

eDNA Metabarcoding for Biodiversity Assessment in *Posidonia oceanica* Meadows: A Multi-Marker, Multi-Matrix Approach to Support Marine Spatial Planning

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The integration of comprehensive, high-resolution spatio-temporal data is critical for effective Marine Spatial Planning (MSP), as traditional monitoring methods are often too slow, invasive, or localized to guide adaptive management. The urgent need to align MSP with conservation objectives—such as the 30×30 target—necessitates the adoption of non-invasive, high-throughput technologies.

Environmental DNA (eDNA) metabarcoding has emerged as a powerful tool for characterizing marine biodiversity, offering strong potential to inform MSP and adaptive management. By coupling water and sediment matrices, it captures complementary signals, from transient, waterborne genetic material to more persistent benthic archives, enabling comprehensive biodiversity assessments suitable for conservation prioritization.

We present results from two seasonal sampling campaigns (November 2024, June 2025) conducted at ten sites along the W coast of Sardinia (Italy), in *Posidonia oceanica* meadows within the "Penisola del Sinis - Isola di Mal di Ventre" Marine Protected Area (MPA), adjacent Special Areas of Conservation and proposed ecological corridors. Water and sediment samples were processed using five molecular markers (18S V4, COI Leray, 12S MiFishU, Tele02, Elas02) targeting eukaryotes, metazoans and vertebrates, and analysed through a tiered taxonomic assignment workflow.

This approach recovered an unprecedentedly rich biodiversity dataset, with each marker-matrix combination capturing distinct and largely non-overlapping windows of diversity. Water eDNA reflected mobile and pelagic communities and showed clear seasonal turnover, while sediment eDNA provided more temporally stable signals from benthic fauna taxa. Detections included flagship and indicator species, overexploited commercial fish, Mediterranean endemics, IUCN-listed threatened taxa, invasive non-native species, and harmful algal bloom organisms.

Forthcoming functional diversity analyses and protection-gradient comparisons across different areas will further inform on the effect of protection and the role of corridors. Overall, our findings demonstrate that integrated multi-marker, multi-matrix eDNA monitoring provides a robust, cost-effective and repeatable evidence base for refining MPA zonation and operationalising biodiversity mainstreaming within MSP.