6.95186619394522**
Control meadow	Root	**Pseudomonadales**	**3.83966977613847**
Control meadow	Root	Microtrichales	3.52446537860288
Control meadow	Root	Bacteroidales	2.50222226763254
Control meadow	Root	Burkholderiales	1.60258702604395
Control meadow	Root	Bacillales	1.56478293298458
Control meadow	Root	Solirubrobacterales	0.857744219549825
Control meadow	Root	Frankiales	0.853146424448009
Control meadow	Root	Gaiellales	0.762212254656545
Control meadow	Root	Flavobacteriales	0.757614459554729
Control meadow	Root	Micrococcales	0.715212571393541
Control meadow	Root	Enterobacterales	0.615082811398445
Control meadow	Root	Rhodobacterales	0.491453209771847
Control meadow	Root	Lachnospirales	0.393366914266447
Control meadow	Root	Not_Assigned	16.9975376252899
Control meadow	Root	Others	20.7759034667375

Table S3.10. Results of the PERMANOVA main test for Observed ASVs alpha diversity index for the factors "Sample nature", "Transplantation method", and their interaction "Sample natureXTransplantation method". Bold face values are significant at p<0.05.

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	42.486	42.486	1.5231	0.21	996
Transplantation method	4	312.68	78.171	2.8023	**0.041**	998
Sample naturexTransplantation method	4	510.75	127.69	4.5774	**0.002**	999
Res	95	2650	27.895			
Total	104	3361.4				

Table S3.11. *Results of the PERMANOVA pair-wise test within the factor "Transplantation method" for Observed ASVs alpha diversity index. Bold face values are significant at p<0.05.*

Groups	t	P(perm)	Unique perms
Donor population. BESE element	2.2203	**0.026**	998
Donor population. Coconut fiber mat	3.2272	**0.004**	994
Donor population. Iron staple	2.8778	**0.01**	995
Donor population. Control meadow	2.4395	**0.027**	998
BESE element. Coconut fiber mat	0.82816	0.385	996
BESE element. Iron staple	0.48242	0.631	997
BESE element. Control meadow	0.040943	0.965	996
Coconut fiber mat. Iron staple	0.39377	0.703	996

Coconut fiber mat. Control meadow	0.96766	0.349	998
Iron staple. Control meadow	0.5998	0.531	995

Table S3.12. Results of the PERMANOVA pair-wise test within the interaction between the factors "Sample nature" and "Transplantation method" for Observed ASVs alpha diversity index. Bold face values are significant at $p<0.05$.

Within level ‘leaf’ of factor ‘Sample nature’				
Groups	t	P(perm)	Unique perms	P(MC)
Donor population. BESE element	0.2755	0.8	998	0.803
Donor population. Coconut fiber mat	0.4133	0.688	985	0.722
Donor population. Iron staple	0.0027471	0.998	992	0.996
Donor population. Control meadow	0.93783	0.356	979	0.358
BESE element. Coconut fiber mat	0.20723	0.831	993	0.813
BESE element. Iron staple	0.35187	0.734	996	0.726
BESE element. Control meadow	0.85366	0.413	998	0.416
Coconut fiber mat. Iron staple	0.55967	0.581	992	0.608
Coconut fiber mat. Control meadow	0.56119	0.562	987	0.592
Iron staple. Control meadow	1.1694	0.264	995	0.227

Within level 'root' of factor 'Sample nature'				
Groups	t	P(perm)	Unique perms	P(MC)
Donor population. BESE element	3.1008	**0.013**	413	0.013
Donor population. Coconut fiber mat	5.7579	**0.001**	793	0.001
Donor population. Iron staple	3.4688	**0.005**	968	0.005
Donor population. Control meadow	5.7307	**0.001**	996	0.001
BESE element. Coconut fiber mat	1.149	0.248	568	0.273
BESE element. Iron staple	0.31679	0.77	970	0.763
BESE element. Control meadow	0.8282	0.419	995	0.391
Coconut fiber mat. Iron staple	0.87128	0.414	992	0.408
Coconut fiber mat. Control meadow	0.82139	0.43	997	0.432
Iron staple. Control meadow	0.42929	0.673	998	0.688

Table S3.13. Results of the PERMANOVA main test for exponentiated Shannon diversity index for the factors "Sample nature", "Transplantation method", and their interaction "Sample natureXTransplantation method".

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	0.38682	0.38682	1.8539	0.174	997
Transplantation method	4	0.61231	0.15308	0.73363	0.557	999
Sample naturexTransplantation method	4	1.053	0.26325	1.2617	0.266	999
Res	95	19.823	0.20866			
Total	104	22.121				

Table S3.14. Results of the PERMANOVA main test for Simpson diversity index for the factors "Sample nature", "Transplantation method", and their interaction "Sample natureXTransplantation method".

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	0.052376	0.052376	1.7282	0.181	999
Transplantation method	4	0.025749	0.006437	0.2124	0.945	998
Sample naturexTransplantation method	4	0.09636	0.02409	0.79487	0.512	998

Res	95	2.8791	0.030307
Total	104	3.0707	

Table S3.15. Results of the PERMANOVA main test for Observed ASVs alpha diversity index for the factors "Sample nature", "Sample origin", and their interaction "Sample natureXSample origin". Bold face values are significant at p<0.05.

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	25.717	25.717	0.9597	0.318	999
Sample origin	4	400.74	100.18	3.7387	**0.005**	999
Sample naturexSample origin	3	306.03	102.01	3.8067	**0.015**	998
Res	96	2572.5	26.797			
Total	104	3361.4				

Table S3.16. Results of the PERMANOVA pair-wise test within the factor "Sample origin" for Observed ASVs alpha diversity index. Bold face values are significant at p<0.05.

Groups	t	P(perm)	Unique perms
Donor pop. - IC. Donor pop. - SF	1.6621	0.119	996
Donor pop. - IC. Storm-fragment	1.782	0.082	998
Donor pop. - IC. Intermatte cutting	0.69278	0.521	996
Donor pop. - IC. Control meadow	1.8291	0.084	997
Donor pop. - SF. Storm-fragment	3.6055	**0.002**	998
Donor pop. - SF. Intermatte cutting	3.2561	**0.005**	998
Donor pop. - SF. Control meadow	2.8377	**0.008**	997
Storm-fragment. Intermatte cutting	0.14068	0.9	998
Storm-fragment. Control meadow	0.5732	0.539	996
Intermatte cutting. Control meadow	0.66099	0.51	995

Table S3.17. Results of the PERMANOVA pair-wise test within the interaction between the factors "Sample nature" and "Sample origin" for Observed ASVs alpha diversity index. Bold face values are significant at p<0.05.

Within level 'leaf' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Donor pop. - IC. Donor pop. - SF	1.3885	0.195	126	0.213
Donor pop. - IC. Storm-fragment	1.9701	0.059	959	0.07
Donor pop. - IC. Intermatte cutting	0.85637	0.417	954	0.399
Donor pop. - IC. Control meadow	1.5551	0.149	781	0.15
Donor pop. - SF. Storm-fragment	0.30297	0.766	970	0.77
Donor pop. - SF. Intermatte cutting	1.1926	0.24	927	0.239
Donor pop. - SF. Control meadow	0.050943	0.956	788	0.965
Storm-fragment. Intermatte cutting	1.5241	0.128	996	0.129
Storm-fragment. Control meadow	0.44462	0.647	995	0.654
Intermatte cutting. Control meadow	1.5223	0.147	998	0.145

Within level 'root' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms
Donor pop. - SF. Storm-fragment	5.0479	**0.002**	943
Donor pop. - SF. Intermatte cutting	3.2485	**0.007**	975
Donor pop. - SF. Control meadow	5.7307	**0.001**	999

Storm-fragment. Intermatte cutting	0.79552	0.418	995
Storm-fragment. Control meadow	0.34978	0.725	996
Intermatte cutting. Control meadow	0.66058	0.517	996

Table S3.18. Results of the PERMANOVA main test for exponentiated Shannon diversity index for the factors "Sample nature", "Sample origin", and their interaction "Sample natureXSample origin".

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	0.53514	0.53514	2.6993	0.101	996
Sample origin	4	1.3126	0.32815	1.6552	0.171	999
Sample naturexSample origin	3	0.82472	0.27491	1.3867	0.253	999
Res	96	19.032	0.19825			
Total	104	22.121				

Table S3.19. Results of the PERMANOVA main test for Simpson diversity index for the factors "Sample nature", "Sample origin", and their interaction "Sample natureXSample origin".

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
Sample nature	1	0.060368	0.060368	2.0435	0.15	993
Sample origin	4	0.10231	0.025577	0.86578	0.465	998
Sample naturexSample origin	3	0.058464	0.019488	0.65967	0.584	999
Res	96	2.836	0.029542			
Total	104	3.0707				

Table S3.20. Results of the PERMANOVA main test for ASVs composition for the factors "Sample nature", "Transplantation method", and their interaction "Sample natureXTransplantation method". Bold face values are significant at p<0.05.

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms	P(MC)
Sample nature	3	98848	32949	10.418	**0.001**	999	0.001
Transplantation method	5	24577	4915.3	1.5542	**0.003**	995	0.002
Sample nature xTransplantation method	5	23282	4656.4	1.4723	**0.003**	997	0.003
Res	136	4,30E+05	3162.6				
Total	149	6,23E+05					

Table S3.21. Results of the PERMANOVA pair-wise test within the factor "Sample nature" for ASVs structure. Bold face values are significant at p<0.05.

Groups	t	P(perm)	Unique perms	P(MC)
Leaf, root	2.5548	**0.001**	999	0.001
Leaf, sediment	2.2895	**0.001**	995	0.001
Leaf, water	2.6793	**0.001**	999	0.001
Root, sediment	2.5033	**0.001**	997	0.001
Root, water	2.8627	**0.001**	999	0.001
Sediment, water	4.8673	**0.001**	996	0.001

Table S3.22. Results of the PERMANOVA pair-wise test within the factor "Transplantation method " for ASVs structure. Bold face values are significant at p<0.05.

Groups	t	P(perm)	Unique perms	P(MC)
Donor population, BESE element	1.2285	0.079	997	0.099

Donor population, Coconut fiber mat	1.4705	**0.007**	997	0.016
Donor population, Iron staple	1.4995	**0.015**	999	0.021
Donor population, Control meadow	1.6338	**0.001**	999	0.006
Donor population, Dead matte	No test, df = 0			
BESE element, Coconut fiber mat	0.94506	0.557	999	0.553
BESE element, Iron staple	0.95653	0.515	999	0.532
BESE element, Control meadow	1.2696	**0.037**	999	0.062
BESE element, Dead matte	No test, df = 0			
Coconut fiber mat, Iron staple	0.81167	0.922	999	0.83
Coconut fiber mat, Control meadow	1.2506	**0.043**	999	0.056
Coconut fiber mat, Dead matte	No test, df = 0			
Iron staple, Control meadow	1.0618	0.257	998	0.272
Iron staple, Dead matte	No test, df = 0			
Control meadow, Dead matte	1.2	0.05	997	0.085

Table S3.23. Results of the PERMANOVA pair-wise test for the interaction "Sample natureXTransplantation method" comparing the different levels of the factor "Sample nature" for bacterial community structure. Bold face values are significant at $p<0.05$.

Within level 'leaf' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Donor population, BESE element	1.0655	0.229	998	0.311
Donor population, Coconut fiber mat	1.0372	0.297	988	0.375
Donor population, Iron staple	0.91454	0.589	997	0.555
Donor population, Control meadow	0.99091	0.35	989	0.408
BESE element, Coconut fiber mat	0.8549	0.842	995	0.708
BESE element, Iron staple	0.86494	0.752	998	0.694
BESE element, Control meadow	0.90681	0.632	997	0.548
Coconut fiber mat, Iron staple	0.87021	0.835	996	0.654
Coconut fiber mat, Control meadow	0.95491	0.483	982	0.512
Iron staple, Control meadow	0.84911	0.802	997	0.71

Within level 'root' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Donor population, BESE element	1.2606	0.098	406	0.159
Donor population, Coconut fiber mat	1.6963	**0.002**	838	0.015
Donor population, Iron staple	1.6521	**0.003**	972	0.013
Donor population, Control meadow	1.9317	**0.001**	995	0.002
BESE element, Coconut fiber mat	0.94844	0.479	842	0.48
BESE element, Iron staple	0.97677	0.457	966	0.461
BESE element, Control meadow	1.3644	**0.03**	995	0.053
Coconut fiber mat, Iron staple	0.85706	0.754	994	0.652
Coconut fiber mat, Control meadow	1.3591	**0.034**	998	0.061
Iron staple, Control meadow	1.1337	0.193	999	0.213

Within level 'sediment' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Control meadow, Dead matte	1.3858	**0.004**	998	0.016

Within level 'water' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Control meadow, Dead matte	1.7753	**0.001**	966	0.004

Table S3.24. Results of the PERMANOVA main test for ASVs structure for the factors "Sample nature", "Sample origin", and their interaction "Sample natureXSample origin". Bold face values are significant at p<0.05.

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms	P(MC)
Sample nature	3	98535	32845	10.43	**0.001**	997	0.001
Sample origin	5	25209	5041.7	1.6011	**0.001**	995	0.001
Sample naturexSample origin	4	18603	4650.7	1.4769	**0.011**	997	0.011
Res	137	4,31E+05	3149				
Total	149	6,23E+05					

Table S3.25. Results of the PERMANOVA pair-wise test within the factor "Sample nature" for ASVs structure. Bold face values are significant at p<0.05.

Groups	t	P(perm)	Unique perms	P(MC)
Leaf, root	2.5486	**0.001**	997	0.001
Leaf, sediment	2.2994	**0.001**	999	0.001
Leaf, water	2.6951	**0.001**	998	0.001
Root, sediment	2.5039	**0.001**	995	0.001
Root, water	2.8594	**0.001**	999	0.001
Sediment, water	4.8673	**0.001**	997	0.001

Table S3.26. Results of the PERMANOVA pair-wise test within the factor "Sample origin" for ASVs structure. Bold face values are significant at p<0.05.

Groups	t	P(perm)	Unique perms	P(MC)
Donor pop. – IC, Donor pop. - SF	1.1524	0.213	997	0.228
Donor pop. – IC, Storm-fragment	1.089	0.208	997	0.28
Donor pop. – IC, Intermatte cutting	1.0664	0.242	997	0.271
Donor pop. – IC, Control meadow	1.148	0.126	998	0.172
Donor pop. – IC, Dead matte	No test, df = 0			
Donor pop. – SF, Storm-fragment	1.58	**0.002**	997	0.005
Donor pop. – SF, Intermatte cutting	1.4537	**0.01**	998	0.02
Donor pop. – SF, Control meadow	1.6034	**0.003**	997	0.005
Donor pop. – SF, Dead matte	No test, df = 0			
Storm-fragment, Intermatte cutting	0.83442	0.898	997	0.808
Storm-fragment, Control meadow	1.1891	0.09	996	0.09
Storm-fragment, Dead matte	No test, df = 0			
Intermatte cutting, Control meadow	1.223	0.055	997	0.081
Intermatte cutting, Dead matte	No test, df = 0			
Control meadow, Dead matte	1.2	0.055	999	0.069

Table S3.27. Results of the PERMANOVA pair-wise test for the interaction "Sample natureXSample origin" comparing the different levels of the factor "Sample nature" for bacterial community structure. Bold face values are significant at p<0.05.

Within level 'leaf' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Donor pop. – IC, Donor pop. - SF	1.1511	0.17	126	0.239
Donor pop. – IC, Storm-fragment	1.1028	0.22	977	0.27
Donor pop. – IC, Intermatte cutting	1.1083	0.2	989	0.27
Donor pop. – IC, Control meadow	1.0797	0.23	841	0.298
Donor pop. – SF, Storm-fragment	0.98465	0.407	977	0.433
Donor pop. – SF, Intermatte cutting	1.1177	0.162	981	0.216
Donor pop. – SF, Control meadow	0.97784	0.337	858	0.452
Storm-fragment, Intermatte cutting	0.92594	0.599	999	0.58

Storm-fragment, Control meadow	0.87371	0.706	998	0.663
Intermatte cutting, Control meadow	0.97113	0.402	997	0.429

Within level 'root' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Donor pop. – SF, Storm-fragment	1.7716	**0.001**	983	0.009
Donor pop. – SF, Intermatte cutting	1.4861	**0.016**	977	0.03
Donor pop. – SF, Control meadow	1.9317	**0.002**	994	0.002
Storm-fragment, Intermatte cutting	0.9217	0.577	999	0.586
Storm-fragment, Control meadow	1.3714	**0.04**	998	0.046
Intermatte cutting, Control meadow	1.34	**0.042**	999	0.058

Within level 'sediment' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Control meadow, Dead matte	1.3858	**0.003**	994	0.009

Within level 'water' of factor 'Sample nature'

Groups	t	P(perm)	Unique perms	P(MC)
Control meadow, Dead matte	1.7753	**0.001**	966	0.005

Table S3.28. Taxonomical levels of the most differentially abundant ASVs from seagrass roots according to the experimental factor 'transplantation method' from the Linear Discriminant Analysis (LDA) Effect Size (LEfSe).

ASV name	Kingdom	Phylum	Class	Order	Family	Genus
ASV19	Bacteria	Proteobacteria	Gammaproteobacteria			
ASV83	Bacteria	Proteobacteria	Gammaproteobacteria	Enterobacterales	Alteromonadaceae	Alteromonas
ASV23	Bacteria	Proteobacteria	Gammaproteobacteria			
ASV27	Bacteria	Proteobacteria	Gammaproteobacteria	Chromatiales	Sedimenticolaceae	Candidatus_Thiodiazotropha
ASV126	Bacteria	Proteobacteria	Gammaproteobacteria	Pseudomonadales		
ASV113	Bacteria	Proteobacteria	Alphaproteobacteria	Rhodobacterales	Rhodobacteraceae	Yoonia-Loktanella
ASV167	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	Aquimarina
ASV12	Bacteria	Proteobacteria	Gammaproteobacteria	Pseudomonadales	Marinomonadaceae	Marinomonas
ASV117	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	Aquimarina
ASV169	Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Stappiaceae	Labrenzia
ASV155	Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Stappiaceae	Labrenzia
ASV79	Bacteria	Desulfobacterota	Desulfobacteria	Desulfobacterales	Desulfosarcinaceae	
ASV80	Bacteria	Proteobacteria	Alphaproteobacteria	Thalassobaculales	Nisaeaceae	Nisaea
ASV292	Bacteria	Proteobacteria	Gammaproteobacteria	Enterobacterales	Alteromonadaceae	Paraglaciecola
ASV129	Bacteria	Proteobacteria	Gammaproteobacteria	Arenicellales	Arenicellaceae	Arenicella
ASV199	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	
ASV228	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	
ASV184	Bacteria	Proteobacteria	Gammaproteobacteria	pltb-vmat-80		
ASV88	Bacteria	Proteobacteria	Alphaproteobacteria			
ASV335	Bacteria	Proteobacteria	Alphaproteobacteria			

Table S3.29. Taxonomical levels of the most differentially abundant ASVs from seagrass roots according to the experimental factor 'sample origin' from the Linear Discriminant Analysis (LDA) Effect Size (LEfSe).

ASV name	Kingdom	Phylum	Class	Order	Family	Genus
ASV19	Bacteria	Proteobacteria	Gammaproteobacteria			
ASV83	Bacteria	Proteobacteria	Gammaproteobacteria	Enterobacterales	Alteromonadaceae	Alteromonas
ASV23	Bacteria	Proteobacteria	Gammaproteobacteria			
ASV27	Bacteria	Proteobacteria	Gammaproteobacteria	Chromatiales	Sedimenticolaceae	Candidatus_Thiodiazotropha
ASV113	Bacteria	Proteobacteria	Alphaproteobacteria	Rhodobacterales	Rhodobacteraceae	Yoonia-Loktanella
ASV167	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	Aquimarina
ASV12	Bacteria	Proteobacteria	Gammaproteobacteria	Pseudomonadales	Marinomonadaceae	Marinomonas
ASV117	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	Aquimarina
ASV169	Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Stappiaceae	Labrenzia
ASV155	Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Stappiaceae	Labrenzia
ASV79	Bacteria	Desulfobacterota	Desulfobacteria	Desulfobacterales	Desulfosarcinaceae	
ASV80	Bacteria	Proteobacteria	Alphaproteobacteria	Thalassobaculales	Nisaeaceae	Nisaea
ASV292	Bacteria	Proteobacteria	Gammaproteobacteria	Enterobacterales	Alteromonadaceae	Paraglaciecola
ASV129	Bacteria	Proteobacteria	Gammaproteobacteria	Arenicellales	Arenicellaceae	Arenicella
ASV199	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	
ASV228	Bacteria	Bacteroidota	Bacteroidia	Flavobacteriales	Flavobacteriaceae	
ASV88	Bacteria	Proteobacteria	Alphaproteobacteria			
ASV378	Bacteria	Proteobacteria	Alphaproteobacteria	Rhodospirillales	Thalassospiraceae	Thalassospira
ASV281	Bacteria	Proteobacteria	Alphaproteobacteria	Rhizobiales	Stappiaceae	Labrenzia
ASV396	Bacteria	Proteobacteria	Alphaproteobacteria	Kordiimonadales	Kordiimonadaceae	Kordiimonas

Table S3.30. *PERMANOVA results on the effect of donor source (i.e. storm-fragment, intermatte cutting and control meadow), months post transplanting and their interaction on P. oceanica physiological and biochemical individual traits. Bold face values are significant at $p<0.05$.*

Factor	df	Variable	pseudo-F	P(perm)
Donor source	2	Fv/Fm	14.871	**0.001**
Months	6		13.864	**0.001**
Donor source *Months	12		4.2701	**0.001**
Donor source	2	α	0.40321	0.683
Months	6		6.6243	**0.001**
Donor source *Months	12		2.6502	**0.006**
Donor source	2	rETRmax	43.228	**0.001**
Months	6		106.68	**0.001**
Donor source *Months	12		7.803	**0.001**
Donor source	2	Ek	9.4408	**0.003**
Months	6		34.214	**0.001**
Donor source *Months	12		2.5689	**0.021**
Donor source	2	C	18.04	**0.001**
Months	6		70.143	**0.001**
Donor source *Months	12		1.6351	0.101
Donor source	2	N	28.551	**0.001**
Months	6		229.58	**0.001**
Donor source *Months	12		2.0953	**0.016**
Donor source	2	P	10.968	**0.001**
Months	6		29.036	**0.001**
Donor source *Months	12		1.6127	0.114
Donor source	2	S	0.77485	0.438
Months	6		570.81	**0.001**
Donor source *Months	12		3.6795	**0.001**
Donor source	2	C:N	16.222	**0.001**
Months	6		191.11	**0.001**
Donor source *Months	12		1.7358	0.053
Donor source	2	C:P	12.71	**0.001**
Months	6		24.405	**0.001**
Donor source *Months	12		1.8542	0.05
Donor source	2	N:P	20.821	**0.001**
Months	6		9.4719	**0.001**
Donor source *Months	12		1.9607	0.058
Donor source	2	TCR	7.7656	**0.001**
Months	3		20.809	**0.001**

Donor source *Months	6		3.2565	**0.007**
Donor source	2	Sucrose	3.5606	**0.036**
Months	3		80.644	**0.001**
Donor source *Months	6		0.46566	0.829
Donor source	2	Starch	7.113	**0.003**
Months	3		2.7984	0.073
Donor source *Months	6		1.4722	0.25

Table S3.31. *Comparison of means (pair-wise tests) of P. oceanica physiological and biochemical traits for the interaction of donor source (s = storm-fragment, i = intermatte cutting, c = control meadows) and months post transplanting (0, 3, 12, 15, 24, 27, 36).*

	Pair-wise tests of donor source * months post transplanting						
	0	3	12	15	24	27	36
Fv/Fm	c, s > i	c = s = i	c > s, i	c > s, i	c = s =i	c > s, i	c > s, i
α	i > s, c	c = s =i	c > s, i	c = s = i	c = s = i	i > c ; i = s ; c = s	c = s = i
rETRmax	i > s, c	c = s =i	i, s > c	c = s =i	i, s > c	c = s =i	c = s =i
Ek	i > s, c	c = s =i	i, s > c	c = s =i	i, s > c	c = s =i	c = s =i
C	c > s > i	c > i ; i = s ; c = s	c > s, i	c > s, i	c > s; i =s ; c = i	c = s =i	c =s =i
N	c = s = i	c = s =i	c, i > s	c > i > s	c, i > s	c > s ; i =s ; c = i	c > s, i
P	i, s > c	i, s > c	i > c, s	i > s ; i = c ; s =c	c = s = i	c = s =i	c > s ; i = s ; c =i
S	s > c, i	c = s = i	c = s = i	c > s, i	c = s = i	i, s > c	c = s = i
C:N	s > i ; c = i ; c = s	c = s = i	s > i, c	s > i, c	s > i ; c = i ; c = s	s > i ; c = i ; c = s	i, s > c
C:P	c > s > i	c > s, i	c > s, i	c > i ; i = s ; c = s	c > s ; s = i ; c = i		
N:P	c > i, s	c > i, s	c > i, s	c > i, s	c > s ; i = s ; c = i	c > i, s	s > i ; c = s ; c = i
TCR	i > s, c	NA	c = s =i	NA	c = s =i	NA	c > s, i
Sucrose	c = s =i	NA	c = s =i	NA	c >s, i	NA	c >s, i
Starch	i > s, c	NA	c = s =i	NA	c = s =i	NA	c >s, i

Table S3.32. *PERMANOVA results on the effect of donor source (i.e. storm-fragment, intermatte cutting and control meadow), months post transplanting and their interaction on P. oceanica physiological and biochemical traits' structure. Bold face values are significant at p<0.05.*

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms	P(MC)
Donor source	2	2295,2	1147,6	6,6672	0,001	999	**0,001**
Months	3	11814	3937,8	22,877	0,001	999	**0,001**
Donor source*Months	6	3178,6	529,76	3,0777	0,001	998	**0,001**
Res	161	27713	172,13				
Total	172	46061					

Table S3.33. *Results of the PERMANOVA pair-wise test for the interaction "Donor source*Months post transplanting" comparing the different levels of the factor "Months post transplanting" for P. oceanica physiological and biochemical traits' structure. Bold face values are significant at p<0.05.*

Within level '0' of factor 'Months post transplanting'

Groups	t	P(perm)	Unique perms	P(MC)
Storm-fragment, Intermatte cutting	2,4433	0,001	739	**0,003**
Storm-fragment, Control meadow	2,3689	0,006	996	**0,005**
Intermatte cutting, Control meadow	5,3424	0,001	996	**0,001**

Within level '12' of factor 'Months post transplanting'

Groups	t	P(perm)	Unique perms	P(MC)
Storm-fragment, Intermatte cutting	0,74853	0,617	999	0,622
Storm-fragment, Control meadow	2,21	0,01	998	**0,01**
Intermatte cutting, Control meadow	2,2427	0,003	999	**0,009**

Within level '24' of factor 'Months post transplanting'

Groups	t	P(perm)	Unique perms	P(MC)
Storm-fragment, Intermatte cutting	1,1436	0,262	999	0,255
Storm-fragment, Control meadow	1,8793	0,027	997	**0,029**
Intermatte cutting, Control meadow	1,0878	0,328	999	0,295

Within level '36' of factor 'Months post transplanting'

Groups	t	P(perm)	Unique perms	P(MC)
Storm-fragment, Intermatte cutting	2,1575	0,01	999	**0,01**
Storm-fragment, Control meadow	3,2078	0,002	999	**0,001**
Intermatte cutting, Control meadow	2,0058	0,01	997	**0,016**

Table S3.34. *Results of similarity percentages analysis (SIMPER), showing the contribution of each physiological and biochemical trait to the average dissimilarity between donor sources (s = storm-fragment, i = intermatte cutting, c = control meadows) at 0, 12, 24 and 36 months post transplanting.*

Before transplantation (0 months)						**12 months after transplanting**					
s vs i		c vs s		c vs i		c vs i		c vs s		c vs i	
Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)
C:P	30.2	C :P	46.4	C:P	42.1	C:P	43.5	C:P	51.8	C:P	48.5
Ek	24.5	TCR	12.3	Ek	19.4	TCR	17.3	TCR	14.2	TCR	15.9
rETRmax	15.4	Ek	11.8	rETRmax	12.9	Ek	11.5	Sucrose	11.2	Ek	10.3
TCR	11.6	Sucrose	10.3	TCR	8.7	Sucrose	10.5	Ek	8.3	Sucrose	9.7
Sucrose	9.1	Starch	9.1	Starch	8.6	Starch	8.6	Starch	5.1	Starch	7.6
Starch	6.4	rETRmax	5.6	Sucrose	4.8	rETRmax	4.5	rETRmax	3.6	rETRmax	3.8
N:P	1.2	N:P	1.9	N:P	1.8	C:N	1.9	N:P	2.5	N:P	2.1
C:N	0.9	C:N	1.4	C:N	0.8	N:P	1.2	C:N	1.9	C:N	1.1
C	0.5	C	1.0	C	0.7	C	1.1	C	1.2	C	0.8
N	0.1	N	0.1	N	0.1	N	0.1	N	0.1	N	0.1
Fv/Fm	<0.1	S	<0.1	Fv/Fm	<0.1	S	<0.1	S	<0.1	Alpha	<0.1
S	<0.1	Alpha	<0.1	Alpha	<0.1	Alpha	<0.1	Alpha	<0.1	S	<0.1
Alpha	<0.1	Fv/Fm	<0.1	S	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1
P	<0.1	P	<0.1	P	<0.1	P	<0.1	P	<0.1	P	<0.1

24 months after transplanting						**36 months after transplanting**					
s vs i		c vs s		c vs i		s vs i		c vs s		c vs i	
Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)	Trait	Contribution to dissimilarity (%)
C:P	57.6	C:P	48.8	C:P	55.4	Ek	34.9	C:P	30.4	Ek	32.8
TCR	13.6	TCR	17.0	TCR	14.8	C:P	29.6	TCR	23.7	C:P	21.7
Starch	11.6	Starch	14.2	Starch	12.5	TCR	13.0	Starch	18.6	TCR	18.4
Ek	5.8	Ek	7.0	Ek	6.5	Starch	10.2	Ek	14.7	Starch	14.2
C:N	2.7	Sucrose	3.4	N:P	2.9	rETRmax	5.7	Sucrose	5.7	Sucrose	4.8

N:P	2.6	N:P	2.8	Sucrose	2.8	Sucrose	3.3	rETRmax	3.9	rETRmax	4.8
rETRmax	2.5	C:N	2.8	rETRmax	2.4	C:N	1.7	C:N	1.5	C:N	1.7
Sucrose	2.4	rETRmax	2.6	C:N	1.7	N:P	1.0	N:P	0.8	N:P	0.9
C	0.9	C	1.0	C	0.7	C	0.4	C	0.5	C	0.5
N	0.1	N	0.2	N	0.1	N	0.1	N	0.1	N	0.1
S	0.1	S	0.1	S	<0.1	Alpha	<0.1	S	<0.1	Alpha	<0.1
alpha	<0.1	Alpha	<0.1	Alpha	<0.1	S	<0.1	Alpha	<0.1	S	<0.1
Fv/Fm	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1	Fv/Fm	<0.1
P	<0.1	P	<0.1	P	<0.1	P	<0.1	P	<0.1	P	<0.1

Bibliographie

A

Abadie, A., & Gobert, S. (2008). La plante de la mer du milieu. La posidonie. Éditions du Livre en Papier. ISBN 978-2-8083-0449-8.

Abadie, A., Gobert, S., Bonacorsi, M., Lejeune, P., Pergent, G., & Pergent-Martini, C. (2015). Marine space ecology and seagrasses. Does patch type matter in *Posidonia oceanica* seascapes? *Ecological Indicators*, 57, 435–446.

Abadie, A., Lejeune, P., Pergent, G., & Gobert, S. (2016). From mechanical to chemical impact of anchoring in seagrasses: The premises of anthropogenic patch generation in *Posidonia oceanica* meadows. *Marine Pollution Bulletin*, 109(1), 61–71.

Abadie, A., Borges, A.V., Champenois, W., & Gobert, S. (2017). Natural patches in *Posidonia oceanica* meadows: the seasonal biogeochemical pore water characteristics of two edge types. *Marine Biology*, 164, 166.

Abadie, A., Pace, M., Gobert, S., & Borg, J. A. (2018). Seascape ecology in *Posidonia oceanica* seagrass meadows: Linking structure and ecological processes for management. *Ecological Indicators*, 87, 1–13.

Abadie, A., Richir, J., Lejeune, P., Leduc, M., & Gobert, S. (2019). Structural changes of seagrass seascapes driven by natural and anthropogenic factors: A multidisciplinary approach. *Frontiers in Ecology and Evolution*, 7, 1–13.

Abadie, A., Boissery, P., & Viala, C. (2020). Study of the *Posidonia oceanica* meadow's upper limit with georeferenced photogrammetry. *Life and Environment*, 70(3–4), 297-301.

Abadie, A., Viala, C., Marre, G., & Boissery, P. (2022). La photogrammétrie sous-marine. Application au suivi des herbiers de posidonie. *Seaviews Publication*, 108p.

Ackerman, J. D., & Okubo, A. (1993). Reduced mixing in a marine macrophyte canopy. *Functional Ecology*, 7(3), 305-309.

Adolph, E.F. (1956). General and specific characteristics of physiological adaptations. *American Journal of Physiology*, 184, 18–28.

Afgan, E., Baker, D., Batut, B., van den Beek, M., Bouvier, D., Čech, M., Chilton, J., Clements, D., Coraor, N., & Grüning, B. A. (2018). The Galaxy platform for accessible, reproducible and collaborative biomedical analyses: 2018 update. *Nucleic Acids Research*, 46(1), 537–544.

Agisoft LLC (2022). Agisoft Metashape User Manual - Professional Edition, Version 1.8. Agisoft LLC.

Aires, T., Serrão, E. A., & Engelen, A. H. (2016). Host and environmental specificity in bacterial communities associated to two highly invasive marine species (genus *Asparagopsis*). *Frontiers in Microbiology*, 7.

Aires, T., Stuij, T.M., Muyzer, G., Serrão, E.A., & Engelen, A.H. (2021) Characterization and comparison of bacterial communities of an invasive and two native Caribbean seagrass species sheds light on the possible influence of the Microbiome on invasive mechanisms. *Frontiers in Microbiology*, 12.

Airoldi, L. & Beck, M.W. (2007). Loss, status and trends for coastal marine habitats of Europe. *Oceanography and Marine Biology - An Annual Review*, 45, 345–405.

Alagna, A., D'Anna, G., Musco, L., Vega Fernández, T., Gresta, M., Pierozzi, N., & Badalamenti, F. (2019). Taking advantage of seagrass recovery potential to develop novel and effective meadow rehabilitation methods. *Marine Pollution Bulletin*, 149, 110578.

Alcoverro, T., Duarte, C.M., and Romero, J. (1995). Annual growth dynamics of *Posidonia oceanica*: contribution of large-scale versus local factors to seasonality. *Marine Ecology Progress Series*, 120, 203-210.

Alcoverro, T., Manzanera, M., & Romero, J. (2000). Nutrient mass balance of the seagrass *Posidonia oceanica*: the importance of nutrient retranslocation. *Marine Ecology Progress Series*, 194, 13-21.

Alcoverro, T., Manzanera, M., & Romero, J. (2001). Annual Metabolic Carbon Balance of the Seagrass *Posidonia oceanica*: The Importance of Carbohydrate Reserves. *Marine Ecology Progress Series*, 211,105–116.

ALF (2005). Fostering the land ethic through the legacy of Aldo Leopold. The Aldo Leopold Foundation. http://www.aldoleopold.org

Almela, E. D., Marbà, N., Álvarez, E., Santiago, R., Martínez, R., & Duarte, C. M. (2008). Patch dynamics of the Mediterranean seagrass *Posidonia oceanica*: Implications for recolonisation process. *Aquatic Botany*, 89(4), 397–403.

Anderson, M.J. (2001). A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, 26 (1).

Anderson, M.J., Gorley, R.N., & Clarke, K.R. (2008). PERMANOVA + for PRIMER: guide to software and statistical methods. Plymouth (UK): PRIMER-E.

Apostolaki, E. T., Caviglia, L., Santinelli, V., Cundy, A. B., Tramati, C. D., Mazzola, A., & Vizzini, S. (2022). The importance of dead seagrass (*Posidonia oceanica*) matte as a biogeochemical sink. *Frontiers in Marine Science*, 9.

Atkinson, J., & Bonser, S. P. (2020). "Active" and "passive" ecological restoration strategies in meta-analysis. *Restoration Ecology*, 28(5), 1032–1035.

Averill, C., Anthony, M.A., Baldrian, P., Finkbeiner, F., van den Hoogen, J., Kiers, T., Kohout, P., Hirt, E., Reuben Smith, G., & Crowther, T.W. (2022) Defending Earth's terrestrial microbiome. *Nature Microbiology*, 7, 1717–1725.

B

Bacci, T., & La Porta, B. (2021). Manual of techniques and procedures for the transplantation of *Posidonia oceanica*. LIFE SEPOSSO (LIFE 16 GIE/IT/000761), Rome, Italy.

Bacci, T., Scardi, M., Tomasello, A., Valiante, L.M., Piazzi, L., Calvo, S., Badalamenti, F., Di Nuzzo, F., Raimondi, V., Assenzo M, Cecchi, E., Penna, M., Bertasi, F., Piazzi, A., & La Porta, B. (2024). Long-term response of *Posidonia oceanica* meadow restoration at the population and plant level: implications for management decisions. *Restoration Ecology*, 33(3), e14360.

Bacon, C. W., & White, J. F. (2016). Functions, mechanisms and regulation of endophytic and epiphytic microbial communities of plants. *Symbiosis*, 68(1–3), 87–98.

Badalamenti, F., Alagna, A., D'Anna, G., Terlizzi, A., & Di Carlo, G. (2011). The impact of dredge-fill on *Posidonia oceanica* seagrass meadows: Regression and patterns of recovery. *Marine Pollution Bulletin*, 62(3), 483–489.

Balata, D., Nesti, U., Piazzi, L. & Cinelli, F. (2007). Patterns of spatial variability of seagrass epiphytes in the north-west Mediterranean Sea. *Marine Biology* 151, 2025-2035.

Balestri, E., & Cinelli, F. (2003). Sexual reproductive success in *Posidonia oceanica*. *Aquatic Botany*, 75.

Balestri, E. (2004). Flowering of the seagrass *Posidonia oceanica* in a north-western Mediterranean coastal area: Temporal and spatial variations. *Marine Biology*, 145(1), 61–68.

Balestri, E., & Lardicci, C. (2008). First evidence of a massive recruitment event in *Posidonia oceanica*: Spatial variation in first-year seedling abundance on a heterogeneous substrate. *Estuarine, Coastal and Shelf Science*, 76(3), 634–641.

Balestri, E., Vallerini, F., & Lardicci, C. (2011). Storm-generated fragments of the seagrass *Posidonia oceanica* from beach wrack - A potential source of transplants for restoration. *Biological Conservation*, 144(5), 1644–1654.

Balestri, E., de Battisti, D., Vallerini, F., & Lardicci, C. (2015). First evidence of root morphological and architectural variations in young *Posidonia oceanica* plants colonizing different substrate typologies. *Estuarine, Coastal, and Shelf Science*, 154, 205–213.

Balestri, E., Vallerini, F., & Lardicci, C. (2017). Recruitment and patch establishment by seed in the seagrass *Posidonia oceanica*: Importance and conservation implications. *Frontiers in Plant Science*, 8.

Barcelona, A., Colomer, J., Soler, M., Gracias, N., & Serra, T. (2021). Meadow fragmentation influences *Posidonia oceanica* density at the edge of nearby gaps. *Estuarine, Coastal and Shelf Science*, 249.

Barnaud, G. (1995). A l'interface de la pratique et de la théorie : l'écologie de la restauration. *Natures, Sciences Sociétés*, 3, 36-50.
Batista, B.D., & Singh, B.K. (2021). Realities and hopes in the application of microbial tools in agriculture. *Microbial Biotechnology*, 14(4), 1258–1268.

Bay D. (1978) Etude "in situ" de la production primaire d'un herbier de posidonies (*Posidonia oceanica* (L.) Delile) de la baie de Calvi-Corse. Thèse de doctorat. Université de Liège, Liège (Belgique).

Bayraktarov, E., Saunders, M. I., Abdullah, S., Mills, M., Beher, J., Possingham, H. P., Mumby, P. J., & Lovelock, C. E. (2016). The cost and feasibility of marine coastal restoration. *Ecological Applications*, 26(4), 1055–1074.

Beattie, G.A., Edlund, A., Esiobu, N., Gilbert, J., Nicolaisen, M.H., Jansson, J.K., Jensen, P., Keiluweit, M., Lennon, J.T., Martiny, J., Minnis, V.R., Newman, D., Peixoto, R., Schadt, C., & van der Meer, J. R. (2025). Soil microbiome interventions for carbon sequestration and climate mitigation. *Environmental Microbiology*, 10(1).

Beck, M.W., Heck, K.L., Able, K.W., Childers, D.L., Eggleston, D.B., Gillanders, B.M., Halpern, B., Hays, C.G., Hoshino, K., Minello, T.J., Orth, R.J., Sheridan, P.F., & Weinstein, M.P. (2021). The identification, conservation, and management of estuarine and marine nurseries for fish and invertebrates. *Bioscience*, 51(8), 633–641.

Bedini, R., Bedini, M., & Salvadori, E. (2020). A new transplanting method of *Posidonia oceanica* (Linnaeus) Delile, 1813 plants. In Monitoring of Mediterranean Coastal Areas. Problems and Measurement Techniques; Bonora, L., Carboni, D., de Vicenzi, M., Eds.; Firenze University Press: Florence, Italy; pp. 492–500.

Beheshti, K.M., Williams, S.L., Boyer, K.E., Endris, C., Clemons, A., Grimes, T., Wasson, K., & Hughes, B.B. (2021). Rapid enhancement of multiple ecosystem services following the restoration of a coastal foundation species. *Ecological Applications*, 31(8), e02466.

Belshe, E.F., Durako, M.J., & Blum, J.E. (2007). Photosynthetic rapid light curves (RLC) of *Thalassia testudinum* exhibit diurnal variation. *Journal of Experimental Marine Biology and Ecology*, 342(2), 253–268.

Belzunce, M., Navarro, R. M., & Rapoport, H. F. (2005). Seed and early plantlet structure of the Mediterranean seagrass *Posidonia oceanica*. *Aquatic Botany*, 82(4), 269-283.

Bergen, S. D., Bolton, S. M., & Fridley, J. L. (2001). Design principles for ecological engineering. *Ecological Engineering*, 18(2), 201–210.

Bergès, L., Roche, P. & Avon, C. (2010). Corridors écologiques et conservation de la biodiversité, intérêts et limites pour la mise en place de la Trame verte et bleue. *Sciences Eaux & Territoires*, 3, 34-39.

Bethoux, J. P., Gentili, B., Morin, P., Nicolas, E., Pierre, C., & Ruiz-Pino, D. (1999). The Mediterranean Sea: a miniature ocean for climatic and environmental studies and a key for the climatic functioning of the North Atlantic. *Progress in Oceanography*, 44(1–3), 131–146.

Bianchi, C. N., & Morri, C. (2000). Marine Biodiversity of the Mediterranean Sea: Situation, Problems and Prospects for Future Research. *Marine Pollution Bulletin*, 40(5), 367–376.

Bianchi, C. N., Morri, C., Chiantore, M., Montefalcone, M., Parravicini, V., & Rovere, A. (2012). Mediterranean Sea biodiversity between the legacy from the past and a future of change. In Life in the Mediterranean Sea: A Look at Habitat Changes (pp. 1–55).

Bockel, T., Marre, G., Delaruelle, G., Holon, F., Boissery, P., Blandin, A., Mouquet, N., & Deter, J. (2023). Anchoring pressure and the effectiveness of new management measures quantified using AIS data and a mobile application. *Marine Pollution Bulletin*, 195, 115511.

Bockel, T., Marre, G., Delaruelle, G., Agel, N., Boissery, P., Guilhaumon, F., Mouquet, N., Mouillot, D., Guilbert, A., & Deter, J. (2024). Early signals of *Posidonia oceanica* meadows recovery in a context of wastewater treatment improvements. *Marine Pollution Bulletin*, 201, 116193.

Bodenhausen, N., Horton, M.W., & Bergelson, J. (2013). Bacterial Communities Associated with the Leaves and the Roots of *Arabidopsis thaliana*. *PLoS ONE,* 8(2), e56329.

Bonacorsi, M., Pergent-Martini, C., Bréand, N., & Pergent, G. (2013). Is *Posidonia oceanica* regression a general feature in the mediterranean sea? *Mediterranean Marine Science*, 14(1), 193–203.

Bonamano, S., Piazzolla, D., Scanu, S., Mancini, E., Madonia, A., Piermattei, V., & Marcelli, M. (2021). Modelling approach for the evaluation of burial and erosion processes on *Posidonia oceanica* meadows. *Estuarine, Coastal and Shelf Science*, 254.

Bonito, G., Reynolds, H., Robeson, M. S., Nelson, J., Hodkinson, B. P., Tuskan, G., et al. (2014). Plant host and soil origin influence fungal and bacterial assemblages in the roots of woody plants. *Molecular Ecology*, 23(13), 3356–3370.

Blignaut, J., Esler, K.J., de Wit, M.P., Le Maitre, D., Milton, S.J., & Aronson, J. (2013). Establishing the links between economic development and the restoration of natural capital. *Current Opinion in Environmental Sustainability*, 5(1), 94–101.

Bolyen, E., Rideout, J.R., Dillon, M.R., Bokulich, N.A., Abnet, C.C., Al-Ghalith, G.A., Alexander, H., Alm, E.J., Arumugam, M., Asnicar, F., Bai, Y., Bisanz, J.E., Bittinger, K., Brejnrod, A., Brislawn, C.J., Brown, C.T., Callahan, B.J., Caraballo-Rodríguez, A.M., Chase, J., ... Caporaso, J.G. (2019). Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2. *Nature Biotechnology*, 37(8), 852–857.

Bonito, G., Reynolds, H., Robeson, M.S., Nelson, J., Hodkinson, B.P., Tuskan G., Schadt, C.W., & Vilgalys, R. (2014). Plant host and soil origin influence fungal and bacterial assemblages in the roots of woody plants. *Molecular Ecology*, 23(13), 3356-3370.

Borg, J.A., Attrill, M. J., Rowden, A. A., Schembri, P. J., & Jones, M. B. (2005). Architectural characteristics of two bed types of the seagrass *Posidonia oceanica* over different spatial scales. *Estuarine, Coastal and Shelf Science*, 62(4), 667–678.

Borsje, B. W., van Wesenbeeck, B. K., Dekker, F., Paalvast, P., Bouma, T. J., van Katwijk, M. M., & de Vries, M. B. (2011). How ecological engineering can serve in coastal protection. *Ecological Engineering*, 37(2), 113–122.

Boudouresque, C.F. & Meinesz, A. (1982) Découverte de l'herbier de Posidonies. *Cahier du Parc national de Port-Cros*, 4, 1–79.

Boudouresque, C.F., Bianconi, C.H., Meinesz, A. (1990). Live *Posidonia oceanica* in a coraligenous algal bank at Sulana bay, Corsica. *Rapport de la Commission Internationale pour l'Exploration Scientifique de la mer Méditerranée*, 32, 11 p.

Boudouresque, C.F., Charbonnel, E., Meinesz, A., Pergent, G., Pergent-Martini, C., Cadiou, G., Bertrandy, M.C., Foret, P., Ragazzi, M., Rico-Raimondino, V., (2000). A monitoring network based on the seagrass *Posidonia oceanica* in the nortwestern Mediterranean Sea. *Biologia Marina Mediterranea*, 7(2), 328-331.

Boudouresque, C.F. (2001). La restauration des écosystèmes à phanérogames marines. In Restauration des Ecosystèmes Côtiers; Drévès, L., Chaussepied, M., Eds.; IFREMER: Paris, France; Volume 29, pp. 65–85

Boudouresque, C.F., & Verlaque, M. (2005). Nature conservation, Marine Protected Areas, sustainable development and the flow of invasive species to the Mediterranean Sea. *Scientific Reports Port-Cros National Park*, Fr, 21, 29–54.

Boudouresque, C.F., Bernard, G., Bonhomme, P., Charbonnel, E., Diviacco, G., Meinesz, A., Pergent, G., Pergent-Martini, C., Ruitton, S., & Tunesi, L. (2006). Préservation et conservation des herbiers à *Posidonia oceanica*. Ramoge pub, Marseille, France. pp. 1–202. ISBN: 2-905540-30-3.

Boudouresque, C.F., Bernard, G., Pergent, G., Shili, A. & Verlaque, M. (2009). Regression of Mediterranean seagrasses caused by natural processes and anthropogenic disturbances and stress: a critical review. *Botanica Marina*, 52, 395-418.

Boudouresque, C.F., Bernard, G., Bonhomme, P., Charbonnel, E., Diviacco, G., Meinesz, A., Pergent, G., Pergent-Martini, C., Ruitton, S., & Tunesi, L. (2012). Protection and conservation of *Posidonia oceanica* meadows. RAMOGE and RAC/SPA publisher, Tunis: 1-202.

Boudouresque, C.F., Pergent, G., Pergent-Martini, C., Ruitton, S., Thibaut, T. & Verlaque, M. (2016). The necromass of the *Posidonia oceanica* seagrass meadow: fate, role, ecosystem services and vulnerability. *Hydrobiologia*, 781, 25–42.

Boudouresque, C. F., Blanfuné, A., Pergent, G., & Thibaut, T. (2021). Restoration of seagrass meadows in the Mediterranean sea: a critical review of effectiveness and ethical issues. *Water*, 13(8), 1034.

Boulenger, A., Roberty, S., Lopez Velosa, M.M., Marengo, M., & Gobert, S. (2024). The use of photo-biological parameters to assess the establishment success of

Posidonia oceanica cuttings after transplantation. *Water*, 16(12), 1702.

Boulenger, A., Chapeyroux, J., Fullgrabe, L., Marengo, M., & Gobert, S. (2025). Assessing *Posidonia oceanica* recolonisation dynamics for effective restoration designs in degraded anchoring sites. *Marine Pollution Bulletin*, 216, 117960.

Boulenger, A., Aires, T., Engelen, A.H., Muyzer, G., Marengo, M., & Gobert, S. (2025b). Microbiome matters: how transplantation methods and donor origins shape the successful restoration of the seagrass *Posidonia oceanica*. *Environmental Microbiome*, 20(99).

Boulenger, A., Marengo, M., Boissery, P., & Gobert, S. (in review). Comparative assessment of transplantation methods and donor sources for the restoration of *Posidonia oceanica* meadows. *Science of the Total Environment*.

Brockerhoff, E.G., Jactel, H., Parrotta, J.A., Quine, C.P., & Sayer, J. (2008). Plantation forests and biodiversity: oxymoron or opportunity? *Biodiversity Conservation*, 17, 925–951.

Brodersen, K.E., Mosshammer, M., Bittner, M.J., Hallstrøm, S., Santner, J., Riemann, L., & Kühl, M. (2024). Seagrass-mediated rhizosphere redox gradients are linked with ammonium accumulation driven by diazotrophs. *Microbiology Spectrum*, 12(4).

Brodersen, K.E., Siboni, N., Nielsen, D.A., Pernice, M., Ralph, P.J., Seymour, J., & Kühl, M. (2018). Seagrass rhizosphere microenvironment alters plant-associated microbial community composition. *Environmental Microbiology*, 20(8), 2854–2864.

Bucher, L.M. (2025). Integrating climate change and anthropogenic pressures in regional-scale site selection for *Posidonia oceanica* restoration in Corsica . Master Thesis. University of Liege, Liege, Belgium.

Buckley, D.H., Huangyutitham, V., Hsu, S.-F., & Nelson, T.A. (2007). Stable isotope probing with $^{15}N_2$ reveals novel non cultivated diazotrophs in soil. *Applied and Environmental Microbiology*, 73(10), 3196–3204.

Buia, M. C., & Mazzella, L. (1991). Reproductive phenology of the Mediterranean seagrasses *Posidonia oceanica* (L.) Delile, *Cymodocea nodosa* (Ucria) Aschers., and *Zostera noltii* Hornem. *Aquatic Botany*, 40.

Buia, M.C., Zupo, V., & Mazzella, L. (1992). Primary production and growth dynamics in *Posidonia oceanica*. *Marine Ecology*, 13(1), 2-16.

C

Callahan, B.J., McMurdie, P.J., Rosen, M. J., Han, A. W., Johnson, A. J. A., & Holmes, S. P. (2016). DADA2: High-resolution sample inference from Illumina amplicon data. *Nature Methods*, 13(7), 581–583.

Calleja, M.L., Marbà, N., & Duarte, C.M. (2007). The relationship between seagrass (*Posidonia oceanica*) decline and sulfide porewater concentration in carbonate sediments. *Estuarine, Coastal and Shelf Science*, 73(3–4), 583–588.

Calvo, E., Simó, R., Coma, R., Ribes, M., Pascual, J., Sabatés, A., Gili, J.M., & Pelejero, C. (2011). Effects of climate change on Mediterranean marine ecosystems: the case of the Catalan Sea. *Climate Research*, 50(1), 1–29.

Calvo, S., Calvo, R., Luzzu, F., Raimondi, V., Assenzo, M., Cassetti, F. P., & Tomasello, A. (2021). Performance assessment of *Posidonia oceanica* (L.) delile restoration experiment on dead matte twelve years after planting—structural and functional meadow features. *Water*, 13(5).

Campagne, C.S., Salles, J.M., Boissery, P., & Deter, J. (2014). The seagrass *Posidonia oceanica*: Ecosystem services identification and economic evaluation of goods and benefits. *Marine Pollution Bulletin*, 97(1–2), 391–400.

Campbell, M.L. (2000). An Empirical Evaluation of *Posidonia australis* (R. Br.) Hook f. Restoration in Western Australia: Development of a Decision-Based Restoration Framework. Ph.D. Thesis, Murdoch University, Perth, WA, Australia.

Cancemi, G., De Falco, G., & Pergent, G. (2003). Effects of organic matter input from a fish farming facility on a *Posidonia oceanica* meadow. *Estuarine, Coastal and Shelf Science*, 56(5–6), 961–968

Cardini, U., Marin-Guirao, L., Montilla, L.M., Marzocchi, U., Chiavarini, S., Rimauro, J., Quero, G.M., Petersen, J.M., Procaccini, G. (2022). Nested interactions between chemosynthetic lucinid bivalves and seagrass promote ecosystem functioning in contaminated sediments. *Frontiers in Plant Science*, 13.

Cappato, A., Canevello, S., Baggiani, B. (2011). Cruise and Recreational Boating in the Mediterranean. Istituto Internazionale delle Communicazioni, Genoa, Italy. Plan Bleu/UNEP MAP regional Activity Centre, 76 p.

Carreño, A. & Lloret, J. (2021). Environmental impacts of increasing leisure boating activity in Mediterranean coastal waters. *Ocean and Coastal Management*, 209, 105693.

Casoli, E., Mancini, G., Ventura, D., Belluscio, A., & Ardizzone, G. (2021). Double trouble: Synergy between habitat loss and the spread of the alien species *Caulerpa cylindracea* (Sonder) in three Mediterranean habitats. *Water*, 13(10), 1342.

Castejón-Silvo, I., & Terrados, J. (2021). Poor success of seagrass *Posidonia oceanica* transplanting in a meadow disturbed by power line burial. *Marine Environmental Research*, 170.

Castro-Fernández, J., Castejón-Silvo, I., Hinz, H., Escandell Westcott, A., & Terrados, J. (2025). Differences in habitat complexity between restored and natural seagrass meadows shape fish community structure. *Nature-Based Solutions*, 7, 100230.

Catucci, E., & Scardi, M. (2020). A Machine Learning approach to the assessment of the vulnerability of *Posidonia oceanica* meadows. *Ecological Indicators*, 108, 105744.

Caye, G., 1980. Sur la morphogénèse et le cycle végétatif de *Posidonia oceanica* (L.) Delile. Thèse de Doctorat. Université Aix-Marseille, Marseille (France).

Caye, G. & Meinesz, A. (1984). Observations sur la floraison et la fructification de *Posidonia oceanica* dans la baie de Villefranche et en Corse du Sud. In : Boudouresque, C.F., Jeudy de Grissac, A., Olivier, J. edits International Workshop on *Posidonia oceanica* beds, GIS Posidonie publ., Fr., 1:193-201

Ceccherelli, G., Oliva, S., Pinna, S., Piazzi, L., Procaccini, G., Marin-Guirao, L., Dattolo, E., Gallia, R., La Manna, G., Gennaro, P., Costa, M. M., Barrote, I., Silva, J., & Bulleri, F. (2018). Seagrass collapse due to synergistic stressors is not anticipated by phenological changes. *Oecologia*, 186(4), 1137–1152.

Chazdon, RL. (2003). Tropical forest recovery: legacies of human impact and natural disturbances. *Perspectives in Plant Ecology, Evolution and Systematics*, 6, 51–71.

Cheminée, A., Le Direach, L., Rouanet, E., Astruch, P., Goujard, A., Blanfuné, A., Bonhomme, D., Chassaing, L., Jouvenel, J., Ruitton, S., Thibaut, T., & Harmelin-Vivien M. (2021). All shallow coastal habitats matter as nurseries for Mediterranean juvenile fish. *Scientific Reports*, 11, 14631.

Chessa, L.A., Fresi, E. & Lorenzi, C. (1995). The state of health of a *Posidonia oceanica* meadow: study method. In: La *Posidonia oceanica*, a contribution to the preservation of a major Mediterranean marine ecosystem. *Revista Marittima*, 78-83.

Choi, Y. D. (2007). Restoration Ecology to the Future: A Call for New Paradigm. *Restoration Ecology*, 15(2), 351–353.

Christiaen, B., McDonald, A., Cebrian, J., & Ortmann, A. C. (2013). Response of the microbial community to environmental change during seagrass transplantation. *Aquatic Botany*, 109, 31–38.

Christianen, M.J.A., van Belzen, J., Herman, P.M.J., van Katwijk, M.M., Lamers, L.P.M., van Leent, P.J.M., & Bouma, T.J. (2013). Low-canopy seagrass beds still provide important coastal protection services. *PLoS ONE*, 8(5).

Clarke, K. R. (1993). Non-parametric multivariate analyses of changes in community structure. *Australian Journal of Ecology*, 18, 117–143.

Cohen-Shacham, E., Walters, G., Janzen, C. and Maginnis, S. (2016). Nature-based Solutions to address global societal challenges. Gland, Switzerland: IUCN. xiii + 97pp.http://dx.doi.org/10.2305/IUCN.CH.2016.13.en (accessed 13 April 2022)

Coll, M., Piroddi, C., Steenbeek, J., Kaschner, K., Lasram, F. B. R., Aguzzi, J., Ballesteros, E., Bianchi, C. N., Corbera, J., Dailianis, T., Danovaro, R., Estrada, M., Froglia, C., Galil, B. S., Gasol, J. M., Gertwage, R., Gil, J., Guilhaumon, F., Kesner-Reyes, K., … Voultsiadou, E. (2010). The biodiversity of the Mediterranean sea: estimates, patterns, and threats. *PLOS ONE*, 5(8), e11842.

Collier, C. J., Lavery, P. S., Ralph, P. J., & Masini, R. J. (2009). Shade-induced response and recovery of the seagrass *Posidonia sinuosa*. *Journal of Experimental Marine Biology and Ecology*, 370(1–2), 89–103.

Coma, R., Ribes, M., Serrano, E., Jiménez, E., Salat, J., & Pascual, J. (2009). Global warming-enhanced stratification and mass mortality events in the mediterranean. *Proceedings of the National Academy of Sciences of the United States of America*, 106(15), 6176–6181.

Conte, C., Rotini, A., Manfra, L., D'Andrea, M. M., Winters, G., & Migliore, L. (2021). The seagrass holobiont: what we know and what we still need to disclose for its Possible use as an ecological indicator. *Water*, 13(4), 406.

Cooke, S.J., & Suski, C.D. (2008). Ecological restoration and physiology: an overdue integration. *BioScience*, 58(10), 957–968.

Cooper G. (1982). Réimplantation de *Posidonia oceanica*. Protection des implants. *Bulletin d'Ecologie*, 13 (1), 65-73.

Corinaldesi, C., Bianchelli, S., Candela, M., Dell'Anno, A., Gambi, C., Rastelli, E., Varrella, S., & Danovaro, R. (2023). Microbiome-assisted restoration of degraded marine habitats: a new nature-based solution? *Frontiers in Marine Science*, 10.

Costa, M. M., Barrote, I., Silva, J., Olivé, I., Alexandre, A., Albano, S., & Santos, R. (2015). Epiphytes modulate *Posidonia oceanica* photosynthetic production, energetic balance, antioxidant mechanisms, and oxidative damage. *Frontiers in Marine Science*, 2, 111.

Costanza, R., D'Arge, R., De Groot, R., Farber, S., Grasso, M., Hannon, B., Limburg, K.,Naeem, S., O'Neill, R.V., Paruelo, J., Raskin, R.G., Sutton, P., van Den Belt, M. (1997). The value of the world's ecosystem services and natural capital. *Nature*, 387, 253–260.

Cozza, R., Rende, F., Ferrari, M., Bruno, L., Pacenza, M., Dattola, L., & Bitonti, M. B. (2019). Biomonitoring of *Posidonia oceanica* beds by a multiscale approach. *Aquatic Botany*, 156, 14–24.

Cruaud, P., Vigneron, A., Fradette, M.S., Charrette, S.J., Rodriguez, M.J., Dorea, C.C., & Culley, A.I. (2017). Open the sterivex™ casing: an easy and effective way to improve DNA extraction yields. *Limnology and Oceanography Methods*, 15(12), 1015-1020.

Crump, B. C., Wojahn, J. M., Tomas, F., & Mueller, R. S. (2018). Metatranscriptomics and amplicon sequencing reveal mutualisms in seagrass microbiomes. *Frontiers in Microbiology*, 9, 1–14.

Cúcio, C., Engelen, A. H., Costa, R., & Muyzer, G. (2016). Rhizosphere microbiomes of European + seagrasses are selected by the plant, but are not species specific. *Frontiers in Microbiology*, 7.

Cullen-Unsworth, L., & Unsworth, R. (2013). Seagrass meadows, ecosystem services, and sustainability. *Environment*, 55(3), 14–28.

Cummings, M. E., & Zimmerman, R. C. (2003). Light harvesting and the package effect in the seagrasses *Thalassia testudinum* Banks ex König and *Zostera marina* L.: optical constraints on photoacclimation. *Aquatic Botany*, 75(3), 261-274.

Cunha, A.H., Marbá, N.N., van Katwijk, M.M., Pickerell, C., Henriques, M., Bernard, G., Ferreira, M.A., Garcia, S., Garmendia, J.M., & Manent, P. (2012). Changing paradigms in seagrass restoration. *Restoration Ecology*, 20, 427–430.

Currie, L.A. (1999). Nomenclature in evaluation of analytical methods including detection and quantification capabilities (IUPAC Recommendations 1995). *Analytica Chimica Acta*, 391(2), 105–126.

D

Daleo, P., Fanjul, E., Casariego, A. M., Silliman, B. R., Bertness, M. D., & Iribarne, O. (2007). Ecosystem engineers activate mycorrhizal mutualism in salt marshes. *Ecology Letters*, 10(10), 902–908.

Danovaro, R., Aronson, J., Bianchelli, S., Boström, C., Chen, W., Cimino, R., Corinaldesi, C., Cortina-Segarra, J., D'Ambrosio, P., Gambi, C., Garrabou, J., Giorgetti, A., Grehan, A., Hannachi, A., Mangialajo, L., Morato, T., Orfanidis, S., Papadopoulou, N., Ramirez-Llodra, E., … Fraschetti, S. (2025). Assessing the success of marine ecosystem restoration using meta-analysis. *Nature Communications*, 16(1), 3062.

Dattolo, E., Ruocco, M., Brunet, C., Lorenti, M., Lauritano, C., D'Esposito, D., de Luca, P., Sanges, R., Mazzuca, S., & Procaccini, G. (2014). Response of the seagrass *Posidonia oceanica* to different light environments : Insights from a combined molecular and photo-physiological study. *Marine Environmental Research*, 101, 225–236.

Dattolo, E., Marín-Guirao, L., Ruiz, J. M., & Procaccini, G. (2017). Long-term acclimation to reciprocal light conditions suggests depth-related selection in the marine foundation species *Posidonia oceanica*. *Ecology and Evolution*, 7(4), 1148–1164.

Dauby, P., & Poulicek, M. (1995). Methods for removing epiphytes from seagrasses: SEM observations on treated leaves. *Aquatic Botany*, 52, 217-228.

de Boer, W.F. (2007) Seagrass–sediment interactions, positive feedbacks and critical thresholds for occurrence: a review. *Hydrobiologia*, 591, 5–24.

De Groot, R. S., Blignaut, J., Van Der Ploeg, S., Aronson, J., Elmqvist, T., & Farley, J. (2013). Benefits of Investing in Ecosystem

Restoration. *Conservation Biology*, 27(6), 1286–1293.

De Falco, G., Simeone, S. & Baroli, M. (2008). Management of beach-cast *Posidonia oceanica* seagrass on the island of Sardinia (Italy, western Mediterranean). *Journal of Coastal Research*, 24 (4), 69-75.

Delgado, O., Ruiz, J., Perez, M., Romero, J., & Ballesteros, E., (1999). Effects of fish farming on seagrass (*Posidonia oceanica*) in a Mediterranean bay: seagrass decline after organic loading cessation. *Oceanologia Acta*, 22(1), 109-117.

de los Santos, C. B., Krause-Jensen, D., Alcoverro, T., Marbà, N., Duarte, C. M., van Katwijk, M. M., Pérez, M., Romero, J., Sánchez-Lizaso, J. L., Roca, G., Jankowska, E., Pérez-Lloréns, J. L., Fournier, J., Montefalcone, M., Pergent, G., Ruiz, J. M., Cabaço, S., Cook, K., Wilkes, R. J., … Santos, R. (2019). Recent trend reversal for declining European seagrass meadows. *Nature Communications*, 10(1), 1–8.

De Luca, M., Piazzi, L., Guala, I., Cinti, M. F., Marras, P., Pansini, A., Pinna, F., Puccini, A., Santonastaso, A., Stelletti, M., Stipcich, P., & Pascucci, V. (2025). Restoration *of Posidonia oceanica* meadow using cuttings from an area impacted by harbor extension project. *Journal of Marine Science and Engineering*, 13(1), 3.
Den Hartog C. (1970). The seagrasses of the world. North Holland publ. Co, Amsterdam, Pays-Bas: 1-275.

Den Hartog, C. (1977). Structure, function, and classification in seagrass communities. In: McRoy CP and Helfferich (eds) A Scientific Perspective, pp 89–121. Marcel Dekker, New York.

Den Hartog, C., & Kuo, J. (2006). Taxonomy and Biogeography of Seagrasses. In Seagrasses: Biology, Ecology and Conservation; Larkum, A.W.D., Orth, R.J., Duarte, C., Eds.; Springer: Dordrecht, The Netherlands, 2006; pp. 1–23.

Descamp, P., Cornu, T., Bougerol, M., Boissery, P., Ferlat, C., Delaruelle, G., Deter, J., & Gobert, S. (2017). Experimental transplantation of *Posidonia oceanica*. 13th International MEDCOAST Congress on Coastal and Marine Sciences, Engineering, Management and Conservation, MEDCOAST 2017, 1(1), 471–482.

Descamp, P., Personnic, S., Gobert, S., Boulenger, A., Leduc, M., Delaruelle, G., Barroil, A., Marre, G., Holon, F., & Deter, J. (2025). Seagrass sod transplantation: A relevant tool for preventing the destruction of meadows in coastal construction projects. *Environmental Challenges*, 101087.

Deter, J., Lozupone, X., Inacio, A., Boissery, P., & Holon, F. (2017). Boat anchoring pressure on coastal seabed: Quantification and bias estimation using AIS data. *Marine Pollution Bulletin*, 123(1–2), 175–181.

de Vogel, F.A., Goudriaan, M., Zettler, E.R., Niemann, H., Eich, A., Weber, M., Lott, C., & Amaral-Zettler, L.A. (2024). Biodegradable plastics in Mediterranean coastal environments feature contrasting microbial succession. *Science of the Total Environment*, 928.

Dhariwal, A., Chong, J., Habib, S., King, I. L., Agellon, L. B., and Xia, J. (2017). MicrobiomeAnalyst: a web-based tool for comprehensive statistical, visual and meta-analysis of microbiome data. *Nucleic Acids Research*, 45 (1), 180–188.

Diaz-Almela, E., Marbà, N., & Duarte, C. M. (2007). Consequences of Mediterranean warming events in seagrass (*Posidonia oceanica*) flowering records. *Global Change Biology*, 13(1), 224–235.

Diaz-Gil, C., Grau, A., Grau, A. M., Palmer, M., Cabrera, R., Jordà, G., & Catalán, I. A. (2019). Changes in the juvenile fish assemblage of a Mediterranean shallow *Posidonia oceanica* seagrass nursery area after half century. *Mediterranean Marine Science*, 20(3).

Di Carlo, G., Badalamenti, F., Jensen, A.C., Koch, E.W., & Riggio, S. (2005). Colonisation process of vegetative fragments of *Posidonia oceanica* (L.) Delile on rubble mounds. *Marine Biology*, 147(6), 1261–1270.

Drius, M., Bongiorni, L., Depellegrin, D., Menegon, S., Pugnetti, A., & Stifter, S. (2019). Tackling challenges for Mediterranean sustainable coastal tourism: An ecosystem service perspective. *Science of The Total Environment*, 652, 1302–1317.

Duarte, C.M. (1991). Seagrass depth limits. *Aquatic Botany,* 40(4), 363–377.

Duarte, C. M., & Cebrián, J. (1996). The fate of marine autotrophic production. *Limnology and Oceanography*, 41(8), 1758–1766.

Duarte, C. M., & Chiscano, C. L. (1999). Seagrass biomass and production: a reassessment. *Aquatic Botany*, 65(1-4), 159-174.

Duarte, C.M. (2004). How can beaches be managed with respect to seagrass litter? In Borum, J., Duarte, C.M., Krause-Jansen, D. & Greeve, T.M., (eds.), European Seagrasses: An Introduction to Monitoring and Management. M&MS Project Publisher, pp. 83-84.

Duarte, C.M., Middelburg, J.J., & Caraco, N. (2005). Major role of marine vegetation on the oceanic carbon cycle. *Biogeosciences*, 2(1-8).

Duarte, C.M., Dennison, W.C., Orth, R.J. W., & Carruthers, T.J.B. (2008). The charisma of coastal ecosystems: Addressing the imbalance. *Estuaries and Coasts*, 31(2), 233–238.

Duarte, C.M., Marbà, N., Gacia, E., Fourqurean, J.W., Beggins, J., Barrón, C., & Apostolaki, E.T. (2010). Seagrass community metabolism: Assessing the carbon sink capacity of seagrass meadows. *Global Biogeochemical Cycles*, 24(4).

Duarte, C.M., Agusti, S., Barbier, E., Britten, G.L., Castilla, J. C., Gattuso, J. P., Fulweiler, R. W., Hughes, T.P., Knowlton, N., Lovelock, C.E., Lotze, H.K., Predragovic, M., Poloczanska, E., Roberts, C., & Worm, B. (2020). Rebuilding marine life. *Nature*, 580(7801), 39–51.

Duffy, J.E. (2006). Biodiversity and the functioning of seagrass ecosystems. *Marine Ecology Progress Series*, 311, 233–250.

Dutoit, T. (2011). La société internationale pour la restauration écologique, une association pour promouvoir la restauration des écosystèmes au niveau mondial. *Sciences, Eaux et Territoires, 5*, 6-9.

E

Ehrenfeld, J.G. (2000). Defining the limits of restoration: The need for realistic goals. *Restoration Ecology*, 8(1), 2–9.

Egan, S., Harder, T., Burke, C., Steinberg, P., Kjelleberg, S., & Thomas, T. (2013). The seaweed holobiont: Understanding seaweed–bacteria interactions. *FEMS Microbiology Reviews, 37*(4), 462–476.

Elliott, M., Burdon, D., Hemingway, K. L., & Apitz, S. E. (2007). Estuarine, coastal and marine ecosystem restoration: Confusing management and science – A revision of concepts. *Estuarine, Coastal and Shelf Science*, 74(3), 349–366.

Escandell-Westcott, A., Riera, R., & Hernández-Muñoz, N. (2023). *Posidonia oceanica* restoration review: Factors affecting seedlings. *Journal of Sea Research*, 191, 102337.

Estrada, M. (1996). Primary production in the northwestern Mediterranean. *Scientia Marina*, 60(2), 55–64.

Evrard, V., Kiswara, W., Bouma, T.J., & Middelburg, J.J. (2005) Nutrient dynamics of seagrass ecosystems: 15 N evidence for the importance of particulate organic matter and root systems. *Marine Ecology Progress Series*, 295, 49–55.

Ewanchuk, P.J. & Williams, S.L. (1996). Survival and re-establishment of vegetative fragments of eelgrass (*Zostera marina*). *Canadian Journal of Botany*, 74, 1584–1590.

F

Fabbrizzi, E., Giakoumi, S., De Leo, F., Tamburello, L., Chiarore, A., Colletti, A., Coppola, M., Munari, M., Musco, L., Rindi, F., Rizzo, L., Savinelli, B., Franzitta, G., Grech, D., Cebrian, E., Verdura, J., Bianchelli, S., Mangialajo, L., Nasto, I., … Fraschetti, S. (2023). The challenge of setting restoration targets for macroalgal forests under climate changes. *Journal of Environmental Management*, 326, 116834.

Fahimipour, A.K., Kardish, M.R., Lang, J. M., Green, J.L., Eisen, J.A., & Stachowicz, J.J. (2017). Global scale structure of the eelgrass microbiome. *Applied and Environmental Microbiology*, 83(12).

Foden, J. & Brazier, D.P. (2007). Angiosperms (seagrass) within the EU water framework directive: a UK perspective. *Marine Pollution Bulletin*, 55(1-6), 181-195.

Fontaine, Q., Fullgrabe, L., Borrossi, L., Chapat, M., Orticoni, S., Bitaly, M., Epaminondas, S., Poch, M., Agostini, A., Paoli Lecca, N., & Marengo, M. (2024). Etude de la fréquentation plaisancière en Corse en 2024. Rapport final avril 2025. Contrat OEC/STARESO. E05-24, 488 p.

Fourqurean, J.W., Duarte, C.M., Kennedy, H., Marbà, N., Holmer, M., Mateo, M.A., Apostolaki, E.T., Kendrick, G.A., Krause-Jensen, D., McGlathery, K. J., & Serrano, O. (2012). Seagrass ecosystems as a globally significant carbon stock. *Nature Geoscience*, 5(7), 505–509.

Frasca, S., Alabiso, A., D’Andrea, M.M., Cattaneo, R., & Migliore, L. (2024). Diversity and composition of *Posidonia oceanica*-associated bacterial and fungal communities: effect of boat-induced mechanical stress in the Villefranche-sur-Mer Bay (France). *Diversity*, 16(10), 604.

Fraser, M.W., Statton, J., Hovey, R.K., Laverock, B., & Kendrick, G.A. (2016). Seagrass derived organic matter influences biogeochemistry, microbial communities, and seedling biomass partitioning in seagrass sediments. *Plant and Soil*, 400, 133–146.

Fuggle, R.E., Gribben, P.E., & Marzinelli, E.M. (2023). Experimental evidence root-associated microbes mediate seagrass response to environmental stress. *Journal of Ecology*, 111(5), 1079–1093.

Fullgrabe, L., Grosjean, P., Gobert, S., Lejeune, P., Leduc, M., Engels, G., Dauby, P., Boissery, P., & Richir, J. (2020). Zooplankton dynamics in a changing environment: A 13-year survey in the northwestern Mediterranean Sea. *Marine Environmental Research, 159,* 104962.

Fullgrabe, L., Fontaine, Q., Marengo, M., Donnay, A., Sirjacobs, D., Iborra, L., Cnudde, S., Melllet, N., Seveno, J., Boulenger, A., …, & Lejeune, P. (2022). STARECAPMED (STAtion of Reference and Research on Change of Local and Global Anthropogenic Pressures on Mediterranean Ecosystems Drifts)—Year 2021. Research Report, STARESO: Calvi, France, 123p.

Fullgrabe, L., Fontaine, Q., Richir, J., Martelli, X., Boulenger, A., Borrossi, L., Woerhel, A., Balsaux, L., Iborra, L., Seimandi, T., Cnudde, S., Leduc, M., Dumas, M., Seveno, J., Chapat, M., Donnay, A., Boissery, P., Gobert, S., Lejeune, P. & Marengo, M. (2024). STARECAPMED (STAtion of Reference and rEsearch on Change of local and global

Anthropogenic Pressures on Mediterranean Ecosystems Drifts) – Année 2022-2023. Rapport de recherches, STARESO, 133 p.

G

Gacia, E., Granata, T.C., & Duarte, C.M. (1999). An approach to measurement of particle flux and sediment retention within seagrass (*Posidonia oceanica*) meadows. *Aquatic Botany*, 65(1–4), 255–268.

Gacia, E. & Duarte, C.M. (2001). Sediment retention by a Mediterranean *Posidonia oceanica* meadow: The balance between deposition and resuspension. *Estuarine, Coastal and Shelf Science* , 52(4), 505–514.

Gallmetzer, I., Pfuldfelder, B., Zekely, J. & Ott, J.A. (2005). Macrofauna diversity in *Posidonia oceanica* detritus: distribution and diversity of mobile macrofauna in shallow sublittoral accumulations of *Posidonia oceanica* detritus. *Marine Biology*, 147(2), 517-523.

Galil, B.S., & Zenetos, A. (2002). A Sea Change — Exotics in the Eastern Mediterranean Sea. Invasive Aquatic Species of Europe. *Distribution, Impacts and Management*, 325–336.

Gallet, S., Jaunatre, R., Regnery, B., Alignan, J.-F., Heckenroth, A., Muller, I., Bernez, I., Combroux, I., Glasser, T., Jund, S., Lelièvre, S., Malaval, S., Moussard, S., Vécrin-Stablo, M.-P., & Buisson, E. (2017). L'écologie de la restauration en France. Dynamique actuelle et rôle d'un réseau multi-acteurs, REVER. *Naturae*, 7, 1–11.

Galushko, A.S., & Rozanova, E.P. (1991). *Desulfobacterium cetonicum* sp. nov.: A sulfate-reducing bacterium which oxidizes fatty acids and ketones. *Microbiology*, 60(6), 742–746.

Gambi, M.C., Nowell, A.R.M., & Jumars, P.A. (1990). Flume observations on flow dynamics in *Zostera marina* (eelgrass) beds. *Marine Ecology Progress Series*, 61(1/2), 159–169.

Gamble C., Debney, A., Glover, A., Bertelli, C., Green, B., Hendy, I., Lilley, R., Nuuttila, H., Potouroglou, M., Ragazzola, F., Unsworth, R. and Preston, J, (eds) (2021). Seagrass Restoration Handbook. Zoological Society of London, UK., London, UK.

Ganthy, F., Soissons, L., Sauriau, P.-G., Verney, R., & Sottolichio, A. (2015). Effects of short flexible seagrass *Zostera noltei* on flow, erosion and deposition processes determined using flume experiments. *Sedimentology*, 62(4), 997–1023.

Garcias-Bonet, N., Marbà, N., Holmer, M., & Duarte, C. M. (2008). Effects of sediment sulfides on seagrass *Posidonia oceanica* meristematic activity. *Marine Ecology Progress Series*, 372, 1–6.

Garcias-Bonet, N., Arrieta, J.M., de Santana, C.N., Duarte, C.M., & Marbà, N. (2012). Endophytic bacterial community of a Mediterranean marine angiosperm (*Posidonia oceanica*). *Frontiers in Microbiology*, 3, 342.

Garcias-Bonet, N., Arrieta, J.M., Duarte, C.M., & Marbà, N. (2016). Nitrogen-fixing bacteria in Mediterranean seagrass (*Posidonia oceanica*) roots. *Aquatic Botany*, 131, 57–60.

Garcia-Castellanos, D., Estrada, F., Jiménez-Munt, I., Gorini, C., Fernández, M., Vergés, J., & De Vicente, R. (2009). Catastrophic flood of the Mediterranean after the Messinian salinity crisis. *Nature*, 462(7274), 778–781.

García-Martínez, M., Kuo, J., Kilminster, K., Walker, D., Rosselló-Mora, R., & Duarte, C. M. (2005). Microbial colonization in the seagrass *Posidonia* spp. roots. *Marine Biology Research*, 1(6), 388–395.

García-Martínez, M., López-López, A., Calleja, M. L., Marbà, N., & Duarte, C. M. (2009). Bacterial community dynamics in a seagrass (*Posidonia oceanica*) meadow sediment. *Estuaries and Coasts*, 32(2), 276–286.

García-Nieto, A.P., Geijzendorffer, I.R., Baró, F., Roche, P.K., Bondeau, A., & Cramer, W. (2018). Impacts of urbanization around Mediterranean cities: Changes in ecosystem service supply. *Ecological Indicators*, 91, 589–606.

Garrabou, J., Gómez-Gras, D., Medrano, A., Cerrano, C., Ponti, M., Schlegel, R., Bensoussan, N., Turicchia, E., Sini, M., Gerovasileiou, V., Teixido, N., Mirasole, A., Tamburello, L., Cebrian, E., Rilov, G., Ledoux, J.B., Souissi, J. Ben, Khamassi, F., Ghanem, R., … Harmelin, J.G. (2022). Marine heatwaves drive recurrent mass mortalities in the Mediterranean Sea. *Global Change Biology*, 28(19), 5708–5725.

Gauthier-Clerc, M., Mesléard, F., & Blondel, J. (2014). Sciences de la conservation (1ere edition), de boeck.

Genot, I., Caye, G., Meinesz, A., & Orlandini, M. (1994). Role of chlorophyll and carbohydrate contents in survival of *Posidonia oceanica* cuttings transplanted to different depths. *Marine Biology*, 119(1), 23–29.

Gera, A., Alcoverro, T., Mascaró, O., Pérez, M., & Romero, J. (2012). Exploring the utility of *Posidonia oceanica* chlorophyll fluorescence as an indicator of water quality within the European Water Framework Directive. *Environmental Monitoring and Assessment*, 184(6), 3675–3686.

Gera, A., Pagès, J.F., Romero, J., & Alcoverro, T. (2013). Combined effects of fragmentation and herbivory on *Posidonia oceanica* seagrass ecosystems. *Journal of Ecology*, 101, 1053–1061.

Giakoumi, S., Halpern, B.S., Michel, L.N., Gobert, S., Sini, M., Boudouresque, C.F., Gambi, M.C., Katsanevakis, S., Lejeune, P., Montefalcone, M., Pergent, G., Pergent-Martini, C., Sanchez-Jerez, P., Velimirov, B., Vizzini, S., Abadie, A., Coll, M., Guidetti, P., Micheli, F., & Possingham, H. P. (2015). Towards a framework for assessment and management of cumulative human impacts on marine food webs. *Conservation Biology*, 29(4), 1228–1234.

Gianni, F., Bartolini, F., Airoldi, L., Ballesteros, E., Francour, P., Guidetti, P., Meinesz, A., Thibaut, T., & Mangialajo, L. (2013). Conservation and restoration of marine forests in the Mediterranean Sea and the potential role of Marine Protected Areas. *Advances in Oceanography and Limnology*, 4(2), 83–101.

Giorgi, F. (2006). Climate change hot-spots. *Geophysical Research Letters*, 33(8), 8707.

Giorgi, F., & Lionello, P. (2008). Climate change projections for the Mediterranean region. *Global and Planetary Change*, 63(2–3), 90–104.

Giraud, G. (1977). Contribution à la description et à la phénologie quantitative des herbiers à *Posidonia oceanica* (L.) Delile. Thèse Doctorat 3ième cycle, Université Aix-Marseille II, France, 150 pp.

Giraud, G. (1979). Sur une méthode de mesure et de comptage des structures foliaires de *Posidonia oceanica* (Linnaeus) Delile. *Bulletin du Museum d'Histoire naturelle de Marseille*, 39, 33-39.

Gobert, S. (2002). Variations spatiale et temporelle de l'herbier à *Posidonia oceanica* (L.) Delile. Thèse Doctorat 3ième cycle, Université De Liège, Liège, Belgique, 226 pp.

Gobert, S., Laumont, N., & Bouquegneau, J.-M. (2002). *Posidonia oceanica* meadow: a low nutrient high chlorophyll (LNHC) system? *BMC Ecology*, 2, 9.

Gobert, S., Lepoint, G., Bouquegneau, J. M., Vangeluwe, D., Eisinger, M., Paster, M., Schuhmaker, H., & Van Treeck, P. (2005). Restoration of seagrass meadows: Means and limitations. Proceedings of the 7th International Conference on the Mediterranean Coastal Environment, MEDCOAST 2005, 1(September 2001), 461–472

Gobert, S., Lejeune, P., Lepoint, G., & Bouquegneau, J.M. (2005). C, N, P concentrations and requirements of flowering *Posidonia oceanica* shoots. *Hydrobiologia*, 533(1), 253–259.

Gobert, S., Cambridge, M.L., Velimirov, B., Pergent, G., Lepoint, G., Bouquegneau, J.M., Dauby, P., Pergent-Martini, C., & Walker, D.I. (2006). Biology of *Posidonia*. In Seagrasses: Biology, Ecology, and Conservation. Springer, Dordrecht, The Netherlands.

Gobert, S., Sartoretto, S., Rico-Raimondino, V., Andral, B., Chery, A., Lejeune, P., & Boissery, P. (2009). Assessment of the ecological status of Mediterranean French coastal waters as required by the Water Framework Directive using the Posidonia oceanica Rapid Easy Index: PREI. *Marine Pollution Bulletin*, 58(11), 1727–1733.

Gobert, S., Lepoint, G., Silva, J., Santos, R., Lejeune, P., Dujardin, P., Delvaux, B., Cornelis, J.-T., & Richir, J. (2015). A consensual Diving-PAM protocol to monitor *Posidonia oceanica* photosynthesis. *PeerJ*.

Gobert, S., Lepoint, G., Pelaprat, C., Remy, F., Lejeune, P., Richir, J., & Abadie, A. (2016). Temporal evolution of sand corridors in a *Posidonia oceanica* seascape: A 15-year study. *Mediterranean Marine Science*, 17(3), 777–784.

Gobert, S., Lefebvre, L., Boissery, P., & Richir, J. (2020). A non-destructive method to assess the status of *Posidonia oceanica* meadows. *Ecological Indicators,* 119, 106838.

González-Correa, J. M., Bayle, J. T., Sánchez-Lizaso, J. L., Valle, C., Sánchez-Jerez, P., & Ruiz, J.M. (2005). Recovery of deep *Posidonia oceanica* meadows degraded by trawling. *Journal of Experimental Marine Biology and Ecology*, 320(1), 65–76.

Govers, L.L., Suykerbuyk, W., Hoppenreijs, J.H.T., Giesen, K., Bouma, T.J., & van Katwijk, M.M. (2015). Rhizome starch as indicator for temperate seagrass winter survival. *Ecological Indicators*, 49, 53–60.

Gracia, A., Rangel-Buitrago, N., Oakley, J. A., & Williams, A. T. (2018). Use of ecosystems in coastal erosion management. *Ocean and Coastal Management*, 156, 277–289.

Graham, O.J., Adamczyk, E.M., Schenk, S., Dawkins, P., Burke, S., Chei, E., Cisz, K., Dayal, S., Elstner, J., Hausner, A.L.P., Hughes, T., Manglani, O., McDonald, M., Mikles, C., Poslednik, A., Vinton, A., Wegener Parfrey, L., & Harvell, C.D. (2024). Manipulation of the seagrass-associated microbiome reduces disease severity. *Environmental Microbiology*, 26(2).

Grime, J.P. (1974). Vegetation classification by reference to strategies. *Nature*, 250, 26–31.

Grossiord, C., Buckley, T.N., Cernusak, L. A., Novick, K.A., Poulter, B., Siegwolf, R.T. W., Sperry, J. S., & McDowell, N.G. (2020). Plant responses to rising vapor pressure deficit. *New Phytologist*, 226(6), 1550–1566.

Guerrero-Meseguer, L., Marín, A., & Sanz-Lázaro, C. (2017). Future heat waves due to climate change threaten the survival of *P. oceanica* seedlings. *Environmental Pollution*, 230, 40–45.

Guinaldo, T., Voldoire, A., Waldman, R., Saux Picart, S., & Roquet, H. (2023). Response of the sea surface temperature to heatwaves during the France 2022 meteorological summer. *Ocean Science*, 19 (3), 629–647.

H

Halpern, B.S., Walbridge, S., Selkoe, K.A. (2008). A global map of human impact on marine ecosystems. *Science*, 319, 946–948.

Haney, C.H., Samuel, B.S., Bush, J., & Ausubel, F.M. (2015). Associations with rhizosphere bacteria can confer an adaptive advantage to plants. *Nature Plants*, 1, 15051.

Hartman, K., van der Heijden, M.G.A., Wittwer, R.A., Banerjee, S., Walser, J.C., & Schlaeppi, K. (2018). Cropping practices manipulate abundance patterns of root and soil Microbiome members paving the way to smart farming. *Microbiome*, 6, 14.

Heck, K.L., Carruthers, T.J.B., Duarte, C. M., Randall Hughes, A., Kendrick, G., Orth, R.J., & Williams, S.W. (2008). Trophic transfers from seagrass meadows subsidize diverse marine and terrestrial consumers. *Ecosystems*, 11(7), 1198–1210.

Heiri, O., Lotter, A.F., & Lemcke, G. (2001). Loss on ignition as a method for estimating organic and carbonate content in sediments: Reproducibility and comparability of results. *Journal of Paleolimnology*, 25, 101–110.

Hemminga, M.A., Marbà, N. & Stapel, J. (1999). Leaf nutrient resorption, leaf lifespan and the retention of nutrients in seagrass systems. *Aquatic Botany*, 65(1-4), 0–158.

Hemminga, M.A., & Duarte, C.M. (2000). *Seagrass ecology*. Cambridge University Press, Cambridge, UK.

Hemminga, M.A., & Duarte, C.M. (2000). Taxonomy and distribution. In Seagrass Ecology; Hemminga, M.A., Duarte, C.M., Eds.; Cambridge University Press: Cambridge, UK, 2000; pp. 1–26.

Hendriks, I. E., Bouma, T. J., Morris, E. P., & Duarte, C. M. (2010). Effects of seagrasses and algae of the *Caulerpa* family on hydrodynamics and particle-trapping rates. *Marine Biology*, 157(3), 473–481.

Hensel, E., Patrick, C. J., Wilson, S. J., Song, B., Reay, W. G., & Orth, R. J. (2024). Incorporating generalist seagrasses enhances habitat restoration in a changing environment. *Journal of Applied Ecology, 61*(6), 1469–1480.

Hobbs, R. J. (2007). Setting effective and realistic restoration goals: Key directions for research. *Restoration Ecology*, 15(2), 354–357.

Holmer, M., Andersen, F. Ø., Nielsen, S. L., & Boschker, H. T. S. (2001). The importance of mineralization based on sulfate reduction for nutrient regeneration in tropical seagrass sediments. *Aquatic Botany*, 71(1), 1–17.
Holmer, M., Duarte, C. M., & Marbá, N. (2003). Sulfur cycling and seagrass (*Posidonia oceanica*) status in carbonate sediments. *Biogeochemistry*, 66(3), 223–239.

Holmer, M., Marbà, N., Lamote, M., & Duarte, C. M. (2009). Deterioration of sediment quality in seagrass meadows (*Posidonia oceanica*) invaded by macroalgae (*Caulerpa* sp.). *Estuaries and Coasts*, 32(3), 456–466.

Holon, F., Delaruelle, G., Boissery, P., Descamp, P., Deter, J., (2013). TEMPO : Réseau de surveillance des herbiers à Posidonie par télémétrie acoustique, Observatoire de la mer: www.observatoiremer.fr.

Holon, F., Boissery, P., Guilbert, A., Freschet, E., & Deter, J. (2015). The impact of 85 years of coastal development on shallow seagrass beds (*Posidonia oceanica* L. (Delile)) in South Eastern France: A slow but steady loss without recovery. *Estuarine, Coastal and Shelf Science*, 165, 204–212.

Holon, F., Marre, G., Parravicini, V., Mouquet, N., Bockel, T., Descamp, P., Tribot, A.S., Boissery, P., & Deter, J.

(2018). A predictive model based on multiple coastal anthropogenic pressures explains the degradation status of a marine ecosystem: Implications for management and conservation. *Biological Conservation*, 222(April), 125–135.

Hopkins, T.S. (1985). Physics of the sea. In: Margalef, R., ed. Key environments: Western Mediterranean. New York: Pergamon Press.

Horn, L.E., Paling ,E.I., & van Keulen, M. (2009). Photosynthetic recovery of transplanted *Posidonia sinuosa*, Western Australia. *Aquatic Botany*, 90, 149–156.

Houngnandan, F., Kéfi, S., & Deter, J. (2020). Identifying key-conservation areas for *Posidonia oceanica* seagrass beds. *Biological Conservation*, 247, 108546.

Huber, S.C., & Israel, D.W. (1982). Biochemical basis for partitioning of photosynthetically fixed carbon between starch and sucrose in soybean (*Glycine max* Merr.) leaves. *Plant Physiology*, 69, 691–696.

Hyndes, G.A., Nagelkerken, I., Mcleod, R. J., Connolly, R. M., Lavery, P.S., & Vanderklift, M.A. (2014). Mechanisms and ecological role of carbon transfer within coastal seascapes. *Biological Reviews*, 89(1), 232–254.

I

Imhoff, J.F. (2005). *Chromatiales ord. nov.* In: Brenner, D.J., et al. Bergey's Manual® of Systematic Bacteriology. Springer, Boston, MA.

IPBES. IPBES Global Assessment Summary for Policymakers. https://www.ipbes.net/news/ipbes-global-assessment-summary-policymakers-pdf (2019) (accessed 21/03/2022).

IPCC (2021). Climate Change 2021: The Physical Science Basis. Contribution of Working Group I to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change[Masson-Delmotte, V., P. Zhai, A. Pirani, S.L. Connors, C. Péan, S. Berger, N. Caud, Y. Chen, L. Goldfarb, M.I. Gomis, M. Huang, K. Leitzell, E. Lonnoy, J.B.R. Matthews, T.K. Maycock, T. Waterfield, O. Yelekçi, R. Yu, and B. Zhou (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

IPCC (2023). Summary for Policymakers. In: Climate Change 2023: Synthesis Report. Contribution of Working Groups I, II and III to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change [Core Writing Team, H. Lee and J. Romero (eds.)]. IPCC, Geneva, Switzerland, pp. 1-34.

Irving, A. D., Tanner, J. E., & Collings, G. J. (2014). Rehabilitating seagrass by facilitating recruitment: improving chances for success. *Restoration Ecology*, 22(2), 134–141.

Ismael, M., Charras, Q., Leschevin, M., Herfurth, D., Roulard, R., Quéro, A., Rusterucci, C., Domon, J.-M., Jungas, C., Vermeris, W., & Rayon, C. (2023). Seasonal variation in cell wall composition and carbohydrate metabolism in the seagrass *Posidonia oceanica* growing at different depths. *Plants*, 12(17), 3155.

J

Jackson, M. B., Ishizawa, K., & Ito, O. (2009). Evolution and mechanisms of plant tolerance to flooding stress. *Annals of Botany*, 103(2), 137–142.

Jamaludin, M.R. (2015). Carbon storage and preservation in seagrass meadows. Thesis in Environmental Science. Edith Cowan University – Western Australia, p. 188.

James, K., Macreadie, P.I., Burdett, H.L., Davies, I., & Kamenos, N.A. (2024). It's time to broaden what we consider a 'blue carbon ecosystem'. *Global Change Biology*, 30(5), e17261.

Jeyabaskaran, R., Jayasankar, J., Ambrose, T. V., Vineetha Valsalan, K.C., Divya, N.D., Raji, N., Vysakhan, P., John, S., Prema, D., Kaladharan, P., & Kripa, V. (2018). Conservation of seagrass beds with special reference to associated species and fishery resources. *Journal of the Marine Biological Association of India*, 60(1), 62–70.

Jiang, Z., Cui, L., Liu, S., Zhao, C., Wu, Y., Chen, Q., Yu, S., Li, J., He, J., Fang, Y., Premarathne Maha Ranvilage, C. I., & Huang, X. (2020). Historical changes in seagrass beds in a rapidly urbanizing area of Guangdong Province: Implications for conservation and management. *Global Ecology and Conservation*, 22.

Jiao, S., Chen, W., & Wei, G. (2022). Core microbiota drive functional stability of soil microbiome in reforestation ecosystems. *Global Change Biology*, 28(3), 1038–1047.

Jones, B.L., & Unsworth, R.K.F. (2016). The perilous state of seagrass in the British Isles. *Royal Society Open Science*, 3(1).

Jongen, R., Marzinelli, E.M., Bugnot, A.B., Ferguson, A., Fraser, M.W., Glasby, T.M., Jackson, E.L., Martin, B.C., Sherman, C.D.H., Sinclair, E.A., Trevathan-Tackett, S.M., Vergès, A., Waycott, M., Wright, J.T., Kendrick, G.A., & Gribben, P.E. (2024). Integrating belowground interactions into seagrass restoration strategies. *Oceanography and Marine Biology*, 62, 192–214.

K

Kaal J., Serrano O., del Río J.C., & Rencoret J. (2018). Radically different lignin composition in Posidonia species may link to differences in organic carbon sequestration capacity. *Organic Geochemistry*, 124, 247–256.

Kämpf, C., & Pfennig, N. (1980). Capacity of *Chromatiaceae* for chemotrophic growth. Specific respiration rates of *Thiocystis violacea* and *Chromatium vinosum*. *Archives of Microbiology*, 127(2), 125-135.

Katsanevakis, S., Issaris, Y., Poursanidis, D., & Thessalou-Legaki, M. (2010). Vulnerability of marine habitats to the invasive green alga *Caulerpa racemosa* var. *cylindracea* within a marine protected area. *Marine Environmental Research*, 70(2), 210–218.

Katsanevakis, S., Coll, M., Piroddi, C., Steenbeek, J., Ben Rais Lasram, F., Zenetos, A., & Cardoso, A.C. (2014). Invading the Mediterranean Sea: Biodiversity patterns shaped by human activities. *Frontiers in Marine Science*, 1.

Kelty, M.J. (2006). The role of species mixtures in plantation forestry. *Forest Ecology and Management*, 233, 195–204.

Kendrick, G.A., Duarte, C.M., & Marba, N. (2005). Clonality in seagrasses, emergent properties of seagrass landscapes. *Marine Ecology Progress Series*, 290, 291–296.

Kendrick, G.A., Marbà, N., & Duarte, C.M. (2005). Modelling formation of complex topography by the seagrass *Posidonia oceanica*. *Estuarine, Coastal and Shelf Science*, 65(4), 717–725.

Kessler, A.J., Wawryk, M., Marzocchi, U., Roberts, K.L., Wong, W.W., Risgaard-Petersen, N., Meysman, F.J.R., Glud, R.N., & Cook, P.L.M. (2019). Cable bacteria promote DNRA through iron sulfide dissolution. *Limnology and Oceanography*, 64(3), 1228–1238.

Kilminster, K.L., Walker, D.I., Thompson, P.A., & Raven, J. A. (2006). Limited nutritional benefit to the seagrass *Halophila ovalis*, in culture, following sediment organic matter enrichment. *Estuarine, Coastal and Shelf Science*, 68(3–4), 675–685.

Kiparissis, S., Fakiris, E., Papatheodorou, G., Geraga, M., Kornaros, M., Kapareliotis, A., & Ferentinos, G. (2011). Illegal trawling and induced invasive algal spread as collaborative factors in a *Posidonia oceanica* meadow degradation. *Biological Invasions*, 13(3), 669–678.

Kjeldsen, K.U., Schreiber, L., Thorup, C.A., Boesen, T., Bjerg, J.T., Yang, T., Dueholm, M.S., Larsen, S., Risgaard-Petersen, N., Nierychlo, M., Schmid, M., Bøggild, A., van de Vossenberg, J., Geelhoed, J.S., Meysman, F.J.R., Wagner, M., Nielsen, P.H., Nielsen, L.P., & Schramm, A. (2019). On the evolution and physiology of cable bacteria. *Proceedings of the National Academy of Sciences*, 116(38), 19116–19125.

König, S., Gros, O., Heiden, S. E., Hinzke, T., Thürmer, A., Poehlein, A., Meyer, S., Vatin, M., Mébéguié-A-Méboué, D., Tocny, J., Ponnudurai, R., Daniel, R., Becher, D., Schweder, T., & Markert, S. (2016). Nitrogen fixation in a chemoautotrophic lucinid symbiosis. *Nature Microbiology,* 2, 16193.

Kraemer, G.P., & Alberte, R.S. (1995). Impact of daily photosynthetic period on protein synthesis and carbohydrate stores in *Zostera marina* L. (eelgrass) roots: implications for survival in light-limited environments. *Journal of Experimental Marine Biology and Ecology*, 185, 191–202.

Kraemer, G. P., Mazzella, L., & Alberte, R. S. (1997). Nitrogen assimilation and partitioning in the Mediterranean seagrass *Posidonia oceanica*. *Marine Ecology*, 18(2), 175–188.

Kuo, J., & Kirkman, H. (1987). Floral and seedling morphology and anatomy of *Thalassodendron pachyrhizumden* Hartog (Cymodoceaceae). *Aquatic Botany*, 29, 1–7

L

Laegdsgaard, P. (2006). Ecology, disturbance and restoration of coastal saltmarsh in Australia: a review. *Wetlands Ecology and Management*, 14, 379–399.

Laffoley, D., & Grimsditch, G. (2009). The management of natural coastal carbon sinks. IUCN, Gland, Switzerland, p. 53pp.

Lai, S., Cheok, Z.Y., Bouma, T.J., Yaakub, S.M., & Todd, P.A. (2020). The relative importance of light and hydrodynamics in the decay and transport of vegetative seagrass fragments. *Estuarine, Coastal and Shelf Science*, 232.

Lamb, D., Erskine, P.D., & Parrotta, J.A. (2005). Restoration of degraded tropical forest landscapes. *Science*, 310, 1628–1632.

Lapeyra, J., Abadie, A., & Lejeune, P. (2016). Assessing edge-effects in *Posidonia oceanica* seagrass meadows: A multidisciplinary approach. Zoology 2016, Annual Meeting of the Zoological Society, Antwerp, Belgium.

Larkum, A. W.D., Orth, R.J., & Duarte, C. M. (2006). *Seagrasses: Biology, ecology and conservation*. Springer: Dordrecht, The Netherlands.

Larkum, A.W., Drew, E.A., & Ralph, P.J. (2007). Photosynthesis and metabolism in seagrasses at the cellular level. In *Seagrasses: Biology, Ecology and Conservation*; Springer: Dordrecht, The Netherlands, pp.323–345.

Larkum, A.W.D., Kendrick, G.A., & Ralph, P. J. (2018). Seagrasses of Australia: Structure, ecology and conservation. In Seagrasses of Australia: Structure, Ecology and Conservation. Springer International Publishing.

Lassauque, J. (2009). Ecophysiological early bio-indicators of anthropic-induced stress on *Posidonia oceanica* meadows. Thèse de doctorat. Université de Nice Sophia Antipolis, Nice (France).

Lau, W.W.Y. (2013). Beyond carbon: Conceptualizing payments for ecosystem services in blue forests on carbon and other marine and coastal ecosystem services. *Ocean & Coastal Management*, 83, 5–14.

Le Livre Bleu des engagements du Grenelle de la Mer. (2009). https://www.vie-publique.fr/sites/default/files/rapport/pdf/094000356.pdf (accessed 15 March 2022).

Leather, S.R., & Quicke, D.J.L. (2009). Do shifting baselines in natural history knowledge threaten the environment? *The Environmentalist*, 30(1), 1–2.

Lebeis, S.L., Paredes, S.H., Lundberg, D.S., Breakfield, N., Gehring, J., McDonald, M., Malfatti, S., Glavina del Rio, T., Jones, C. D., Tringe, S.G., & Dangl, J.L. (2015). Salicylic acid modulates colonization of the root microbiome by specific bacterial taxa. *Science*, 349(6250).

Lehnen, N., Marchant, H.K., Schwedt, A., Milucka, J., Lott, C., Weber, M., Dekaezemacker, J., Seah, B.K.B., Hach, P. F., Mohr, W., & Kuypers, M.M.M. (2016). High rates of microbial dinitrogen fixation and sulfate reduction associated with the Mediterranean seagrass *Posidonia oceanica*. *Systematic and Applied Microbiology*, 39(7), 476–483.

Lekha, K. R. (2004). Field instrumentation and monitoring of soil erosion in coir geotextile stabilised slopes—A case study. *Geotextiles and Geomembranes, 22*(5), 399–413.

Lejeusne, C., Chevaldonné, P., Pergent-Martini, C., Boudouresque, C.F., & Pérez, T. (2010). Climate change effects on a miniature ocean: the highly diverse, highly impacted Mediterranean Sea. *Trends in Ecology & Evolution*, 25(4), 250–260.

Lepoint, G., Millet, S., Dauby, P., Gobert, S., & Bouquegneau, J. M. (2002). Annual nitrogen budget of the seagrass *Posidonia oceanica* as determined by in situ uptake experiments. *Marine Ecology Progress Series*, 237, 87–96.

Lepoint, G., Vangeluwe, D., Eisinger, M., Paster, M., Van Treeck, P., Bouquegneau, J. M., & Gobert, S. (2004). Nitrogen dynamics in *Posidonia oceanica* cuttings: Implications for transplantation experiments. *Marine Pollution Bulletin*, 48(5–6), 465–470.

Les, D.H., Cleland, M.A., & Waycott, M. (1997). Phylogenetic Studies in Alismatidae, II: Evolution of Marine Angiosperms (Seagrasses) and Hydrophily. *Systematic Botany*, 22(3), 443–463.

Lester, S.E., Dubel, A.K., Hernán, G., McHenry, J., & Rassweiler, A. (2020). Spatial Planning Principles for Marine Ecosystem Restoration. *Frontiers in Marine Science*, 7.

Li, H., Liu, J., Zhang, L., Che, X., Zhang, M., & Zhang, T. (2024). A pilot restoration of *Enhalus acoroides* by transplanting dislodged rhizome fragments and its effect on the microbial diversity of submarine sediments. *Journal of Environmental Management*, 359.

Lilburn, T.G., Kim, K.S., Ostrom, N.E., Byzek, K.R., Leadbetter, J. R., & Breznak, J. A. (2001). Nitrogen fixation by symbiotic and free-living spirochetes. *Science*, 292(5526), 2495–2498.

Lim, S.J., Davis, B.G., Gill, D.E., Walton, J., Nachman, E., Engel, A.S., Anderson, L.C., & Campbell, B. J. (2019). Taxonomic and functional heterogeneity of the gill microbiome in a symbiotic coastal mangrove lucinid species. *The ISME Journal*, 13(4), 902–920.

Lin, L., Wang, X., Cao, L., & Xu, M. (2019). Lignin catabolic pathways reveal unique characteristics of dye-decolorizing peroxidases in *Pseudomonas putida*. *Environmental Microbiology*, 21(6), 1847–1863.

Lindenmayer, D.B., & Hobbs, R.J. (2004). Fauna conservation in Australian plantation forests – a review. *Biological Conservation*, 119, 151–168.

Liu, S., Jiang, Z., Deng, Y., Wu, Y., Zhang, J., Zhao, C., Huang, D., Huang, X., & Trevathan-Tackett, S. M. (2018). Effects of nutrient loading on sediment bacterial and pathogen communities within seagrass meadows. *Microbiology Open*, 7(5).

Lobet, G., Pagès, L., & Draye, X. (2011). A novel image-analysis toolbox enabling quantitative analysis of root system architecture. *Plant Physiology*, 157 (1), 29–39.

Lopez y Royo, C., Pergent, G., Alcoverro, T., Buia, M.C., Casazza, G., et al (2011). The seagrass *Posidonia oceanica* as indicator of coastal water quality: experimental intercalibration of classification systems. *Ecological Indicators*, 11(2), 557-563.

Lugo, A.E. (1997). The apparent paradox of reestablishing species richness on degraded lands with tree monocultures. *Forest Ecology and Management*, 99, 9–19.

Lundberg, D.S., Lebeis, S.L., Paredes, S.H., Yourstone, S., Gehring, J., Malfatti, S., Tremblay, J., Engelbrektson, A., Kunin, V., Rio, T.G., Edgar, R.C., Eickhorst, T., Ley, R. E., Hugenholtz, P., Tringe, S.G., & Dangl, J. L. (2012). Defining the core *Arabidopsis thaliana* root microbiome. *Nature*, 488 (7409), 86–90.

M

Madonia, A., Caporale, G., Penna, M., Bonamano, S., & Marcelli, M. (2021). Assessment of the photosynthetic response of *Posidonia oceanica* (Linneaus) Delile, 1813 along a depth gradient in the northern Tyrrhenian sea (Latium, Italy). *Geosciences*, 11(5).

Major, K.M., & Dunton, K.H (2002). Variations in light-harvesting characteristics of the seagrass, *Thalassia testudinum* : Evidence for photoacclimation. *Journal of Experimental Marine Biology and Ecology*, 275,173–189.

Malkin, S., & Cardini, U. (2021). Facilitative interactions on the rise: cable bacteria associate with diverse aquatic plants. *New Phytologist*, 232(5), 1897–1900.

Mancini, G., Casoli, E., Ventura, D., Jona Lasinio, G., Belluscio, A., & Ardizzone, G.D. (2021). An experimental investigation aimed at validating a seagrass restoration protocol based on transplantation. *Biological Conservation*, 264.

Mancini, G., Ventura, D., Casoli, E., Belluscio, A., & Ardizzone, G.D. (2022). Transplantation on a *Posidonia oceanica* meadow to facilitate its recovery after the Concordia shipwrecking. *Marine Pollution Bulletin*, 179, 113683.

Mancini, G., Mastrantonio, G., Pollice, A., Lasinio, G.J., Belluscio, A., Casoli, E., Pace, D.S., Ardizzone, G., & Ventura, D. (2023). Detecting trends in seagrass cover through aerial imagery interpretation: Historical dynamics of a *Posidonia oceanica* meadow subjected to anthropogenic disturbance. *Ecological Indicators*, 150, 110209.

Mancini, G., Ventura, D., Casoli, E., Michelangeli, F., Panarello, G., Belluscio, A., & Ardizzone, G. (2024). *Posidonia oceanica* (L.) Delile, 1813 seedlings as a potential integrative tool for restoring damaged meadows. *Restoration Ecology*, 32(5).

Mantelin, S., & Touraine, B. (2004). Plant growth-promoting bacteria and nitrate availability: Impacts on root development and nitrate uptake. *Journal of Experimental Botany*, 55(394), 27–34.

Marbà, N., Duarte, C.M., Cebrian, J., Gallegos, M.E., Olesen, B. & Sand-Jensen, K. (1996). Growth and population dynamics of *Posidonia oceanica* on the Spanish Mediterranean coast: elucidating seagrass decline. *Marine Ecology Progress Series*, 37, 203-213.

Marbà, N., & Duarte, C.M. (1998). Rhizome elongation and seagrass clonal growth.

Marine Ecology Progress Series, 174, 269–280.

Marbà, N., Duarte, C.M., Holmer, M., Martínez, R., Basterretxea, G., Orfila, A., Jordi & A., Tintoré, J. (2002). Effectiveness of protection of seagrass (*Posidonia oceanica*) populations in Cabrera National Park (Spain). *Environmental Conservation*, 29(4).

Marbà, N., Hemminga, M.A., Mateo, M.A., Duarte, C.M., Mass, Y.E.M., Terrados, J., & Gacia, E. (2002). Carbon and nitrogen translocation between seagrass ramets. *Marine Ecology Progress Series*, 226, 287-300.

Marbà, N., Holmer, M., Gacia, E., & Barron, C. (2006). Seagrass beds and coastal biogeochemistry. In Seagrasses: Biology, Ecology and Conservation. Springer, Dordrecht, The Netherlands.

Marbà, N., Santiago, R., Diaz-Almela, E., Alvarez, E. & Duarte, C.M. (2006). Seagrass (*Posidonia oceanica*) vertical growth as an early indicator of fish farm-derived stress. Estuarine, *Coastal and Shelf Science*, 67, 475-483.

Marbà, N., Díaz-Almela, E., & Duarte, C. M. (2014). Mediterranean seagrass (*Posidonia oceanica*) loss between 1842 and 2009. In *Biological Conservation*, 176, 183–190.

Marbà, N., Arias-Ortiz, A., Masqué, P., Kendrick, G. A., Mazarrasa, I., Bastyan, G. R., Garcia-Orellana, J., & Duarte, C. M. (2015). Impact of seagrass loss and subsequent revegetation on carbon sequestration and stocks. Journal of Ecology, 103(2), 296–302.

Marco-Méndez, C., Marbà, N., Amores, Á., Romero, J., Minguito-Frutos, M., García, M., Pagès, J. F., Prado, P., Boada, J., Sánchez-Lizaso, J. L., Ruiz, J. M., Muñoz-Ramos, G., Sanmartí, N., Mayol, E., Buñuel, X., Bernardeau-Esteller, J., Navarro-Martinez, P. C., Marín-Guirao, L., Morell, C., … Alcoverro, T. (2024). Evaluating the extent and impact of the extreme Storm Gloria on *Posidonia oceanica* seagrass meadows. *Science of the Total Environment*, 908.

Marín-Guirao, L., Entrambasaguas, L., Ruiz, J. M., & Procaccini, G. (2019). Heat-stress induced flowering can be a potential adaptive response to ocean warming for the iconic seagrass *Posidonia oceanica*. *Molecular Ecology*, 28(10), 2486–2501.

Marre, G., Deter, J., Holon, F., Boissery, P., & Luque, S. (2020). Fine-scale automatic mapping of living *Posidonia oceanica* seagrass beds with underwater photogrammetry. *Marine Ecology Progress Series*, 643, 63–74.

Marre, G., Holon, F., Luque, S., Boissery, P., & Deter, J. (2019). Monitoring Marine Habitats With Photogrammetry: A Cost-Effective, Accurate, Precise and High-Resolution Reconstruction Method. *Frontiers in Marine Science*, 6, 1–15.

Martin, D. M. (2017). Ecological restoration should be redefined for the twenty-first century. Restoration Ecology, 25(5), 668–673.

Martin, B. C., Gleeson, D., Statton, J., Siebers, A. R., Grierson, P., Ryan, M. H., & Kendrick, G. A. (2018). Low light availability alters root exudation and reduces putative beneficial microorganisms in seagrass roots. *Frontiers in Microbiology*, 8.

Martin, B. C., Alarcon, M. S., Gleeson, D., Middleton, J. A., Fraser, M. W., Ryan, M. H., Holmer, M., Kendrick, G. A., & Kilminster, K. (2020). Root microbiomes as indicators of seagrass health. *FEMS Microbiology Ecology*, 96, 201.

Marullo, S., Serva, F., Iacono, R., Napolitano, E., di Sarra, A., Meloni, D., Monteleone, F., Sferlazzo, D., De Silvestri, L., de Toma, V., Pisano, A., Bellacicco, M., Landolfi, A., Organelli, E., Yang, C., & Santoleri, R. (2023). Record-breaking persistence of the 2022/23 marine heatwave in the Mediterranean Sea.

Environmental Research Letters, 18(11), 114041.

Masson-Delmotte, V., Zhai, P., Portner, H.-O., Roberts, D., Skea, J., Calvo, E., Priyadarshi, B., Shukla, R., Ferrat, M., Haughey, E., Luz, S., Neogi, S., Pathak, M., Petzold, J., Pereira, J.P., Vyas, P., Huntley, E., Kissick, K., Belkacemi, M.,& Malley, J. (2019). IPCC, 2019: Climate Change and Land: an IPCC special report on climate change, desertification, land degradation, sustainable land management, food. https ://spiral.imperial.ac.uk/bitstream/10044/1/ 76618/2/SRCCL-Full-Report-Compil ed-191128.pdf

Mateo, M.A. & Romero, J. (1996). Evaluating seagrass leaf litter decomposition: an experimental comparison between litter-bag and oxygen-uptake methods. *Journal of Experimental Marine Biology and Ecology*, 202(2), 97–106.

Mateo, M.A., & Romero, J. (1997). Detritus dynamics in the seagrass *Posidonia oceanica*: elements for an ecosystem carbon and nutrient budget. *Marine Ecology Progress Series*, 151, 43-53.

Mateo, M.A., Romero, J., Perez, M., Littler, M.M., & Littler, D.S. (1997). Dynamics of millenary organic deposits resulting from the growth of the Mediterranean seagrass *Posidonia oceanica*. *Estuarine, Coastal and Shelf Science*, 44, 103-110.

Mateo, M.-Á., Sánchez-Lizaso, J.-L., & Romero, J. (2003). *Posidonia oceanica* 'banquettes': A preliminary assessment of the relevance for meadow carbon and nutrients budget. *Estuarine, Coastal and Shelf Science*, 56(1), 85–90.

Mateo, M.A., Cebrian, J., Dunton, K., Mutchler, T. (2006). Carbon flux in seagrass ecosystems. In: Orth, R.J., Duarte, C.M. (Eds.), Seagrass: Biology, Ecology and Conservation, Larkum AWD. Springer, New York, pp. 159–192.

Maxwell, P.S., Eklöf, J. S., van Katwijk, M. M., O'Brien, K.R., de la Torre-Castro, M., Boström, C., Bouma, T.J., Krause-Jensen, D.,Unsworth, R.K.F., van Tussenbroek, B.I., & van der Heide, T. (2017).The fundamental role of ecological feedback mechanisms for the adaptive management of seagrass ecosystems—A review. *Biological Reviews of the Cambridge Philosophical Society,* 92, 1521–1538.

Mazella, L. & Ott, J.A. (1984). Seasonal changes in some features of *Posidonia oceanica* (L.) Delile leaves and epiphytes at different depths. In : Boudouresque, C.F., Jeudy de Grissac, A., Olivier, J. edits International Workshop on *Posidonia oceanica* beds, GIS Posidonie publ., Fr., 1, 119-127

McCauley, D.J., Pinsky, M.L., Palumbi, S.R., Estes, J.A., Joyce, F.H., & Warner, R.R. (2015). Marine defaunation: Animal loss in the global ocean. *Science*, 347(6219), 1255641.

McDonald, T., Gann, G.D., Jonson, J., & Dixon, K.W. (2016). International standards for the practice of ecological restoration – including principles and key concepts. Society for Ecological Restoration, Washington D.C.

McKenzie, L.J., Nordlund, L.M., Jones, B.L., Cullen-Unsworth, L.C., Roelfsema, C., & Unsworth, R.K.F. (2020). The global distribution of seagrass meadows. *Environmental Research Letters*, 15(7).

McLeod, E., Chmura, G.L., Bouillon, S., Salm, R., Björk, M., Duarte, C. M., Lovelock, C.E., Schlesinger, W.H., & Silliman, B.R. (2011). A blueprint for blue carbon: Toward an improved understanding of the role of vegetated coastal habitats in sequestering CO_2. *Frontiers in Ecology and the Environment*, 9(10), 552–560.

McNeil, B.I., Matear, R.J., & At, D.J.M. (2007). Climate change feedbacks on future oceanic acidification. *Chemical and Physical Meteorology*, 59(2), 191–198.

MEDTRIX (2025). Cahier de surveillance #23 : Mars, avril et mai 2025. *MEDTRIX – La plateforme de surveillance des eaux côtières et des écosystèmes de Méditerranée.*

Meinesz, A. & Lefevre, J.R. (1984). Régénération d'un herbier à *Posidonia oceanica* quarante années après sa destruction par une bombe dans la rade de Villefranche (Alpes-Maritimes). In: Boudouresque, C.F., Jeudy de Grissac, A., Olivier, J. edits. International Workshop on *Posidonia oceanica* beds, GIS Posidonie publ., Fr., 1, 39-44.

Meinesz, A., Lefevre, L.R. & Astier, J.M. (1991). Impact of coastal development on the infralittoral zone along the southeastern Mediterranean shore of continental France. *Marine Pollution Bulletin*, 23, 0–347.

Meinesz, A., Molenaar, H., Bellone, E., & Loques, F. (1992). Vegetative Reproduction in *Posidonia oceanica* I. Effects of Rhizome Length and Transplantation Season in Orthotopic Shoots. *Marine Ecology*, 13(2), 163–174.

Metropolis, N., & Ulam, S. (1949). The monte carlo method. *Journal of the American Statistical Association*, 44, 335–341.

Micheli, F., Halpern, B.S., Walbridge, S., Ciriarco, S., Ferretti, F., Fraschetti, S., Lewison, R., Nykjaer, L., & Rosenberg, A.A. (2013). Cumulative human impacts on Mediterranean and Black Sea marine ecosystems: assessing current pressures and opportunities. *PloS One*, 8, e79889.

Milazzo, M., Badalamenti, F., Ceccherelli, G., & Chemello, R. (2004). Boat anchoring on *Posidonia oceanica* beds in a marine protected area (Italy, western Mediterranean): Effect of anchor types in different anchoring stages. *Journal of Experimental Marine Biology and Ecology*, 299(1), 51–62.

Millennium Ecosystem Assessment MEA (2005). Ecosystem and Human Well-Being, General Synthesis. Island Press, Washington D.C

Mitsch, W.J. (2012). What is ecological engineering? *Ecological Engineering*, 45, 5–12.

Mitsch, W.J., & Jørgensen, S.E. (2003). Ecological engineering: A field whose time has come. *Ecological Engineering*, 20(5), 363–377.

Mohapatra, M., Dash, S. P., & Rastogi, G. (2024). Genomic approaches for the investigation of seagrass rhizosphere microbiome and bioprospecting potential: A field study from Chilika Lagoon, Odisha, India. *Applications of Metagenomics: Agriculture, Environment, and Health*, 10, 201–234.

Mohr, W., Lehnen, N., Ahmerkamp, S., Marchant, H.K., Graf, J.S., Tschitschko, B., Yilmaz, P., Littmann, S., Gruber-Vodicka, H., Leisch, N., Weber, M., Lott, C., Schubert, C.J., Milucka, J., & Kuypers, M.M.M. (2021). Terrestrial-type nitrogen-fixing symbiosis between seagrass and a marine bacterium. *Nature*, 600(7887), 105–109.

Moksnes, P.-O., Röhr, M.E., Holmer, M., Eklöf, J.S., Eriander, L., Infantes, E., & Boström, C. (2021). Major impacts and societal costs of seagrass loss on sediment carbon and nitrogen stocks. *Ecosphere*, 12(7).

Molenaar, H. (1992). Etude de la transplantation de boutures de *Posidonia oceanica* (L.) Delile, phanérogames marines. Modélisation de l'architecture et du mode de croissance. Thèse de doctorat. Université de Nice, Nice (France).

Molenaar, H., & Meinesz, A. (1992). Vegetative reproduction in *Posidonia oceanica*. Effects of depth changes on transplanted orthotopic shoots. *Marine Ecology*, 13, 175–185.

Molenaar, H., Meinesz, A., & Caye, G. (1993). Vegetative reproduction in *Posidonia oceanica*. Survival and development in different morphological types of transplanted cuttings. *Botanica Marina*, 36(6), 481–488.

Molenaar, H., & Meinesz, A. (1995). Vegetative Reproduction in *Posidonia oceanica*: Survival and Development of Transplanted Cuttings According to Different Spacings, Arrangements and Substrates. *Botanica Marina*, 38, 313–322.

Molenaar H., Barthelemy D., Reffye De P., Meinesz A., Mialet I., 2000. Modelling architecture and growth pattern of *Posidonia oceanica*. *Aquatic Botany*, 66, 85-99.

Molinier, R. & Picard, J. (1952). Recherches sur les herbiers de phanérogames marines du littoral méditerranéen français. *Annales de l'institut océanographique*, 27(3), 157-234.

Monnier, B., Pergent, G., Valette-Sansevin, A., Boudouresque, C.F., Mateo, M.A., Pergent- Martini, C. (2020). The *Posidonia oceanica* matte: a unique coastal carbon sink for climate change mitigation and implications for management. *Vie & Milieu*, 70, 3-4.

Monnier, B., Pergent, G., Mateo, M. Á., Clabaut, P., & Pergent-Martini, C. (2022). Quantification of blue carbon stocks associated with *Posidonia oceanica* seagrass meadows in Corsica (NW Mediterranean). *Science of the Total Environment*, 838.

Montefalcone, M., Lasagna, R., Bianchi, C. N., Morri, C., & Albertelli, G. (2006). Anchoring damage on *Posidonia oceanica* meadow cover: A case study in Prelo cove (Ligurian Sea, NW Mediterranean). *Chemistry and Ecology*, 22.

Montefalcone, M., Albertelli, G., Morri, C., & Bianchi, C. N. (2007). Urban seagrass: Status of *Posidonia oceanica* facing the Genoa city waterfront (Italy) and implications for management. *Marine Pollution Bulletin*, 54(2), 206–213.

Montefalcone, M., Chiantore, M., Lanzone, A., Morri, C., Albertelli, G., & Bianchi, C. N. (2008). BACI design reveals the decline of the seagrass *Posidonia oceanica* induced by anchoring. *Marine Pollution Bulletin*, 56(9), 1637–1645.

Montefalcone, M. (2009). Ecosystem health assessment using the Mediterranean seagrass *Posidonia oceanica*: A review. *Ecological Indicators*, 9(4), 595–604.

Montefalcone, M., Giovannetti, E., Morri, C., Peirano, A., & Bianchi, C. N. (2013). Flowering of the seagrass *Posidonia oceanica* in NW Mediterranean: is there a link with solar activity?. *Mediterranean Marine Science*, 14(2), 416–423.

Montefalcone, M. (2024). Challenges in restoring Mediterranean seagrass ecosystems in the Anthropocene. *Environments, 11*(5), 86.

Morando, M., & Capone, D.G. (2016). Intraclude heterogeneity in nitrogen utilization by marine prokaryotes revealed using stable isotope probing coupled with tag sequencing (Tag-SIP). *Frontiers in Microbiology*, 7, 1932.

Morris, R.L., Konlechner, T.M., Ghisalberti, M., & Swearer, S.E. (2018). From grey to green: Efficacy of eco-engineering solutions for nature-based coastal defence. *Global Change Biology*, 24(5), 1827–1842.

Mtwana Nordlund, L., Koch, E.W., Barbier, E.B., & Creed, J.C. (2016). Seagrass ecosystem services and their variability across genera and geographical regions. *PLoS ONE*, 11(10).

N

Nam, V.N., Sasmito, S.D., Murdiyarso, D., Purbopuspito, J., & MacKenzie, R.A. (2016). Carbon stocks in artificially and naturally regenerated mangrove ecosystems in the Mekong Delta. *Wetlands Ecology and Management*, 24(2), 231–244.

Nitsch, C.K., Walters, L.J., Sacks, J.S., Sacks, P.E., & Chambers, L.G. (2021). Biodegradable material for oyster reef restoration: First-year performance and biogeochemical considerations in a coastal lagoon. *Sustainability*, 13(13).

O

Odum, H.T., & Odum, B. (2003). Concepts and methods of ecological engineering. *Ecological Engineering*, 20, 339–361.

Offret, C., Desriac, F., Le Chevalier, P., Mounier, J., Jégou, C., & Fleury, Y. (2016). Spotlight on antimicrobial metabolites from the marine bacteria *Pseudoalteromonas*: Chemodiversity and ecological significance. *Marine Drugs,* 14(7), 129.

Olesen, B., Marbà, N., Duarte, C.M., Savela, R.S., & Fortes, M.D. (2004). Recolonization dynamics in a mixed seagrass meadow: The role of clonal versus sexual processes. *Estuaries*, 27, 770–780.

Orth, R. J., Lefcheck, J. S., McGlathery, K. S., Aoki, L., Luckenbach, M. W., Moore, K. A., Oreska, M. P. J., Snyder, R., Wilcox, D. J., & Lusk, B. (2020). Restoration of seagrass habitat leads to rapid recovery of coastal ecosystem services. *Science Advances*, 6(41), eabc6434.

Osvatic, J.T., Wilkins, L.G.E., Leibrecht, L., & Yuen, B. (2021). Global biogeography of chemosynthetic symbionts reveals both localized and globally distributed symbiont groups. *Proceedings of the National Academy of Sciences,* 118(29), e2104378118.

Öztürk, B. (2021). Non-indigenous species in the Mediterranean and the Black Sea. Studies and Reviews No. 87 (General Fisheries Commission for the Mediterranean). FAO.

P

Pandolfi, J.M. (2008). Succession. In S.E. Jørgensen & B.D. Fath (Eds.), *Encyclopedia of ecology*, 3416–3424.

Pansini, A., La Manna, G., Pinna, F., Stipcich, P., & Ceccherelli, G. (2021). Trait gradients inform predictions of seagrass meadows changes to future warming. *Scientific reports*, 11, 18107.

Pansini, A., Bosch-Belmar, M., Berlino, M., Sarà, G., & Ceccherelli, G. (2022). Collating evidence on the restoration efforts of the seagrass *Posidonia oceanica*: current knowledge and gaps. *Science of The Total Environment*, 851, 158320.

Pansini, A., Deroma, M., Guala, I., Monnier, B., Pergent-Martini, C., Piazzi, L., Stipcich, P., & Ceccherelli, G. (2024). The resilience of transplanted seagrass traits encourages detection of restoration success. *Journal of Environmental Management*, 357, 120744.

Papworth, S. K., Rist, J., Coad, L., & Milner-Gulland, E. J. (2009). Evidence for shifting baseline syndrome in conservation. *Conservation Letters*, 2(2), 93–100.

Pasqualini, V., Pergent-Martini, C., Clabaut, P., & Pergent, G. (1998). Mapping of *Posidonia oceanica* using Aerial Photographs and Side Scan Sonar: Application off the Island of Corsica (France). *Estuarine, Coastal and Shelf Science*, 47.

Pasqualini, V., Pergent-Martini, C., & Pergent, G. (1999). Environmental impact identification along the Corsican coast (Mediterranean sea) using image processing. *Aquatic Botany*, 65, 311–320.

Patriquin, D.G. (1972). The origin of nitrogen and phosphorus for growth of the marine angiosperm *Thalassia testudinum*. *Marine Biology*, 15, 35–46.

Paulsen, S.S., Strube, M.L., Bech, P.K., Gram, L., & Sonnenschein, E. C. (2019). Marine chitinolytic *Pseudoalteromonas* represents an untapped reservoir of bioactive potential. *mSystems, 4*(4), e00060-19.

Pauly, D. (1995). Anecdotes and the shifting baseline syndrome of fisheries. *Trends in Ecology and Evolution*, 10, 430.

Pedersen, A.-G. U., Berntsen, J., & Lomstein, B.A. (1999). The effect of eelgrass decomposition on sediment carbon and nitrogen cycling: A controlled laboratory experiment. *Limnology and Oceanography*, 44(8), 1978–1991.

Peirano, A., Damasso, V., Montefalcone, M., Morri, C., & Bianchi, C.N. (2005). Effects of climate, invasive species and anthropogenic impacts on the growth of the seagrass *Posidonia oceanica* (L.) Delile in Liguria (NW Mediterranean Sea). *Marine Pollution Bulletin*, 50(8), 817–822.

Pereda-Briones, L., Tomas, F., & Terrados, J. (2018). Field transplantation of seagrass (*Posidonia oceanica*) seedlings: Effects of invasive algae and nutrients. *Marine Pollution Bulletin*, 134, 160–165.

Pérez, M., García, T., Invers, O., & Ruiz, J. M. (2008). Physiological responses of the seagrass *Posidonia oceanica* as indicators of fish farm impact. *Marine Pollution Bulletin*, 56(5), 869–879.

Pergent, G. & Pergent-Martini, C. (1991). Leaf renewal cycle and primary production of *Posidonia oceanica* in the Bay of Lacco Ameno (Ischia, Italy) using lepidochronological analysis. *Aquatic Botany*, 42(1), 49-66.

Pergent, G., & Kempf, M. (1993). L'environnement marin côtier en Tunisie. https://archimer.ifremer.fr/doc/00105/21592/

Pergent, G., Romero, J., Pergent-Martini, C., Mateo, M.-A., & Boudouresque, C.-F. (1994). Primary production, stocks and fluxes in the Mediterranean seagrass *Posidonia oceanica*. *Marine Ecology Progress Series*, 106(1).
Pergent, G., Pergent-Martini, C. & Boudouresque, C.F. (1995). Utilisation de l'herbier de *Posidonia oceanica* comme indicateur biologique de la qualité du milieu littoral en Méditerranée : état des connaissances. *Mésogée*, 54, 3-27.

Pergent, G., Rico-Raimondino, V. & Pergent-Martini, C. (1997). Fate of primary production in *Posidonia oceanica* meadows of the Mediterranean. *Aquatic Botany*, 59(3-4), 307–321.

Pergent, G., Bazairi, H., Bianchi, C. N., Boudouresque, C. F., Buia, M. C., Calvo, S., Clabaut, P., Harmelin-Vivien, M., Angel Mateo, M., Montefalcone, M., Morri, C., Orfanidis, S., Pergent-Martini, C., Semroud, R., Serrano, O., Thibaut, T., Tomasello, A., & Verlaque, M. (2014). Climate change and Mediterranean seagrass meadows: A synopsis for environmental managers. *Mediterranean Marine Science*, 15(2), 462–473.

Pergent, G., Pergent-Martini, C., Bein, A., Dedeken, M., Oberti, P., Orsini, A., Santucci, J.-F., & Short, F., (2015). Dynamic of *Posidonia oceanica* seagrass meadows in the northwestern Mediterranean: Could climate change be to blame? *Comptes Rendus Biologies*, 338(7), 484–493.

Pergent-Martini, C., Rico-Raimondino, V. & Pergent, G. (1994). Primary production of *Posidonia oceanica* in the Mediterranean Basin. *Marine Biology*, 120(1), 9–15.

Pergent-Martini, C., Pasqualini, V., Pergent, G., & Ferrat, L. (2002). Effect of a newly set up wastewater-treatment plant on a marine phanerogam seagrass bed - A medium-term monitoring program. *Bulletin of Marine Science*, 71(3), 1227–1236.

Pergent-Martini, C., Leoni, V., Pasqualini, V., Ardizzone, G. D., Balestri, E., Bedini, R., Belluscio, A., Belsher, T., Borg, J., Boudouresque, C. F., Boumaza, S., Bouquegneau, J. M., Buia, M. C., Calvo, S., Cebrian, J., Charbonnel, E., Cinelli, F., Cossu, A., Di Maida, G., … Velimirov, B. (2005). Descriptors of *Posidonia oceanica* meadows: Use and application. *Ecological Indicators*, 5(3), 213–230.

Pergent-Martini, C., Pergent, G., Monnier, B., Boudouresque, C.F., Mori, C., & Valette-Sansevin, A. (2021a). Contribution of *Posidonia oceanica* meadows in the context of climate change mitigation in the Mediterranean Sea. *Marine Environmental Research*, 165, 105236.

Pergent-Martini, C., Torchia, G., Rais, C., Villata, C., Bouafif, C., Burzio, P., Stirpe, S., Managanza, A., & Bezzo, T., (2021b). Comparative Legal Analysis on Protection of *Posidonia* Meadows in the Mediterranean Sea : a Benchmark on Regulations on Anchoring (Report Golder-Okianos & OFB, N◦20144905/13206, 122 p).

Pergent-Martini, C., Monnier, B., Lehmann, L., Barralon, E., & Pergent, G. (2022). Major regression of *Posidonia oceanica* meadows in relation with recreational boat anchoring: a case study from Sant'Amanza bay. *Journal of Sea Research*, 110490.

Pergent-Martini, C., André, S., Castejon, I., Deter, J., Frau, F., Gerakaris, V., Mancini, G., Molenaar, H., Montefalcone, M., Oprandi, A., Pergent, G., Poursanidis, D., Royo, L., Terrados, J., Tomasello, A., Ventura, D., Villers, F., (2024). Guidelines for *Posidonia oceanica* restoration. Report Cooperation agreement Mediterranean Posidonia Network (MPN), French Biodiversity Agency (OFB) & University of Corsica Pasquale Paoli (UCPP) N°OFB-22-1310. 133 p. + Appendices.

Personnic, S., Boudouresque, C. F., Astruch, P., Ballesteros, E., Blouet, S., Bellan-Santini, D., Bonhomme, P., Thibault-Botha, D., Feunteun, E., Harmelin-Vivien, M., Pergent, G., Pergent-Martini, C., Pastor, J., Poggiale, J. C., Renaud, F., Thibaut, T., & Ruitton, S. (2014). An ecosystem-based approach to assess the status of a mediterranean ecosystem, the Posidonia oceanica seagrass meadow. *PLoS ONE*, 9(6).

Petersen, J. M., Kemper, A., Gruber-Vodicka, H., Cardini, U., van der Geest, M., Kleiner, M., Bulgheresi, S., Mußmann, M., Herbold, C., Seah, B. K. B., Antony, C. P., Liu, D., Belitz, A., & Weber, M. (2016). Chemosynthetic symbionts of marine invertebrate animals are capable of nitrogen fixation. *Nature Microbiology*, 2, 16195.

Pfeiffer, N. (2007). La réhabilitation de la Réserve de la Grand'Mare. *Faune Sauvage*, 278, 22–25.

Piazzi, L., Balestri, E., Magri, M., & Cinelli, F. (1998). Experimental transplanting of *Posidonia oceanica* (L.) Delile into a disturbed habitat in the Mediterranean Sea. *Botanica Marina*, 41(1–6), 593–601.

Piazzi, L., Acunto, S., & Cinelli, F. (1999). In situ survival and development of *Posidonia oceanica* (L.) Delile seedlings. *Aquatic Botany*, 63, 103–112.

Piazzi L., Balestri E., Balata D., & Cinelli F. (2000). Pilot transplanting experiment of *Posidonia oceanica* (L.) Delile to restore a damaged coastal area in the Mediterranean Sea. *Biologia Marina Mediterranea*, 7(2), 409-411.

Piazzi, L., Acunto, S., Frau, F., Atzori, F., Cinti, M. F., Leone, L. M., & Ceccherelli, G. (2021). Environmental engineering techniques to restore degraded *Posidonia oceanica* meadows. *Water*, 13(5), 1–10.

Piazzolla, D., Scanu, S., Mancuso, F. P., Bosch-Belmar, M., Bonamano, S., Madonia, A., Scagnoli, E., Tantillo, M. F., Russi, M., Savini, A., Fersini, G., Sarà, G., Coppini, G., Marcelli, M., & Piermattei, V. (2024). An integrated approach for the benthic habitat mapping based on innovative surveying technologies and ecosystem functioning measurements. *Scientific Reports*, 14, 5888.

Picard, J. (1965). Importance, répartition et rôle du matériel organique végétal issu des prairies de posidonie. *Rapport Procès-Verbal Réunion Commission Internationale Exploration Scientifique Méditerranée*, 18(2), 91-92.

Pirrotta, M., Tomasello, A., Scannavino, A., Di Maida, G., Luzzu, F., Bellissimo, G., Bellavia, C., Costantini, C., Orestano, C., Sclafani, G., & Calvo, S. (2015). Transplantation assessment of degraded *Posidonia oceanica* habitats: Site selection and long-term monitoring. *Mediterranean Marine Science*, 16(3), 591–604.

Pisano, A., Marullo, S., Artale, V., Falcini, F., Yang, C., Leonelli, F. E., Santoleri, R., & Nardelli, B. B. (2020). New Evidence of Mediterranean Climate Change and Variability from Sea Surface Temperature Observations. *Remote Sensing*, 12(1), 132.

Plan Bleu (2022). État des lieux du tourisme en Méditerranée, projet communautaire Interreg Med sur le tourisme durable.

Platt, T., Gallegos, C.L., & Harrison, W.G. (1980). Photoinhibition of photosynthesis in natural assemblages of marine phytoplankton. *Journal of Marine Research*, 38, 687–701.

Poole, P., Ramachandran, V. & Terpolilli, J. (2018). Rhizobia: from saprophytes to endosymbionts. *Nature Reviews Microbiology*, 16, 291–303.

Prambauer, M., Wendeler, C., Weitzenböck, J., & Burgstaller, C. (2019). Biodegradable geotextiles – An overview of existing and potential materials. *Geotextiles and Geomembranes, 47*(1), 48–59.

Préfecture maritime de Méditerranée, 2016. Arrêté Préfectoral N°155/2016 réglementant le mouillage des navires dans les eaux intérieures et territoriales françaises de Méditerranée [WWW Document]. https://www.premar-mediterranee.gouv.fr/arretes.html (accessed 18.2.24)

Préfecture maritime de Méditerranée, 2019. Arrêté Préfectoral N°123/2019 réglementant le mouillage des navires dans les eaux intérieures et territoriales françaises de Méditerranée [WWW Document]. https://www.premar-mediterranee.gouv.fr/arretes.html (accessed 18.2.24)

Procaccini, G., Dattolo, E., & Ruocco, M. (2023). Genetic diversity and connectivity in the Mediterranean seagrass *Posidonia oceanica*: state of art and future directions. *Cahiers de Biologie Marine*, 64, 105–114.

Pugnaire, F.I., Morillo, J.A., Peñuelas, J., Reich, P.B., Bardgett, R.D., Gaxiola, A., Wardle, D.A., & van der Putten, W.H. (2019). Climate change effects on plant-soil feedbacks and consequences for biodiversity and functioning of terrestrial ecosystems. *Science Advances*, 5(11), 1–12.

R

Raitsos, D.E., Beaugrand, G., Georgopoulos, D., Zenetos, A., Pancucci-Papadopoulou, A.M., Theocharis, A., & Papathanassiou, E. (2010). Global climate change amplifies the entry of tropical species into the eastern Mediterranean Sea. *Limnology and Oceanography*, 55(4), 1478–1484.

Ralph, P.J., Polk, S.M., Moore, K.A., Orth, R.J., & Smith, W.O. (2002). Operation of the xanthophyll cycle in the seagrass *Zostera marina* in response to variable irradiance. *Journal of Experimental Marine Biology and Ecology*, 271(2), 189-207.

Ralph, P.J., & Gademann, R. (2005). Rapid light curves: a powerful tool to assess photosynthetic activity. *Aquatic Botany*, 82, 222-237.

Raniello, R. & Procaccini, G. (2002). Ancient DNA in the seagrass Posidonia oceanica. *Marine Ecology Progress Series*, 227, 169-173.

Randell, A.S., Tanner, J.E., Wos-Oxley, M. L., Catalano, S.R., Keppel, G., & Oxley, A.P.A. (2023). The root of influence: root-associated bacterial communities alter resource allocation in seagrass seedlings. *Frontiers in Marine Science*, 10.

Rautenbach, S.A., Pieraccini, R., Nebel, K., & Engelen, A.H. (2024). Marine biodegradation of natural potential carrier substrates for seagrass restoration. *Marine Ecology*.

Reay, M.K., Charteris, A.F., Jones, D.L., & Evershed, R.P. (2019). ^{15}N-amino sugar stable isotope probing (^{15}N-SIP) to trace the assimilation of fertiliser-N by soil bacterial and fungal communities. *Soil Biology and Biochemistry*, 138, 107599.

Rende, S.F., Bosman, A., Menna, F., Lagudi, A., Bruno, F., Severino, U., Montefalcone, M., Irving, A. D., Raimondi, V., Calvo, S., Pergent, G., Pergent-Martinì, C., & Tomasello, A. (2022). Assessing seagrass restoration actions through a micro-bathymetry survey approach (Italy, Mediterranean Sea). *Water* , 14(8), 1285.

Rezek, R.J., Furman, B.T., Jung, R.P., Hall, M.O., & Bell, S.S. (2019). Long-term performance of seagrass restoration projects in Florida, USA. *Scientific Reports*, 9(1), 1–11.

Richir, J., & Gobert, S. (2014). A reassessment of the use of *Posidonia oceanica* and *Mytilus galloprovincialis* to biomonitor the coastal pollution of trace elements: New tools and tips. *Marine Pollution Bulletin*, 89(1–2), 390–406.

Roca, G., Romero, J., Columbu, S., Farina, S., Pagès, J.F., Gera, A., Inglis, G., & Alcoverro, T. (2014). Detecting the impacts of harbour construction on a seagrass habitat and its subsequent recovery. *Ecological Indicators*, 45, 9–17.

Roca, G., Alcoverro, T., Krause-Jensen, D., Balsby, T. J. S., Van Katwijk, M. M., Marbà, N., Santos, R., Arthur, R., Mascaró, O., Fernández-Torquemada, Y., Pérez, M., Duarte, C. M., & Romero, J. (2016). Response of seagrass indicators to shifts in environmental stressors: A global review and management synthesis. *Ecological Indicators*, 63, 310–323.

Roca, G., Palacios, J., Ruíz-Halpern, S., & Marbà, N. (2022). Experimental carbon emissions from degraded Mediterranean seagrass (*Posidonia oceanica*) meadows under current and future summer temperatures. *Journal of Geophysical Research: Biogeosciences*, 127(9).

Rodrigues, R.R., Lima, R.A., Gandolfi, S., & Nave, A.G. (2009) On the restoration of high diversity forests: 30 years of experience in the Brazilian Atlantic Forest. *Biological Conservation*, 142, 1242–1251.

Rolando, J.L., Kolton, M., Song, T., Liu, Y., Pinamang, P., Conrad, R., Morris, J. T., Konstantinidis, K.T., & Kostka, J.E. (2024). Sulfur oxidation and reduction are coupled to nitrogen fixation in the roots of the salt marsh foundation plant *Spartina alterniflora*. *Nature Communications*, 15(1).

Romero, J., Perez, M., Mateo, M.A., & Sala, E. (1994). The belowground organs of the Mediterranean seagrass *Posidonia oceanica* as a biogeochemical sink. *Aquatic Botany*, 47, 13–19.

Rubio-Portillo, E., Martin-Cuadrado, A.-B., Ramos-Esplá, A.Á., & Antón, J. (2021). Metagenomics Unveils *Posidonia oceanica* "Banquettes" as a Potential Source of Novel Bioactive Compounds and Carbohydrate Active Enzymes (CAZymes). *MSystems*, 6(5).

Ruiz-Jaen, M.C., & Aide, T.M. (2005). Restoration success: How is it being measured? *Restoration Ecology*, 13(3), 569–577.

Ruiz, J.M., Perez, M. & Romero, J. (2001). Effects of fish farm loading on seagrass (*Posidonia oceanica*) distribution, growth and photosynthesis. *Marine Pollution Bulletin*, 42, 749-760.

Ruiz, J.M., Romero, J. (2001). Effects of in situ experimental shading on the Mediterranean seagrass *Posidonia oceanica*. *Marine Ecology Progress Series*, 215, 107–120.

Ruiz, J.M., & Romero, J. (2003). Effects of disturbances caused by coastal constructions on spatial structure, growth dynamics and photosynthesis of the seagrass *Posidonia oceanica*. *Marine Pollution Bulletin*, 46(12), 1523–1533.

Ruiz, J.M., & Romero, J. (2003). Effects of in situ experimental shading on the Mediterranean seagrass *Posidonia oceanica*. *Marine Ecology Progress Series*, 215, 107-120.

Ruju, A., Ibba, A., Porta, M., Buosi, C., Passarella, M., & De Muro, S. (2018). The role of hydrodynamic forcing, sediment transport processes and bottom substratum in the shoreward development of *Posidonia oceanica* meadow. *Estuarine, Coastal and Shelf Science*, 212, 63–72.

S

Saaty, T.L. (2008). Decision making with the analytic hierarchy process. *International Journal of Services Sciences*, 1(1), 83–98.

Salinas, C., Duarte, C.M., Lavery, P.S., Masque, P., Arias-Ortiz, A., Leon, J.X., Callaghan, D., Kendrick, G.A., & Serrano, O. (2020). Seagrass losses since mid-20th century fuelled CO_2 emissions from soil carbon stocks. *Global Change Biology*, 26(9), 4772–4784.

Sánchez-Lizaso, J.L., Fernández-Torquemada, Y., & González-Correa, J.M. (2009). Evaluation of the viability of *Posidonia oceanica* transplants associated with a marina expansion. *Botanica Marina*, 52, 471–476.

Saunders, M.I., Doropoulos, C., Bayraktarov, E., Babcock, R.C., Gorman, D., Eger, A.M., Vozzo, M.L., Gillies, C.L., Vanderklift, M.A., Steven, A.D.L., Bustamante, R.H., & Silliman, B.R. (2020). Bright spots in coastal marine ecosystem restoration. *Current Biology*, 30(24), 1500–1510.

Saura, S. (2002). Effects of minimum mapping unit on land cover data spatial configuration and composition. *International Journal of Remote Sensing*, 23, 4853–4880.

Scannavino, A., Pirrotta, M., Tomasello, A., Di Maida, G., Luzzu, F., Bellavia, C., Bellissimo, G., Costantini, C., Orestano, C., Sclafani, G., & Calvo, S. (2014). In Proceedings of the 5th Mediterranean Symposium on Marine Vegetation, Portorož, Slovenia, 27–28 October 2014.

Scholz, V.V., Martin, B.C., Meyer, R., Schramm, A., Fraser, M.W., Nielsen, L.P., Kendrick, G.A., Risgaard-Petersen, N., Burdorf, L.D.W., & Marshall, I.P.G. (2021). Cable bacteria at oxygen-releasing roots of aquatic plants: a widespread and diverse plant-microbe association. *The New Phytologist*, 232(5), 2138–2151.

Sebastián, M., Ortega-Retuerta, E., Gómez-Consarnau, L., Zamanillo, M., Álvarez, M., Arístegui, J., & Gasol, J.M. (2021). Environmental gradients and physical barriers drive the basin-wide spatial structuring of Mediterranean Sea and adjacent eastern Atlantic Ocean prokaryotic communities. *Limnology and Oceanography*, 66(12), 4077–4095.

Secretariat of the Convention on Biological Diversity (2010). Global Biodiversity Outlook 3. Montréal (94p).

Seddon, N., Daniels, E., Davis, R., Chausson, A., Harris, R., Hou-Jones, X., Huq, S., Kapos, V., Mace, G.M., Rizvi, A.R., Reid, H., Roe, D., Turner, B., & Wicander, S. (2020). Global recognition of the importance of nature-based solutions to the impacts of climate change. *Global Sustainability*, 3.

Segata, N., Izard, J., Waldron, L., Gevers, D., Miropolsky, L., Garrett, W.S., & Huttenhower, C. (2011). Metagenomic biomarker discovery and explanation. *Genome Biology*, 12(1).

SER (Society for Ecological Restoration, Science and Policy Working Group). (2004). The SER international primer on ecological restoration. SER, Tucson, Arizona. Available from https://cdn.ymaws.com/www.ser.org/resource/resmgr/custompages/publications/SER_Primer/ser_primer.pdf (accessed 15 March 2022).

Serrano, O., Mateo, M.A., Renom, P., & Julià, R. (2012). Characterization of soils beneath a *Posidonia oceanica* meadow. *Geoderma*, 185–186, 26–36.

Serrano, O., Lavery, P.S., Rozaimi, M., & Mateo, M.Á. (2014). Influence of water depth on the carbon sequestration capacity of seagrasses. *Global Biogeochemical Cycles*, 28(9), 950–961.

Sharma, A., Kumar, V., Shahzad, B., Ramakrishnan, M., Singh Sidhu, G. P., Bali, A. S., Handa, N., Kapoor, D., Yadav, P., Khanna, K., Bakshi, P., Rehman, A., Kohli, S. K., Khan, E. A., Parihar, R. D., Yuan, H., Thukral, A. K., Bhardwaj, R., & Zheng, B. (2020). Photosynthetic response of plants under different abiotic stresses: A review. *Journal of Plant Growth Regulation*, 39(2), 509–531.

Short, F.T., & Wyllie-Echeverria, S. (1996). Natural and human-induced disturbance of seagrasses. *Environmental Conservation*, 23(1), 17–27.

Short, F., Carruthers, T., Dennison, W., & Waycott, M. (2007). Global seagrass distribution and diversity: A bioregional model. *Journal of Experimental Marine Biology and Ecology*, 350(1–2), 3–20.

Shugart, H.H.Jr. (2024). Phenomenon of succession. In S. A. Levin (Ed.), *Encyclopedia of biodiversity*, 6, pp. 272–281.

Skliris, N. (2014). Past, present and future patterns of the thermohaline circulation and characteristic water masses of the mediterranean sea. The Mediterranean Sea: Its History and Present Challenges, 9789400767, 29–48.

Skovhus, T.L., Ramsing, N.B., Holmström, C., Kjelleberg, S., & Dahllöf, I. (2004). Real-time quantitative PCR for assessment of abundance of *Pseudoalteromonas* species in marine samples. *Applied and Environmental Microbiology*, 70(4), 2373–2382.

Sleeman, J.C., Kendrick, G.A., Boggs, G.S., Hegge, B.J., 2005. Measuring fragmentation of seagrass landscapes: which indices are most appropriate for detecting change? *Marine and Freshwater Research*, 58, 851–864.

Smale, D.A., Wernberg, T., Oliver, E.C.J., Thomsen, M., Harvey, B.P., Straub, S.C., Burrows, M.T., Alexander, L.V., Benthuysen, J.A., Donat, M.G., Feng, M., Hobday, A.J., Holbrook, N.J., Perkins-Kirkpatrick, S.E., Scannell, H.A., Sen Gupta, A., Payne, B.L., & Moore, P.J. (2019). Marine heatwaves threaten global biodiversity and the provision of ecosystem services. *Nature Climate Change*, 9, 306–312.

Soga, M., & Gaston, K.J. (2018). Shifting baseline syndrome: causes, consequences, and implications. *Frontiers in Ecology and the Environment*, 16(4), 222–230.

Sogin, E.M., Michelhod, D., Gruber-Vodicka, H.R., Bourceau, P., Geier, B., Meier, D.V., Seidel, M., Ahmerkamp, S., Schorn, S., D'Angelo, G., Procaccini, G., Dubilier, N., & Liebeke, M. (2022). Sugars dominate the seagrass rhizosphere. *Nature Ecology & Evolution*, 6, 866–877.

Somot, S., Sevault, F., & Déqué, M. (2006). Transient climate change scenario simulation of the Mediterranean Sea for the twenty-first century using a high-resolution ocean circulation model. *Climate Dynamics*, 27(7–8), 851–879.

Spalding, M.D., Ruffo, S., Lacambra, C., Meliane, I., Hale, L. Z., Shepard, C.C., & Beck, M.W. (2014). The role of ecosystems in coastal protection: Adapting to climate change and coastal hazards. *Ocean and Coastal Management*, 90, 50–57.

Stewart-Sinclair, P.J., Purandare, J., Bayraktarov, E., Waltham, N., Reeves, S., Statton, J., Sinclair, E.A., Brown, B.M., Shribman, Z.I., & Lovelock, C.E. (2020). Blue Restoration – Building Confidence and Overcoming Barriers. *Frontiers in Marine Science*, 7, 748.

Stipcich, P., Pansini, A., Beca-Carretero, P., Stengel, D. B., & Ceccherelli, G. (2022a). Field thermo acclimation increases the resilience of *Posidonia oceanica* seedlings to marine heat waves. *Marine Pollution Bulletin*, 184, 114230.

Stipcich, P., Apostolaki, E.T., Chartosia, N., Efthymiadis, P.T., Jimenez, C.E., La Manna, G., Pansini, A., Principato, E., Resaikos, V., & Ceccherelli, G. (2022b). Assessment of *Posidonia oceanica* traits along a temperature gradient in the Mediterranean Sea shows impacts of marine warming and heat waves. *Frontiers in Marine Science*, 9.

Stipcich, P., Resaikos, V., & Ceccherelli, G. (2023). Experimental thermocline deepening highlights the resilience of the seagrass *Posidonia oceanica*: An opportunity to investigate shoot adaptability. *Marine Pollution Bulletin*, 189.

Stipcich, P., La Manna, G. & Ceccherelli, G. (2024). Warming-induced flowering and fruiting in the seagrass *Posidonia oceanica* and uncertainties due to context-dependent features. *Marine Biology*, 171, 67.

Stramska, M., & Aniskiewicz, P., (2019). Recent large scale environmental changes in the Mediterranean Sea and their potential impacts on *Posidonia Oceanica*. *Remote Sensing*, 11, 110.

Suding, K. (2011). Toward an Era of Restoration in Ecology: Successes, Failures, and Opportunities Ahead. *Annual Review of Ecology, Evolution and Systematics*, 42, 465-487.

Sun, Y., Mazzotta, M.G., Miller, C.A., Apprill, A., Izallalen, M., Mazumder, S., Perri, S.T., Edwards, B., Reddy, C.M., & Ward, C.P. (2023). Distinct microbial communities degrade cellulose diacetate bioplastics in the coastal ocean. *Applied and Environmental Microbiology*, 89(12).

Sun, J., Zhao, Q., Gao, Y.-N., Long, Q.-G., Yan, W.-J., & Zhang, P.-D. (2024). Restoration of degraded seagrass meadows: Effects of plant growth-promoting rhizobacteria (PGPR) inoculation on *Zostera marina* growth, rhizosphere microbiome and ecosystem functionality. *Journal of Environmental Management*, 371, 123286.

Sureda, A., Box, A., Terrados, J., Deudero, S., & Pons, A. (2008). Antioxidant response of the seagrass *Posidonia oceanica* when epiphytized by the invasive macroalgae *Lophocladia lallemandii*. *Marine Environmental Research*, 66(3), 359–363.

Sutera, A., Bonaviri, C., Spinelli, P., Carimi, F., & De Michele, R. (2024). Fruit encasing preserves the dispersal potential and viability of stranded *Posidonia oceanica* seeds. *Scientific Reports*, 14(1).

T

Tarquinio, F., Hyndes, G.A., Laverock, B., Koenders, A., & Säwström, C. (2019). The seagrass holobiont: understanding seagrass-bacteria interactions and their role in seagrass ecosystem functioning. *FEMS Microbiology Letters*, 366(6), 57.

Tasdemir, D., Scarpato, S., Utermann-Thüsing, C., Jensen, T., Blümel, M., Wenzel-Storjohann, A., Welsch, C., & Echelmeyer, V.A. (2024). Epiphytic and endophytic microbiome of the seagrass *Zostera marina*: Do they contribute to pathogen reduction in seawater? *Science of the Total Environment*, 908.

Telesca, L., Belluscio, A., Criscoli, A., Ardizzone, G., Apostolaki, E. T., Fraschetti, S., Gristina, M., Knittweis, L., Martin, C.S., Pergent, G., Alagna, A., Badalamenti, F., Garofalo, G., Gerakaris, V., Louise Pace, M., Pergent-Martini, C., & Salomidi, M. (2015). Seagrass meadows (*Posidonia oceanica*) distribution and trajectories of change. *Scientific Reports*, 5(1), 12505.

Temmerman, S., Meire, P., Bouma, T.J., Herman, P.M.J., Ysebaert, T., & De Vriend, H.J. (2013). Ecosystem-based coastal defence in the face of global change. *Nature*, 504(7478), 79–83.

Temmink, R.J.M., Christianen, M.J.A., Fivash, G.S., Angelini, C., Boström, C., Didderen, K., Engel, S.M., Esteban, N., Gaeckle, J.L., Gagnon, K., Govers, L.L., Infantes, E., van Katwijk, M.M., Kipson, S., Lamers, L.P.M., Lengkeek, W., Silliman, B. R., van Tussenbroek, B.I., Unsworth, R.K. F., …, van der Heide, T. (2020). Mimicry of emergent traits amplifies coastal restoration success. *Nature Communications*, 11(1), 1–9.

Templado, J. (2014). Future trends of mediterranean biodiversity. The Mediterranean Sea: Its History and Present Challenges, 479–498.

Terrados, J., Marin, A., & Celdran, D. (2013). Use of *Posidonia oceanica* seedlings from beach-cast fruits for seagrass planting. *Botanica Marina*, 56, 185–195.

Thomas, F., Giblin, A.E., Cardon, Z.G., & Sievert, S.M. (2014). Rhizosphere heterogeneity shapes abundance and activity of sulfur-oxidizing bacteria in vegetated salt marsh sediments. *Frontiers in Microbiology*, 5.

Tomasello, A., Di Maida, G., Calvo, S., Pirrotta, M., Borra, M., & Procaccini, G. (2009). Seagrass meadows at the extreme of environmental tolerance: The case of *Posidonia oceanica* in a semi-enclosed coastal lagoon. *Marine Ecology*, 30(3), 288–300.

Touchette, B.W., & Burkholder, J.A.M. (2000). Review of nitrogen and phosphorus metabolism in seagrasses. *Journal of Experimental Marine Biology and Ecology*, 250(1–2), 133–167.

Trevathan-Tackett, S.M., Seymour, J.R., Nielsen, D.A., Macreadie, P.I., Jeffries, T.C., Sanderman, J., Baldock, J., Howes, J.M., Steven, A.D.L., & Ralph, P.J. (2017). Sediment anoxia limits microbial-driven seagrass carbon remineralization under warming conditions. *FEMS Microbiology Ecology*, 93(6).

Trevathan-Tackett, S.M., Sherman, C.D. H., Huggett, M.J., Campbell, A.H., Laverock, B., Hurtado-McCormick, V., Seymour, J.R., Firl, A., Messer, L.F., Ainsworth, T., Negandhi, K.L., Daffonchio, D., Egan, S., Engelen, A. H., Fusi, M., Thomas, T., Vann, L., Hernandez-Agreda, A., ... Macreadie, P. I. (2019). A horizon scan of priorities for coastal marine microbiome research. *Nature Ecology & Evolution, 3*(11), 1509–1520.

Trevathan-Tackett, S.M., Jeffries, T.C., Macreadie, P.I., Manojlovic, B., & Ralph, P. (2020). Long-term decomposition captures key steps in microbial break-down of seagrass litter. *Science of the Total Environment*, 705, 135806.

Tu, T.-H., Lin, E.-J., Hung, C.-C., Chou, W.-C., & Shih, Y.-Y. (2025). The dissolved oxygen variation in seagrasses is influenced by DOC excretion and its associated microbes. *Estuarine, Coastal and Shelf Science*, 313, 109080.

Turner, R. K., Burgess, D., Hadley, D., Coombes, E., & Jackson, N. (2007). A cost-benefit appraisal of coastal managed realignment policy. *Global Environmental Change*, 17(3–4), 397–407.

Turschwell, M.P., Connolly, R.M., Dunic, J.C., Sievers, M., Buelow, C.A., Pearson, R.M., Tulloch, V.J.D., Côté, I.M., Unsworth, R.K.F., Collier, C.J., & Brown, C.J. (2021). Anthropogenic pressures and life history predict trajectories of seagrass meadow extent at a global scale. *Proceedings of the National Academy of Sciences*, 118(45), e2110802118.

U

Udy, J.W., & Dennison, W.C. (1997). Growth and physiological responses of three seagrass species to elevated sediment nutrients in Moreton Bay, Australia. *Journal of Experimental Marine Biology and Ecology*, 217(2), 253–277.

Uhrin, A.V., & Turner, M.G. (2018). Physical drivers of seagrass spatial configuration: the role of thresholds. *Landscape Ecology*, 33(12), 2253–2272.

Uitz, J., Claustre, H., Gentili, B., & Stramski, D. (2010). Phytoplankton class-specific primary production in the world's oceans: Seasonal and interannual variability from satellite observations. *Global Biogeochemical Cycles*, 24(3).

UNEP (United Nations Environment Programme) (2021). Becoming #GenerationRestoration: Ecosystem restoration for people, nature and climate. Nairobi.

United Nations Environment Programme (2021).Conference of the Parties to the Convention on Biological Diversity. Fifteenth Meeting (Part I) Kunming Declaration "Ecological Civilization: Building a Shared Future for All Life on Earth."

UNEP/MAP: State of the Mediterranean Marine and Coastal Environment, UNEP/MAP – Barcelona Convention, Athens, 2012.

United Nations Environment Agency (2019). Resolution Adopted by the General Assembly on 1 March 2019 73/284. United Nations Decade on Ecosystem Restoration (2021-2030).

United Nations Environment Programme, 2019. A new deal for nature–restore the degradedplanet.https://www.unep.org/resources/policy-and-strategy/new-deal-nature

Unsworth, R.K.F., Nordlund, L.M., & Cullen-Unsworth, L.C. (2019). Seagrass meadows support global fisheries production. *Conservation Letters*, 12(1).

Unsworth, R.K.F., Cullen-Unsworth, L.C., Jones, B.L.H., & Lilley, R.J. (2022). The planetary role of seagrass conservation. *Science*, 377(6606), 609–613.

Unsworth, R.K.F., Jones, B.L.H., Bertelli, C.M., Coals, L., Cullen-Unsworth, L.C., Mendzil, A.F., Rees, S.C., Taylor, F., Walter, B., & Evans, A.J. (2024). Ten golden rules for restoration to secure resilient and just seagrass social-ecological systems. *Plants, people, planet*, 7(1), 33-48.

V

Vacchi, M., De Falco, G., Simeone, S., Montefalcone, M., Morri, C., Ferrari, M., & Bianchi, C.N. (2017). Biogeomorphology of the Mediterranean *Posidonia oceanica* seagrass meadows. *Earth Surface Processes and Landforms*, 42(1), 42–54.

Valdez, S.R., Zhang, Y.S., van der Heide, T., Vanderklift, M.A., Tarquinio, F., Orth, R. J., & Silliman, B.R. (2020). Positive

Ecological Interactions and the Success of Seagrass Restoration. *Frontiers in Marine Science*, 7, 91.

Van Der Heide, T., Govers, L.L., De Fouw, J., Olff, H., Van Der Geest, M., Van Katwijk, M.M., Piersma, T., Van De Koppel, J., Silliman, B.R., Smolders, A.J.P., & Van Gils, J.A. (2012). A three-stage symbiosis forms the foundation of seagrass ecosystems. *Science*, 336(6087), 1432–1434.

Van der Heide, T. (2017).The fundamental role of ecological feedback mechanisms for the adaptive management of seagrass ecosystems—A review. *Biological Reviews of the Cambridge Philosophical Society,* 92, 1521–1538.

Van der Heide, T., Temmink, R.J.M., Fivash, G.S., Bouma, T.J., Didderen, K., Esteban, N., Gaeckle, J., Gagnon, K., Infantes, E., Lengkeek, W., Unsworth, R., & Christianen, M.J.A. (2021). Coastal restoration success via emergent trait-mimicry is context dependent. *Biological Conservation*, 264 (109373).

van der Putten, W.H., Bradford, M.A., Pernilla Brinkman, E., van de Voorde, T.F.J., & Veen, G.F. (2016). Where, when and how plant– soil feedback matters in a changing world. *Functional Ecology*, 30(7), 1109– 1121.

Vangeluwe, D., Lepoint, G., Bouquegneau, J.M., & Gobert, S. (2004). Effect of transplantation on *Posidonia oceanica shoots*. *Vie et Milieu*, 54(4), 223–230.

Vangeluwe, D. (2006). Effets de la transplantation sur la biométrie et sur la dynamique des nutriments, du carbone et de la chlorophylle de *Posidonia oceanica* (L.) Delile. Thèse de doctorat. Université de Liège, Liège (Belgique).

van Katwijk, M.M., Thorhaug, A., Marbà, N., Orth, R.J., Duarte, C.M., Kendrick, G.A., Althuizen, I.H.J., Balestri, E., Bernard, G., Cambridge, M.L., Cunha, A., Durance, C., Giesen, W., Han, Q., Hosokawa, S., Kiswara, W., Komatsu, T., Lardicci, C., Lee, K.S., … Verduin, J.J. (2016). Global analysis of seagrass restoration: the importance of large-scale planting. *Journal of Applied Ecology*, 53(2), 567–578.

Ventura, D., Mancini, G., Casoli, E., Pace, D.S., Lasinio, G.J., Belluscio, A., & Ardizzone, G. (2022). Seagrass restoration monitoring and shallow-water benthic habitat mapping through a photogrammetry-based protocol. *Journal of Environmental Management*, 304.

Ventura, D., Grosso, L., Pensa, D., Casoli, E., Mancini, G., Valente, T., Scardi, M., & Rakaj, A. (2023). Coastal benthic habitat mapping and monitoring by integrating aerial and water surface low-cost drones. *Frontiers in Marine Science*, 9.

Vessey, J.K. (2003). Plant growth promoting rhizobacteria as biofertilizers. *Plant and Soil*, 255, 571-586.

Vidondo, B., Duarte, C.M., Middelboe, A.L., Stefansen, K., Lutzen, T., & Nielsen, S.L. (1997). Dynamics of a landscape mosaic: size and age distributions, growth and demography of seagrass *Cymodocea nodosa* patches. *Marine Ecology Progress Series*, 158, 131–138.

Vieira, V.M.N.C.S., Santos, R., Leitão-Silva, D., Veronez, A., Neves, J.M., Nogueira, M., Brito, A., Cereja, R., Creed, J.C., Bertelli, C. M., Samper-Villarreal, J., Bartilotti, C., & Lobo-Arteaga, J. (2024). Seagrass space occupation efficiency is key for their role as ecosystem engineers and ecological indicators. *Communications Earth and Environment*, 5(1).

Vohník, M., Borovec, O., Kolaříková, Z., Sudová, R., & Réblová, M. (2019). Extensive sampling and high-throughput sequencing reveal *Posidoniomyces atricolor* gen. Et sp. Nov. (Aigialaceae, Pleosporales) as the dominant root mycobiont of the dominant Mediterranean seagrass *Posidonia oceanica*. *MycoKeys*, 55, 59–86.

W

Waltham, N.J., Elliott, M., Lee, S.Y., Lovelock, C., Duarte, C.M., Buelow, C., Simenstad, C., Nagelkerken, I., Claassens, L., Wen, C.K.C., Barletta, M., Connolly, R. M., Gillies, C., Mitsch, W.J., Ogburn, M.B., Purandare, J., Possingham, H., & Sheaves, M. (2020). UN Decade on Ecosystem Restoration 2021–2030—What Chance for Success in Restoring Coastal Ecosystems? *Frontiers in Marine Science*, 7, 71.

Wang, L., Tomas, F., & Mueller, R. S. (2020). Nutrient enrichment increases size of *Zostera marina* shoots and enriches for sulfur and nitrogen cycling bacteria in root-associated microbiomes. *FEMS Microbiology Ecology*, 96(8), 1–14.

Wang, L., English, M.K., Tomas, F., & Muellera, R.S. (2021). Recovery and community succession of the *Zostera marina* rhizobiome after transplantation. *Applied and Environmental Microbiology*, 87(3), 1–16.

Ward, E.A., Meek, S.K., Gordon, D.M., Cameron, T.C., Steer, M.D., Smith, D.J., Miliou, A., & Tsimpidis, T. (2020). The use of storm fragments and biodegradable replanting methods allows for a low-impact habitat restoration method of seagrass meadows, in the eastern Aegean Sea. *Conservation Evidence*, 17, 1–6.

Warner, B.G., Clymo, R.S., & Tolonen, K. (1993). Implications of peat accumulation at point escuminac, New Brunswick. *Quarternary Research*, 39, 245–248.

Waycott, M., Duarte, C.M., Carruthers, T.J. B., Orth, R.J., Dennison, W.C., Olyarnik, S., Calladine, A., Fourqurean, J.W., Heck, K. L., Hughes, A.R., Kendrick, G.A., Kenworthy, W.J., Short, F.T., & Williams, S.L. (2009). Accelerating loss of seagrasses across the globe threatens coastal ecosystems. *Proceedings of the National Academy of Sciences*, 106(30), 12377–12381.

Wear, R. J., Tanner, J. E., & Hoare, S. L. (2010). Facilitating recruitment of *Amphibolis* as a novel approach to seagrass rehabilitation in hydrodynamically active waters. *Marine and Freshwater Research*, 61(10), 1123–1133.

Welsh, D.T. (2000). Nitrogen fixation in seagrass meadows: Regulation, plant–bacteria interactions and significance to primary productivity. *Ecology Letters*, 3(1), 58–71.

Wentworth, C.K. (1922). A scale of grade and class terms for clastic sediments. *Journal of Geology*, 30, 377–392.

Williams, S.L., Ambo-Rappe, R., Sur, C., Abbott, J.M., & Limbong, S.R. (2017). Species richness accelerates marine ecosystem restoration in the coral triangle. *Proceedings of the National Academy of Sciences of the United States of America*, 114, 11986–11991.

Wolff, T. (1976). Utilization of seagrass in the deep sea. *Aquatic Botany*, 2, 161-174.

Wood, G., Marzinelli, E.M., Coleman, M.A., Campbell, A.H., Santini, N.S., Kajlich, L., Verdura,J., Wodak, J., Steinberg, P.D., & Vergés, A. (2019). Restoring subtidal marine macrophytes in the Anthropocene: trajectories and future-proofing. *Marine and Freshwater Research*, 70 (7).

Wortley, L., Hero, J.M., & Howes, M. (2013). Evaluating Ecological Restoration Success: A Review of the Literature. *Restoration Ecology*, 21(5), 537–543.

Wright, J.P., & Jones, C.G. (2006). The concept of organisms as ecosystem engineers ten years on: progress, limitations, and challenges. *Bioscience*, 56(3), 203–209.

Y

Yemn, E.W., & Willis, A.J. (1954). The estimation of carbohydrates in plant extracts by anthrone. *Biochemical Journal,* 57, 508–514.

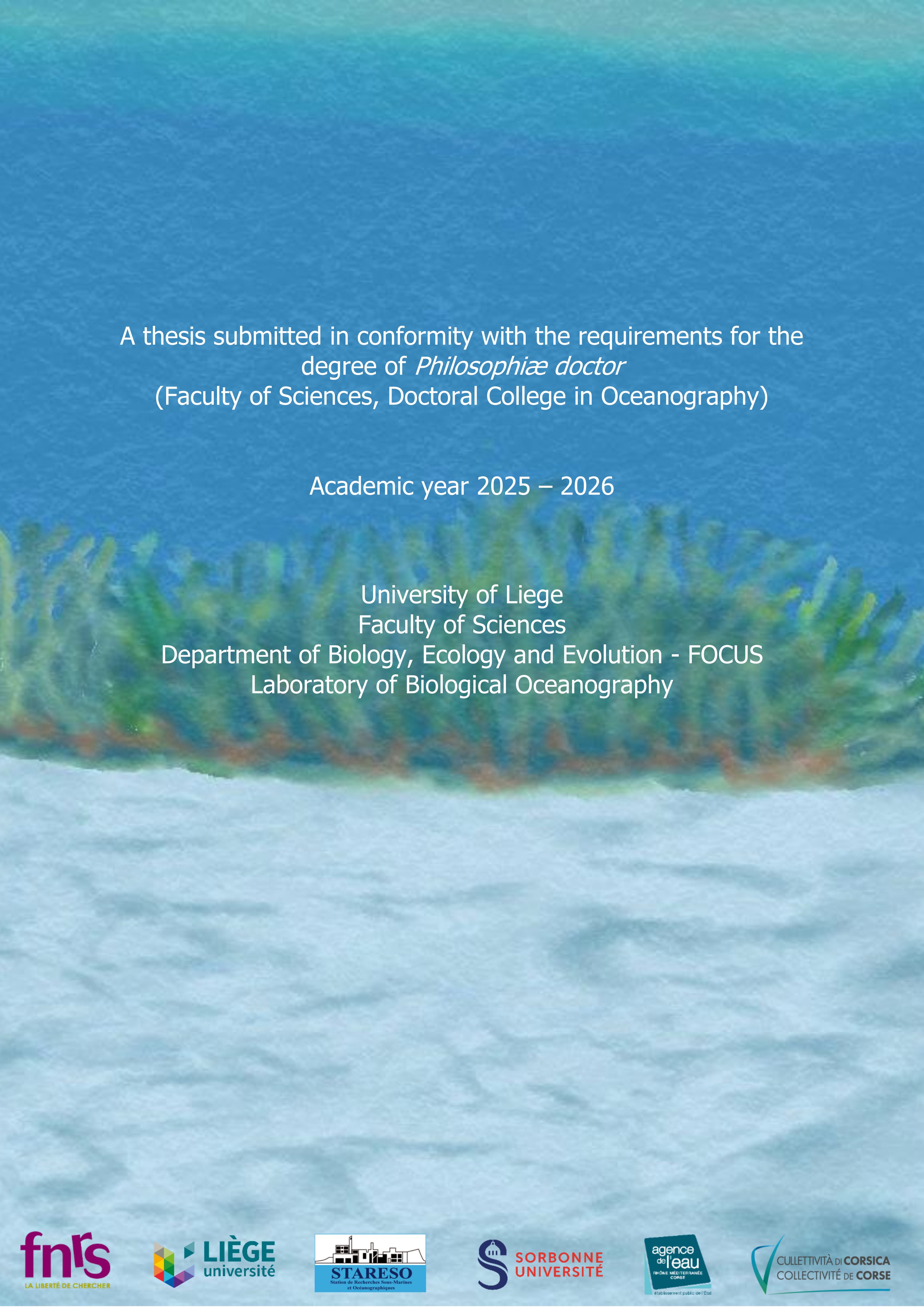
Z

Zenone, A., Giacalone, V.M., Martinez, M., Pipitone, C., Alagna, A., Infantes, E., D'Anna, G., & Badalamenti, F. (2025). Stitching up *Posidonia oceanica* (*L.*) Delile anchorage scars using beach-cast seeds: Results of a six-year study. *Biological Conservation, 303*, 111032.

Zhang, X., Wu, Y., Liu, S., Li, J., Jiang, Z., Luo, H., & Huang, X. (2024). Plant growth and development of tropical seagrass determined rhizodeposition and its related microbial community. *Marine Pollution Bulletin*, 199, 115940.

Zhou, W., Ling, J., Shen, X., Xu, Z., Yang, Q., Yue, W., Liu, H., Suo, A., & Dong, J. (2024). Inoculation with plant growth-promoting rhizobacteria improves seagrass *Thalassia hemprichii* photosynthesis performance and shifts rhizosphere microbiome. *Marine Environmental Research*, 193.

Zimmerman, R.C., Smith, R.D., & Alberte, R.S. (1989). Thermal acclimation and whole-plant carbon balance in *Zostera marina* L. (eelgrass). *Journal of Experimental Marine Biology and Ecology,* 130, 93–109.



A thesis submitted in conformity with the requirements for the
degree of *Philosophiæ doctor*
(Faculty of Sciences, Doctoral College in Oceanography)

Academic year 2025 – 2026

University of Liege
Faculty of Sciences
Department of Biology, Ecology and Evolution - FOCUS
Laboratory of Biological Oceanography

fnrs LA LIBERTÉ DE CHERCHER
LIÈGE université
STARESO Station de Recherches Sous-Marines et Océanographiques
SORBONNE UNIVERSITÉ
agence de l'eau RHÔNE MÉDITERRANÉE CORSE établissement public de l'État
CULLETTIVITÀ DI CORSICA COLLECTIVITÉ DE CORSE