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

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

[a] Laboratory of Oceanology, University of Liege [b] STAtion de REcherche Sous-marines et Océanographiques (STARESO) [c] Centro de Ciências do Mar (CCMAR), Universidade do Algarve [d] Microbial Systems Ecology, University of Amsterdam

*Corresponding author: arnaud.boulenger@uliege.be

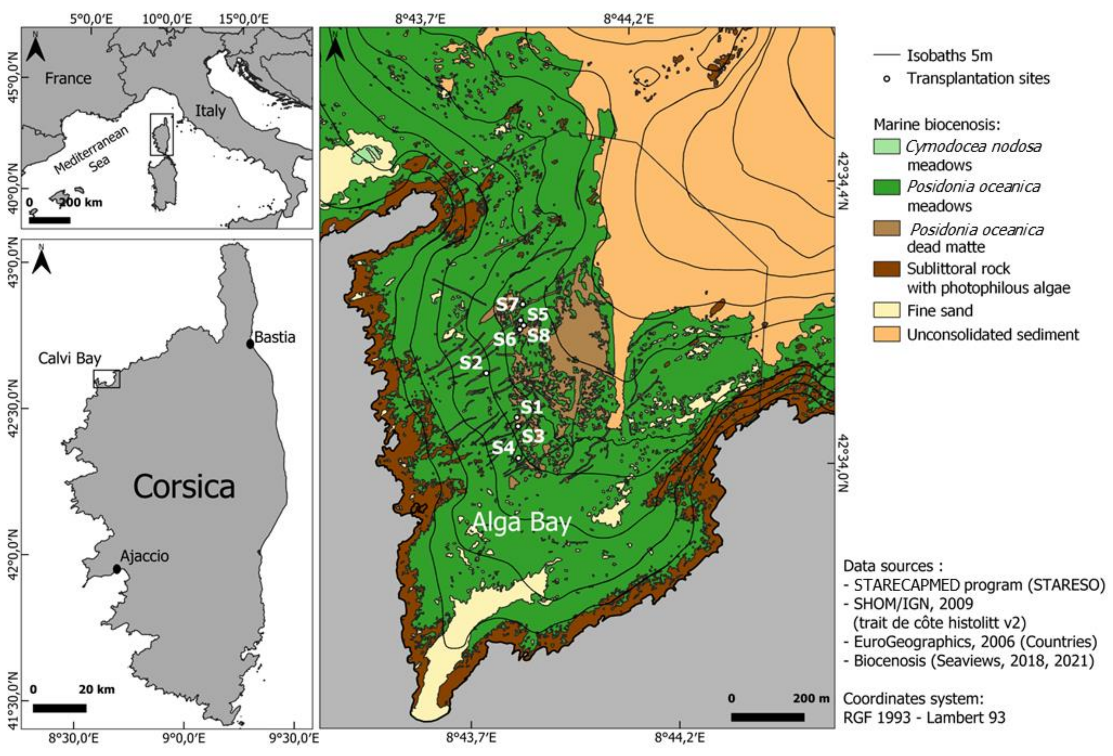
Background

- *P. oceanica* meadows are regressing due to anthropogenic pressures, like anchoring and coastal development.
- Transplantation-based restoration has been explored for decades as a management and mitigation strategy, yet the role of the plant-associated microbiome in restoration success remains largely unknown.

Which transplantation method yields bacterial communities most similar to the control meadows ?

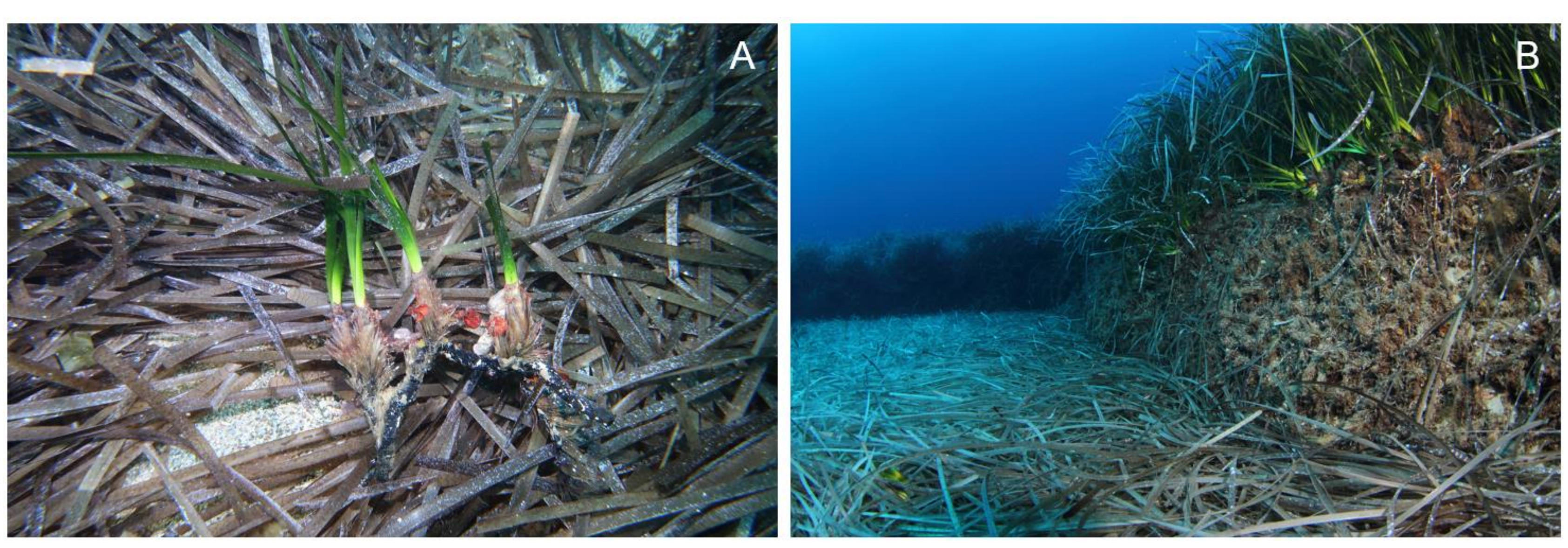
Which donor origin harbours bacterial communities most similar to the control meadows ?

Methods

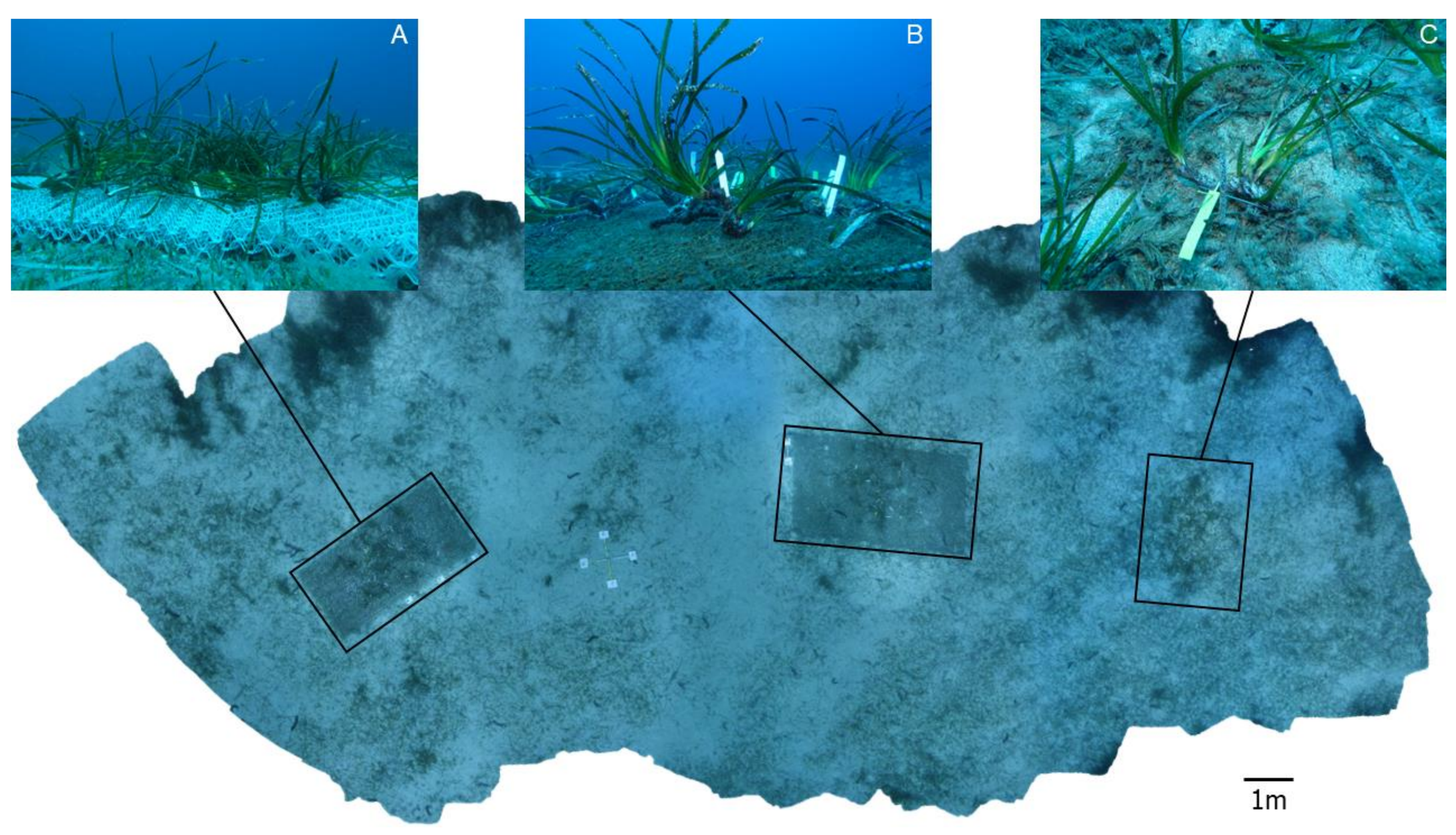


- *P. oceanica* cuttings transplantation on dead matte from boat anchoring degradation in Alga Bay (Corsica)
- 792 plagiotropic fragments transplanted in Spring 2022
- Sampling in Spring 2024 for microbiome analysis by 16S rRNA gene amplicon sequencing

Cuttings from two **donor origins**: (A) Naturally uprooted fragments (i.e. **storm-fragments**), and (B) fragments manually harvested from the erosion side of natural sandy intermattes (i.e. **intermatte cuttings**).



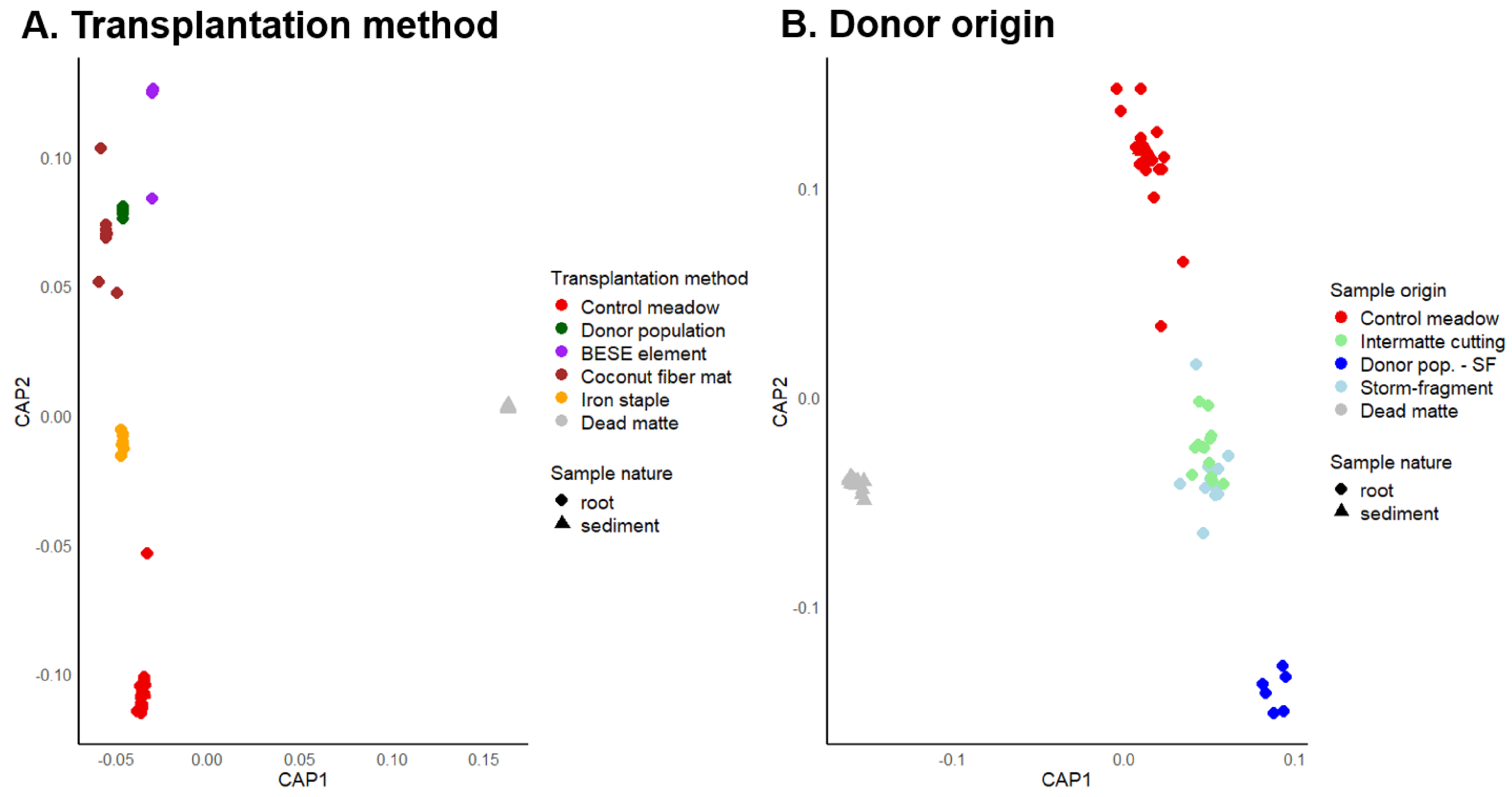
Three different **transplantation methods**: (A) **BESE elements**, (B) **coconut fiber mats**, and (C) **iron staples**.



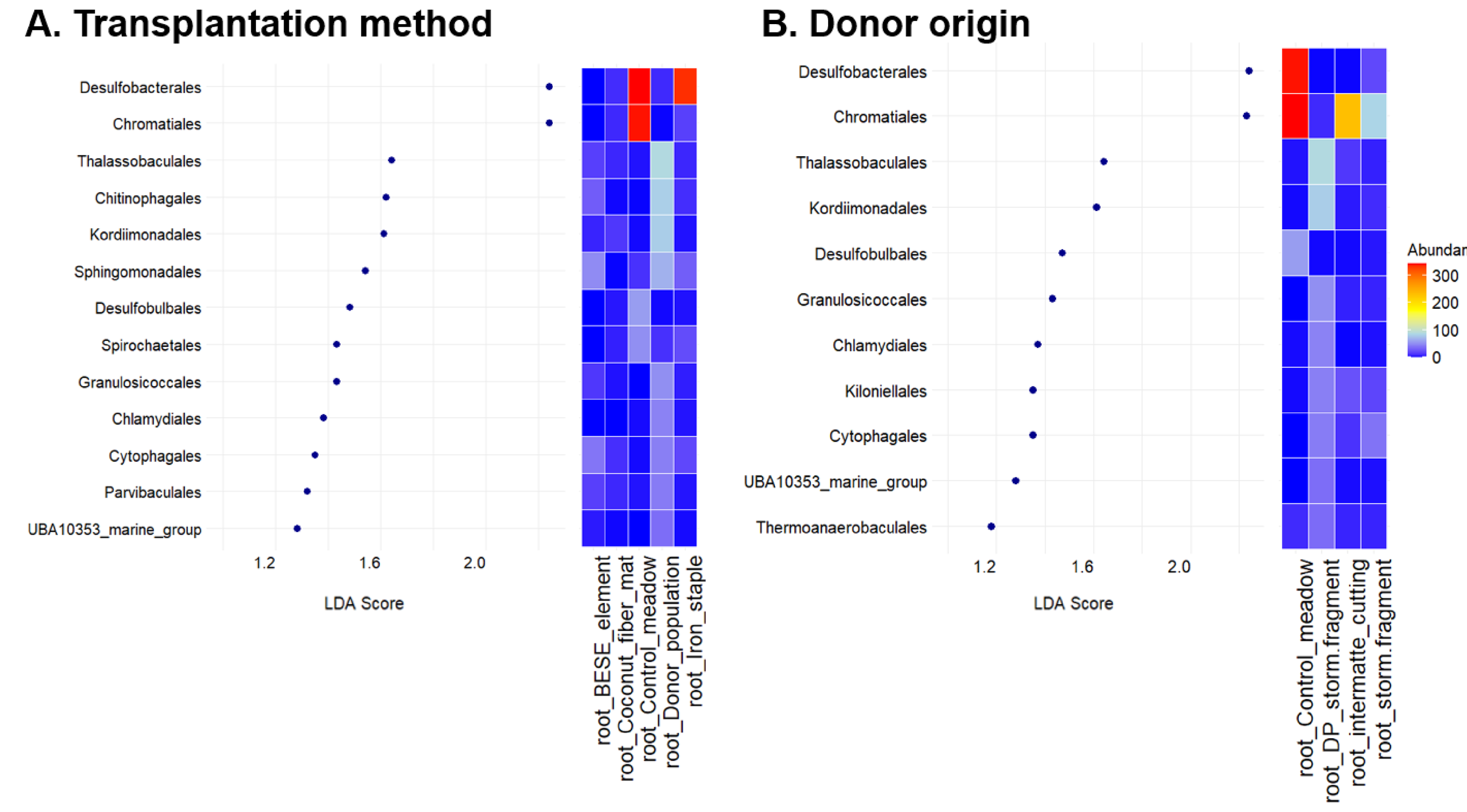
Results

A. The iron staples are more similar to the control meadows compared to the two other transplantation methods.

B. The cuttings transplanted two years ago are more similar to the control meadows compared to their original populations.



Desulfobacterales and ***Chromatiales*** are key bacterial orders in differentiating between control meadows and transplanted cuttings.



Key findings

Our results show that transplantation methods strongly shape the bacterial communities in the seagrass roots. Iron staples promoted bacterial assemblages like those in control meadows, likely due to direct sediment interaction contact facilitating efficient microbial recruitment. In contrast, the physical separation from the sediment imposed by coconut fiber mats and BESE elements delayed microbial stabilization.

Donor populations also influenced the bacterial dynamics; intermatte cuttings showed higher abundances of *Chromatiales* (e.g., genus *Candidatus* Thiodiazotropha), which are associated with sulfur oxidation and nitrogen fixation.

Despite initial differences, transplants progressively recruited microbiomes resembling those of the control meadows, demonstrating the potential for long-term microbial stabilization. These findings underscore the need to integrate microbiome considerations into *P. oceanica* restoration practices to enhance restoration success and improve ecosystem function recovery.

Acknowledgements

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's work is supported by the Ministry of Agriculture, Fisheries, Food Security and Nature through the BIODIVERSA project RESTORESEAS. 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.