



Mediterranean
Action Plan
Barcelona
Convention



#MedSymposia

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7th MEDITERRANEAN SYMPOSIUM ON MARINE VEGETATION

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United Nations Environment Programme
Mediterranean Action Plan
Specially Protected Areas Regional Activity Centre
Boulevard du Leader Yasser Arafat
B.P.337 - 1080 Tunis Cedex - TUNISIA
car-asp@spa-rac.org

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**Proceedings of the
7th Mediterranean Symposium
on Marine Vegetation**

FORWARD

Dear Colleagues,

The United Nations Environment Programme Mediterranean Action Plan – Specially Protected Areas Regional Activity Centre (UNEP/MAP – SPA/RAC) – Barcelona Convention has initiated since year 2000 a series of scientific symposia, dedicated to key habitats and NIS by organising the first Mediterranean Symposium on Marine vegetation. This initiative while aiming essentially to take stock of the recently available scientific data and to promote the cooperation between specialists and key actors working in the Mediterranean, has evolved since then to cover now Coralligenous, Dark Habitats and Non-Indigenous species as well.

These symposia have been initiated as implementation of the UNEP/MAP regional action plans related to (i) **the Conservation of Marine Vegetation in the Mediterranean Sea** (adopted by the Contracting Parties to the Barcelona Convention in 1999 and updated in 2012), (ii) **the Conservation of Coralligenous and other calcareous bio-concretions of Mediterranean** (adopted by the Contracting Parties to Barcelona Convention in 20018 and updated in 2016), (iii) **the conservation of habitats and species associated with seamounts, underwater caves and canyons, aphotic hard beds and chemo-synthetic phenomena in the Mediterranean Sea** (Action Plan for Dark Habitats adopted by the Contracting Parties to the Barcelona Convention in 2013), and (iv) the **Action Plan concerning Species Introduction and Invasive Species** (Adopted by the Contracting Parties to the Barcelona Convention in 2003 and updated in 2016).

The “Mediterranean Symposia on Marine Key Habitats and NIS” are an important output, not only of the UNEP/MAP Medium-Term Strategy for the period 2022-2027 (Decision IG.525/1), but also for NTZ/MPA project “Empowering the legacy: scaling up co-managed and financially sustainable No-Take Zones / Marine Protected Areas”, financed by MAVA foundation under its Mediterranean Strategy and by the EU-funded IMAP-MPA project “Towards achieving the Good Environmental Status of the Mediterranean Sea and Coast through an Ecologically Representative and Efficiently Managed and Monitored Network of Marine Protected Areas” .

For more than two decades, the symposia have provided, within the framework of the MAP Barcelona Convention, a platform for dialogue between the scientific community, managers, and decision makers.

This year, SPA/RAC in collaboration with the Italian Institute for Environmental Protection and Research (ISPRA), the university of Genoa and its Department of Earth, the Environment and Life Sciences of the University of Genoa (DISTAV) and the association “Società Italiana di Biologia Marina” (SIBM), organized a new edition of the Mediterranean Symposia in Genoa, from 19 to 23 September 2022, as follows:

- 7th Mediterranean Symposium on Marine Vegetation (19-20 September 2022)
- 4th Mediterranean Symposium on the conservation of coralligenous and other calcareous bio-concretions (20-21 September 2022)
- 3rd Mediterranean Symposium on the conservation of dark habitats (21-22 September 2022)
- 2nd Mediterranean Symposium on the non-indigenous species (22-23 September 2022)

This edition was also a good opportunity to discuss new topics such as monitoring and definition of Good Environmental Status (GES), monitoring and assessment scale in the Mediterranean and, in this way, contribute to enhancing Science-Policy interface and to strengthening links and cooperation between SPA/RAC and Barcelona Convention system and scientists and scientific institutions in the Mediterranean.

Khalil ATTIA
SPA/RAC Director

Tatjana Hema
UNEP/MAP Coordinator

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PROGRAMME

Monday 19 September 2022

- 8:00-9:00** Participants welcome and registration
- 9:00-10:00** **Opening of the Mediterranean Symposia on Marine Key Habitats and Non-Indigenous Species**
- Opening speeches**
- Prof. Laura CANESI, vice-director of DISTAV
 - Mr. Khalil ATTIA, Director, UNEP/MAP-SPA/RAC
 - Mme Tatjana HEMA, UNEP/MAP Coordinator
 - Her Excellency, Mme Leila CHIKHAOUI MAHDAOUI, Minister of Environment, Tunisia (video message)
- 10:00-10:15** *Coffee break*
- 10:15-10:30** **Keynote conference: "Posidonia oceanica meadow: A major carbon sink in the Mediterranean Sea "** by Gérard PERGENT
- Session 1: Mapping and monitoring (and associated organisms)**
Chair: Monica MONTEFALCONE, Rapporteur: Yassine Ramzi SGHAIER
- 10:30-10:45** "Participative mapping and aerial photographs help to show the strong decline of north eastern *Posidonia oceanica* seagrass beds (French Occitanie region)" by Julie DETER, HOUNGNANDAN F., BENKHABCHECHE F., HARDY P.-Y., GUILBERT A., TRIBOT A.-S., POURTAU M., SUTTON G., BARROIL A., HOLON F., BOISSERY P.
- 10:45-11:00** "A new map atlas of coastal marine biodiversity" by Thomas BOCKEL, BOISSERY P., DELARUELLE G., DETER J., GERVAISE C., HOLON F.
- 11:00-11:15** "3-dimensional mapping for fine-scale cartography of *Posidonia oceanica* seagrass meadow limits with underwater structure from motion photogrammetry" by Daniele VENTURA, CASOLI E., MANCINI G., BELLUSCIO A., PACE D. S., ARDIZZONE G.D.
- 11:15-11:30** "A hierarchical scale-based approach for inferring the ecological quality of a marine ecosystem from its spatial composition and configuration" by Fabrice HOUNGNANDAN, FERY C., BOCKEL T., DELARUELLE G., BOISSERY P., KEFI S., DETER J.
- 11:30-11:45** "Involving managers in the ecosystem-based assessment of marine habitats: a case study in french catalonia)" by Patrick ASTRUCH, SCHOHN T., BELLONI B., CASSETTI O., CABRAL M., RUITTON S., MICHEZ N., MASINSKI I., HARTMANN V., BOUDOURESQUE C.F.
- 11:45-12:00** "Indices from the past: relevance in the status assessment of *Posidonia oceanica* meadows" by Alice OPRANDI, MANCINI I., BIANCHI C.N., MORRI C., AZZOLA A., MONTEFALCONE M.

- 12:00-12:15** **"Benthic habitats id and policy cross-links to improve marine conservation"** by Marie **LA RIVIERE**, DELAVENNE J., DE BETTIGNIES T., MICHEZ N., JANSON A.-L., ANDRES S., GAUDILLAT V.
- 12:15-12:30** **"Need of reference conditions for comparable monitoring of *Posidonia oceanica* at eco-regional scale including the sliding baseline syndrome: a discussion over Foça SEPA"** by Onur **KARAYALI**, AKÇALI B., BİZSEL K.C., KABOĞLU G.
- 12:30-12:45** Discussion
- 12:45-14:30** ***Lunch break***
- Session 2:** **Impacts and disturbances threatening key Mediterranean formations (including global change)**
Chair: Ali **BADREDDINE**, Rapporteur: Atef **LIMAM**
- 14:30-14:45** **"*Posidonia oceanica* restoration, a relevant strategy after boat anchoring degradation?"** by Christine **PERGENT-MARTINI**, ACUNTO S., ANDRÉ S., BARRALON E., CALVO S., CASTEJÓN-SILVO I., CULIOLI J.M., LEHMANN L., MOLENAAR H., MONNIER B., OBERTI P., PEY A., PIAZZI L., SANTONI M.C., TERRADOS J., TOMASELLO A., PERGENT G.
- 14:45-15:00** **"AIS data help to manage anchoring and its impact on French seagrass meadows"** by Thomas **BOCKEL**, HOLON F., MARRE G., DELARUELLE G., BOISSERY P., DETER J
- 15:00-15:15** **"Towards the restoration of canopy-forming brown algae in the Mediterranean Sea: major challenges and wins"** by Valentina **ASNAGHI**, CHIANTORE M., KALEB S., DE LA FUENTE G., SRIJEMSI M., SAVONITTO G., CLAUSING R., CIRIACO S., MEROTTO L., MARCHESE F., KASTELIC L., SEGARICH M., FALACE A.
- 15:15-15:30** **"The perception of *Posidonia oceanica* banquettes by beachgoers in the French Riviera"** by Charles-François **BOUDOURESQUE**, MARINIER M., CAVALIER M., DRIANCOURT T., BLANFUNÉ A., PERRET-BOUDOURESQUE M., THIBAUT T.
- 15:30-15:45** **"*Ex situ* pilot experiment to assess the feasibility of ecological restoration of *Fucus virsoides* in the Gulf of Trieste (Northern Adriatic)"** by Sara **KALEB**, DESCOURVIÈRES E., BANDELJ V., CIRIACO S., ORLANDO-BONACA M., FALACE A.
- 15:45-16:00** ***Coffee break***
- 16:00-16:15** **Welcome speech** by Federico DELFINO, Rector of the University of Genoa
- 16:15-17:15** **Poster Session**
- Session 2:** **Impacts and disturbances threatening key Mediterranean formations (including global change)**
(continued) Chair: Rym **ZAKHAMA**, Rapporteur: Mehdi **AISSI**
- 17:15-17:30** **"The influence of human activities on *Posidonia oceanica* at Tremiti Islands Marine Protected Area (Adriatic Sea, Italy)"** by Andrea **TURSI**,

MASTROTOTARO F., BOTTALICO A., DE GIOSA F., MONTESANTO F.,
LISCO A., CHIMIENTI G.

17:30-17:45 **"Deterioration of precious blue carbon ecosystems in a Natura 2000 site: the impact of anchorage in the seagrass meadows of the National Marine Park of Alonissos Northern Sporades"** by **Dimitris POURSANIDIS**, IOSSFIDIS S., VASSILOPOULOU V.

17:45-18:00 **"The trophic role of the necromass of *Posidonia oceanica* (L) Delile in the surf zone of sandy shores "** by **Francesca ROSSI**, ROMERO G., PEY A., VENTURA P., LOQUES F., DERIJARD B., COTTALORDA J.M., DI FRANCO E., GUIDETTI P., BUSSOTTI S.

18:00-18:15 **"Effect of filamentous macroalgae *Chaetomorpha linum* on *Cymodocea nodosa*: does clonal integration alleviate the magroalgae impacts?"** by **Imen ZRIBI**, ELLOUZI H., MNASRI I., ABEDELKADER N., BEN HMIDA A., DORAI S., DEBEZ A. CHARFI-CHEIKHROUHA F., ZAKHAMA-SRAIEB R.

18:15-18:30 **Discussion**

Tuesday 20 September 2022

8:00-9:30 **Roundtable on "Needs and expectations to reinforce protection and restoration at the regional level"**
Moderator: Christine PERGENT-MARTINI

9:30-9:45 **Awards for best oral communication and poster**

9:45-10:00 **Closure of the Symposium**

KEYNOTE CONFERENCE

G rard PERGENT, MONNIER B., PERGENT-MARTINI C.

EqEL, CNRS SPE 6134, Universit  de Corse, Campus Grimaldi, 20250 Corte, France.

E-mail: pergent_g@univ-corse.fr

POSIDONIA OCEANICA MEADOW: A MAJOR CARBON SINK IN THE MEDITERRANEAN SEA

Abstract

In the last decades, the increasing necessity to reduce atmospheric carbon dioxide (CO₂) concentrations has intensified interest in quantifying the capacity of coastal ecosystems to fix and sequester carbon, referred to commonly as ‘Blue Carbon’ (BC). Among coastal habitats, seagrass meadows are considered as natural carbon sinks due to their capacity to store large amounts of carbon in their sediments over long periods of time.

In the Mediterranean Sea, Posidonia oceanica constitutes extensive meadows considered to be one of the most effective ecosystems in carbon fixation and sequestration but above all, as long-term carbon sinks due to the development of an exceptional structure known as ‘matte’, reaching several meters in height, which can be preserved over millennia.

The synthesis of a hundred measurements provides a basis for estimating the average annual organic carbon (C_{org}) fixation at 1.30 t C_{org} ha⁻¹ yr⁻¹ and sequestration at 0.28 t C_{org} ha⁻¹ yr⁻¹. While this annual carbon fixation represents less than 1% on average of CO₂ releases for all Mediterranean countries, in the large Mediterranean islands the fixation is on average 3.1% and can reach almost 14.4% (Corsica). The mean stock in the top 250 cm of matte (matte mean thickness) was estimated at 723 ± 46 t C_{org} ha⁻¹. This value corresponds to several decades of carbon release accumulated in the matte for Mediterranean countries. Even by integrating inorganic carbon (C_{inorg}) stocks (source of carbon due to calcification) the balance remains positive and the P. oceanica meadow appears as a major carbon sink in the Mediterranean Sea. Nevertheless, the restoration of P. oceanica meadows (and Blue Carbon Ecosystems) with the aim of climate change mitigation must be used with caution due to questionable cost-effectiveness.

Key-words: Seagrass, Carbon sink, *Posidonia oceanica* meadow, Climate change

Context

The strategies to reduce greenhouse gas (GHG), and notably carbon dioxide (CO₂) concentrations, are crucial for climate change mitigation. From the Paris Agreement COP 21 – 2015 to the One Planet Summit for the Ocean – 2022, interest in natural carbon sinks and more especially Blue Carbon ecosystems has intensified (Fig. 1). These Nature-Based Solutions are the subject of widespread discussion and controversy as to their effectiveness and cost (Williamson and Gattuso, 2022).

Carbon fixation in the oceans (Blue Carbon - BC) accounts for more than half of the carbon fixed in the biosphere. Although it mainly corresponds to the action of phytoplankton, coastal vegetated ecosystems play a particular role despite their limited surface area (-0.5% of the surface area of the ocean), (Nellemann *et al.*, 2009). These habitats (mangrove forests, tidal salt marshes and seagrass meadows) fix more than 18% of the BC and store more than 50% as organic carbon (C_{org}) in their sediments for long periods of time.

Among these BC ecosystems, *Posidonia oceanica* plays a major role in the Mediterranean Sea with an extension estimated at more than 2 million hectares (Traganos *et al.*, 2022). If its carbon fixation capacity can be compared to that of terrestrial forests (around $5 \text{ t CO}_{2\text{eq}} \text{ ha}^{-1} \text{ yr}^{-1}$), the sequestration of this C_{org} , in a specific structure called 'matte', is much higher (between 20 and 25%) and it can be stored over millennia (Pergent-Martini *et al.*, 2021).



Fig. 1: Integration of coastal and marine vegetated ecosystems in the international discussions concerning mitigation of climate change.

This matte, constituted by intertwined rhizomes and roots filled with sediment, can reach several meters in height (Kendrick *et al.*, 2005; Fig. 2). As a result, the C_{org} storage below *P. oceanica* meadows has been shown to be 2 500 to 3 000 $\text{t CO}_{2\text{eq}} \text{ ha}^{-1}$ corresponding to the highest C_{org} storage in the soil for major ecosystem types (IUCN, 2021; Monnier *et al.*, 2022).



Fig. 2: Escarpments of *Posidonia oceanica* matte with living shoots.

Carbon fluxes in *Posidonia oceanica* meadows

P. oceanica meadows act as 'sponge' for C_{org} . The seagrass meadows capture this C_{org} from the water column during the photosynthesis process and sequesters it in the matte for a long period of time (centuries to millennia). Furthermore, *P. oceanica* contributes to the burial of inorganic carbon (C_{inorg}), mainly associated with calcifying organisms living within the meadow which may constitute a source of carbon (Fig. 3).

The synthesis of a hundred measurements made throughout the Mediterranean Sea and at depths between 0.5 and 32.0 m provides a basis for estimation of the average primary leaf production (Pergent-Martini *et al.*, 2021). Considering a depth of 15 m, which corresponds to

the average depth of *P. oceanica* meadows in the Mediterranean Sea (UNEP/MAP-RAC/SPA, 2015), the annual primary leaf production (blades and sheaths) is estimated at 343 g DW m⁻² (1.3 t C_{org} ha⁻¹ yr⁻¹), which corresponds to nearly 10 million t CO_{2eq} per year at Mediterranean scale.

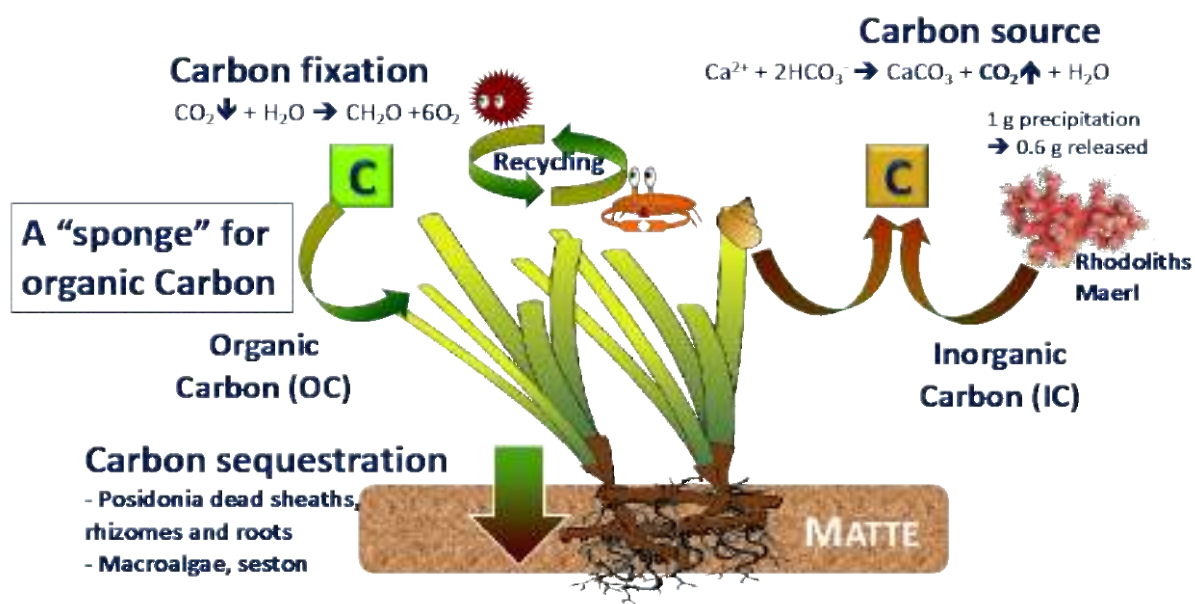


Fig 3: Conceptual model of carbon fluxes in a *Posidonia oceanica* meadow.

The balance between *Posidonia oceanica* fixation and CO₂ country emission varies considerably, with for example 0.12% for France (moderate *P. oceanica* surface area and very high CO₂ release) and 13.2% for Tunisia (very high *P. oceanica* surface area and low CO₂ release). The average for a Mediterranean country is a little less than 1% while this value exceeds 3% for the six main Mediterranean islands.

The sequestration of C_{org} in the matte results mainly from autochthonous (*i.e.* *Posidonia oceanica* dead leaf sheaths, rhizomes and roots, but also epiphytes) and allochthonous sources (*i.e.*, water column seston, adjacent ecosystems – macroalgae). The mean value is estimated at 0.28 t C ha⁻¹ yr⁻¹, that is to say more than 2 million t CO_{2eq} per year at Mediterranean scale (Pergent-Martini *et al.*, 2021).

Values for *P. oceanica* matte thickness were scarce in the Mediterranean Sea. A large-scale assessment was performed along 100 km of the east coast of Corsica (Fig. 4A). This estimate, achieved using geophysical seismic reflection equipment coupled with ground truthing and coring data (Monnier *et al.*, 2021), highlighted a mean matte thickness of 2.5 m (maximum value: 8.7 m). This value is similar to other values recorded by Leduc *et al.* (*in press*) along the west coast of Corsica (2.2 m) and throughout the Mediterranean (Lo Iacono *et al.*, 2008; Tomasello *et al.*, 2009).

Matte datation and carbon content were estimated, along the east coast of Corsica, after the sampling and analysis of 39 cores. The collection of cores was performed under a wide range of environmental conditions (water depth, depositional environment, sediment matrix), with a Kullenberg gravity corer during the oceanographic survey Carbonsink (R/V L'Europe, Ifremer). The mean age of *P. oceanica* matte, performed using radiocarbon (¹⁴C) analysis, was 3 967 cal. yr BP (maximum value: 9 073 cal. yr BP), attesting to *P. oceanica* meadows at the beginning of the Holocene period. The mean C_{org} and C_{inorg} stocks show a significant

variability with depth, sediment matrix and depositional environment. In the top 1.0 m of matte, the mean stock has been estimated at $327 \pm 15 \text{ t C}_{\text{org}} \text{ ha}^{-1}$ and $245 \pm 45 \text{ t C}_{\text{inorg}} \text{ m}^{-2}$ and reached $723 \pm 46 \text{ t C}_{\text{org}} \text{ ha}^{-1}$ and $605 \pm 99 \text{ t C}_{\text{inorg}} \text{ m}^{-2}$ in the top 2.5 m. The balance between C_{org} and C_{inorg} stock found in the top 1.0 and 2.5 m confirms that *P. oceanica* constitutes a net carbon sink (Fig. 4B, 4C). The upscaling of C_{org} stocks in the top 2.5 m of matte along the coastline of Corsica has led to an assessment of 37 million t C_{org} (135 million $\text{t CO}_{2\text{eq}}$) that is to say 77 years of CO_2 emissions by the Corsican population (www.globalcarbonatlas.org).

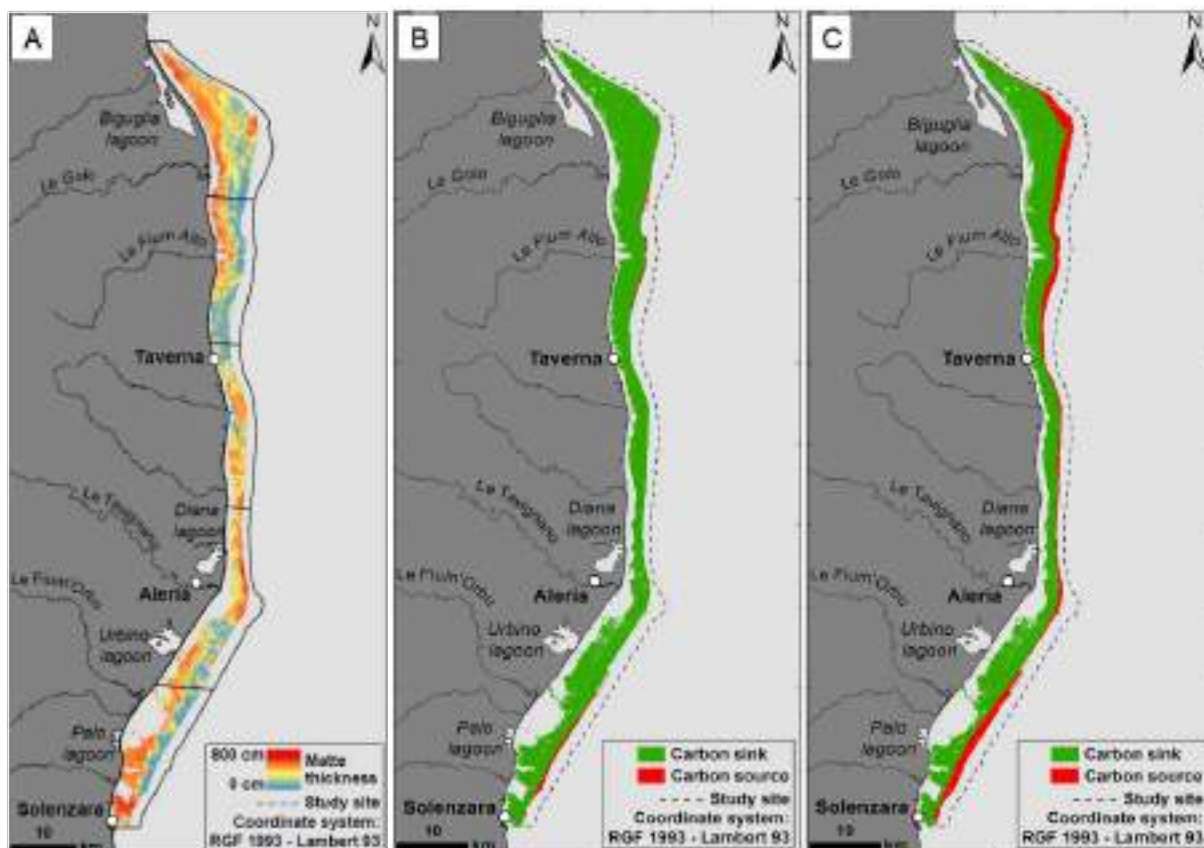


Fig. 4: Prediction map of the *Posidonia oceanica* matte thickness at the study site (A) and contribution of the top 100 cm (B) and 250 cm (C) of *Posidonia oceanica* matte as CO_2 sink (in green) or CO_2 source (in red) based on the balance between C_{org} and C_{inorg} stocks (Monnier *et al.*, 2022).

Take home message

In the present state of knowledge, five main points should be highlighted:

- *Posidonia oceanica* meadows fix and store a large amount of C_{org} and C_{inorg}
- *Posidonia oceanica* meadows act as a net $\text{CO}_{2\text{eq}}$ sink in the Mediterranean Sea
- Conservation of *Posidonia oceanica* meadows appears to be the best strategy to maintain this significant carbon stock and to avoid emissions (carbon dioxide, methane)
- Restoration of *Posidonia oceanica* meadows (and Blue Carbon Ecosystems) must be used with caution as a strategy for reducing climate change (questionable cost-effectiveness)
- Restoration of *Posidonia oceanica* meadows must be integrated within a global strategy integrating all associated ecosystem services and biodiversity

Acknowledgments

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Valentina ASNAGHI , CHIANTORE M., KALEB S., DE LA FUENTE G., SRIJEMSI M., SAVONITTO G., CLAUSING R., CIRIACO S., MEROTTO L., MARCHESE F., KASTELIC L., SEGARICH M., FALACE A.

DiSTAV - University of Genoa (Italy),

E-mail: falace@units.it

TOWARDS THE RESTORATION OF CANOPY-FORMING BROWN ALGAE IN THE MEDITERRANEAN SEA: MAJOR CHALLENGES AND WINS

Abstract

*In the Mediterranean, marine macroalgal forests of the genus *Cystoseira sensu lato* play a valuable role as foundation species. As there is evidence of decline or loss of these habitats due to direct and indirect human impacts, active restoration measures are promoted by European legislation. Within the EU project ROCPOP Life, ex situ restoration was used to repopulate two different *Cystoseira* s.l. species: *Ericaria amentacea* (C. Agardh) Molinari & Guiry and *Gongolaria barbata* (Stackhouse) Kuntze. This non-destructive method consists of three main steps: i) collecting the fertile apices in the donor sites, ii) growing the seedlings on tiles in mesocosms, and iii) outplanting the tiles with juveniles in the field. As the two target species thrive in different habitats (midlittoral for *E. amentacea* and subtidal for *G. barbata*), these species are exposed to different environmental pressures. Therefore, the implementation of ex situ restoration activities was designed according to the requirements of each species and the challenges of their environment. In the case of *E. amentacea*, the tiles with the juveniles were bolted directly onto rocks in the midlittoral, where heat, desiccation and wave exposure pose the greatest challenges. For *G. barbata*, floating structures suspended in the water column and multiple attachment devices with cages were used to avoid grazing, which was the biggest challenge at the recipient site. However, unpredictable challenges such as thermal anomalies, huge storms, and others were also tackled during the grow out phase. Nonetheless, relevant results were achieved regarding validation of the culture protocols, macroalgal growth, welfare and improvement of ecological quality, encouraging further development of this novel approach to marine biodiversity conservation.*

Key-words: ecological restoration, canopy-forming algae, *Ericaria amentacea*, *Gongolaria barbata*, ex situ outplanting, ROCPOP Life

Introduction

Coastal marine ecosystems are subject to multiple stressors such as climate change and local anthropogenic pressures that can lead to loss of biodiversity and functioning and may result in a degraded ecosystem state. Brown canopy-forming macroalgae, kelps and fucoids, play an important role in controlling spatial habitat heterogeneity, productivity and nutrient cycling in these coastal systems, where they create complex habitats with high biodiversity. However, the functioning of these foundation species is under threat, and restoration actions are strongly encouraged by European policies, as well as by the United Nations, which has declared 2021–2030 the Decade on Ecosystem Restoration. In the Mediterranean Sea, the most important canopy-forming algae are the species of the genus *Cystoseira sensu lato* (Fucales), but their decline and loss have been recorded throughout the northern Mediterranean coast (Mancuso *et al.*, 2018). As a result, these

species have become a target for restoration in this biogeographical region, where various non-destructive techniques such as enhancing recruitment potential *in situ* (Verdura *et al.*, 2018; Medrano *et al.*, 2020) or the deployment of laboratory-cultured juveniles on rocky coasts, i.e. *ex situ* outplanting (Sales *et al.*, 2011; De La Fuente *et al.*, 2019), are being used.

Within ROCPOP Life project, aiming to re-establish *Cystoseira s.l.* population at target locations along the Italian coast in order to enhance biodiversity and related ecosystem functions, the *ex situ* method has been chosen and adapted to the environmental challenges faced by the two target species. In the Ligurian Sea, *ex situ* outplanting restoration procedures were applied focusing on the intertidal *Ericaria amentacea* (C. Agardh) Molinari & Guiry. In the North Adriatic (Gulf of Trieste) the target species was *Gongolaria barbata* (Stackhouse) Kuntze. In the present manuscript, key considerations for a successful restoration of these species are discussed.

Material and methods

The *ex situ* outplanting of *E. amentacea* and *G. barbata* was performed in the Cinque Terre MPA (Ligurian Sea) and Miramare MPA (North Adriatic, Gulf of Trieste), respectively, in 4 consecutive reproductive seasons (2018-2021). Apical fronds were collected from healthy and fertile populations in the Portofino MPA (for the Ligurian Sea) and in the Landscape Park Strunjan (Slovenia, for the North Adriatic Sea). After collection, the apices were gently cleaned with tweezers and rinsed with filtered seawater to remove adhering biofouling and detritus. Cleaned apices were kept overnight in the dark and cold, after which spore release and culturing were conducted in environmentally controlled rooms, following the protocol described in Falace *et al.* (2018). Cultures were maintained in the laboratory of the University of Genoa and Trieste, for *E. amentacea* and *G. barbata*, respectively. As culture substrate, round clay tiles (4.5 cm in diameter) with a 0.6 cm hole at the centre (for later attachment with screws) were used. Von Stosch's enriched filtered seawater (VSE) was used as culture medium to enhance the growth rate of the algae, shorten the culture time and increase the size of the juveniles at the time of outplanting. After approximately three weeks of culturing, the clay tiles covered by *E. amentacea* and *G. barbata* juveniles were outplanted in the A Zone of Cinque Terre MPA and Miramare MPA, respectively. The deployment strategy differed for the two species: for the intertidal *E. amentacea* tiles were screwed individually to the rocks, while for the subtidal *G. barbata* dedicated anchoring structures were designed. In particular, the tiles were placed on prefabricated modular frames consisting of a 100×2.5cm flat perforated bar made of galvanised iron with four identical 50×2.5 cm bars arranged perpendicular to the first bar to form a rake-like shape. Each tile was fastened to the bar with a bolt at a fixed distance of about 5 cm from the other tiles, for a total of about 28 tiles per modular structure. The modular frames were specifically designed to mimic the spatial distribution pattern of adult *C. barbata* individuals at the donor site. Herbivore deterrents, consisting of two flexible metal mesh strips (3.5 × 20 cm, 0.5 cm mesh size) crossed in the middle to form an X shape, were added to modular frames. The protective structure was formed by bringing the ends of the strips around the tile like the petals of a flower, creating a cage-like structure around each tile. Another strategy tested to deter grazers was the use of collective cages (about 50 tiles protected by a single cage): the tiles were fixed on a metal support on a 50cm x 50cm concrete plate to which the cage was attached. Another alternative methodology used for the subtidal species was hanging cultivation *in situ* in a lantern net. The tiles were attached to a single-level plastic lantern net (55 cm in

diameter). This type of structure, suspended in the water column, is normally used for holding and fattening oysters and sea cucumbers. In the present study, this device was used as an intermediate step between culturing and outplanting, reducing rearing costs and helping avoid grazing in the field.

Results & Discussion:

Ex situ recruitment enhancement consists of three critical steps: 1) collection of fertile material from the donor site and transfer to hatcheries, 2) growing of juveniles in laboratory cultures, 3) transfer and outplanting at restoration sites. An additional step to be considered is monitoring over time to assess restoration success. The entire process should be ecologically sustainable and adaptable to the specific case studies (Fig. 1).



Species Habitat	Reproduction and Lab culture conditions	Outplanting challenges	Outplanting technique
 <p><i>Ericaria amentacea</i> Midlittoral High hydrodynamics</p>	<ul style="list-style-type: none"> Fertile apices in May-July Multiple tiny receptacles 20°C 125 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ Enriched Medium culture Antibiotics (if needed) 	<ul style="list-style-type: none"> Moisture Heat Desiccation Wave exposure 	<ul style="list-style-type: none"> Rough clay tiles Screws Single attachment Outplanting in strictly protected areas
 <p><i>Gongolaria barbata</i> Sublittoral Low hydrodynamics</p>	<ul style="list-style-type: none"> Reported fertile in April-May Single long subulate receptacles 14°C 125 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ Enriched Medium culture Antibiotics (if needed) 	<ul style="list-style-type: none"> Grazing (fish, molluscs) Sedimentation Heat waves 	<ul style="list-style-type: none"> Rough clay tiles Screws Multiple attachment Antigrazing cages Outplanting in strictly protected areas

Fig. 1: Summary of the main characteristics and challenges of the two *Cystoseira s.l.* species restored in the framework of the ROCPOP Life project

The location of the hatchery for juveniles culturing should be close to the collection site to ensure that healthy apices are brought to the laboratory. The Ligurian donor site is only a few kilometres (about 30 km) away from the laboratory of Genoa University. Landscape Park Strunjan, in the Adriatic Sea, is about 40 minutes away by boat and about an hour away by car. For this reason, transport by boat was initially chosen, which proved to be very fast and effective. Yet, in the framework of ROCPOP Life project, also a transport of apices to a more distant laboratory was tested: fertile apices collected in Punta Chiappa, Portofino MPA, were delivered to the facilities in Trieste (NE Italy) within 24 hours under dark, cold and humid conditions. This transport was successful and the apices were able to release gametes for the start of culture, with similar performance to transport over shorter distance.

A thorough knowledge of the target species is essential to select the optimal growing conditions (e.g., temperature, light intensity, photoperiod, nutrients) to achieve the maximum growth rate in the shortest possible time, thus reducing the time and cost of the process. Furthermore, accelerating the growth rate and achieving a higher yield in a short time brings several advantages: i) it is possible to carry out more than one cultivation during the limited fertile period, resulting in a higher number of recruits outplanted, thus providing the opportunity to restore larger areas; ii) seedlings are outplanted in the field

after they reach a refuge size (protection from at least some categories of grazers, i.e., micro- and meso-grazers such as molluscs or crustaceans), reducing vulnerability and consequently mortality; iii) larger seedlings facilitate monitoring during the initial phase in the field, an advantage that should not be overlooked as this is a crucial phase of restoration.

Under the ROCPOP Life project, six cultures of *E. amentacea* and six of *G. barbata* were performed over the 4 years of the project, and more than 100.000 juveniles of each species have been grown on tiles despite serious environmental issues affecting cultures and the COVID19 pandemic emergency.

After the culture period, when the recruits have reached the desired size, which may take 3-4 weeks, another delicate phase is the transport to the receiving site. The distance between the culturing lab and the receiving site, as well as the outside temperatures, must be considered. Optimal conditions must be ensured during transport to the collection site, especially in summer when air temperatures can reach 30-40 °C. In Liguria, the hatchery is about 80 km away from the receiving site (about 1 hour drive). A very high survival rate of juveniles on the tiles after transport was observed in all translocations performed in ROCPOP. A longer transport from Trieste to Cinque Terre (about 6 hours drive) was tested and was successful. Pictures of each tagged tile taken before leaving the laboratory and after relocation showed no differences in the percentage coverage of juveniles on the tiles before and after transport.

As for the deployment phase, in most restoration studies, whether *in situ* or *ex situ*, adults or tiles with juveniles are attached to the substrate with epoxy glue. However, this technique has several critical aspects: long condensation times, possible release of toxic compounds, mechanical stability, and unsightly aspects. The alternative proposed by ROCPOP Life is to use screws to attach the tiles. This method reduces the time and labour to attach tiles, the aesthetic impact on the rocky shore and strengthens the attachment, preventing possible slippage of the tiles due to wave action.

The environmental conditions of the site (e.g., currents, wave action) and the dispersal (gene flow) of the species used for restoration should be considered when planning the best planting pattern. The planting method and pattern should be species specific and may change significantly if restoration activities occur in the intertidal or subtidal zone.

In the intertidal zone, planting occurs by screwing tiles directly on the rocks within a long but narrow strip due to the microtidal conditions characteristic of most of the Mediterranean basin. In the Ligurian receiving site, the positioning height on the shore proved to be a decisive factor for the survival of the juveniles of *E. amentacea*: the survival rate after 12 weeks in the field on tiles placed in the lower zone was 84%, while only 10% of juveniles survived on tiles placed at a higher level.

In the subtidal zone, the planting pattern should consider a large area of substrate that may extend vertically through the zonation of the species, depending on the *Cystoseira s.l.* species. The use of modular structures for tiles attachment is envisaged given the difficulty of drilling holes by scuba diving. In the framework of ROCPOP Life, different types of modular frames for *G. barbata* were designed and tested in the Northern Adriatic also mitigate the effects of fish grazing, one of the major challenges in restoring subtidal macroalgae. The modular frames proved to be a suitable approach for outplanting subtidal *Cystoseira* species. Bolting the tiles onto the pre-assembled modular structures reduces the time, cost, and manpower requirements compared to other methods, thus offering great upscaling potential. On the other hand, the grazing deterrents, even if successful in protecting outplanted juveniles, require regular maintenance, making this approach not

sustainable for large-scale interventions. The use of suspended lantern net showed promising results, allowing to reduce cultivation time and efforts, enhancing juvenile growth and limiting the presence of grazers, which are typically found on vegetated rocky bottoms.

For large-scale restoration of *Cystoseira s.l.* the optimal pattern for planting should be planned in advance, taking into account the area of restoration and the number of restorations planned in the project. The method consists of planting patches of tiles with recruits that favour a spillover effect over time. In the first restoration effort, cores with recruits are placed far apart so that cores planted in subsequent years are placed between existing cores to close the gap. This method not only allows a large area to be covered, but also increases the success of a single restoration effort. Indeed, distributing recruits among multiple cores can reduce mortality caused by micro-local environmental conditions or biotic interactions (e.g., herbivores) compared to placing all recruits in a single patch.

Monitoring and evaluation are essential to determine if recovery projects are working as expected so that we achieve the intended benefits. Monitoring should be done both in the short and long term. Short term monitoring (< one year) is aimed at evaluating the efficacy of the chosen method, and it should be tailored depending on the ontogenetic phase of the restored individuals. We suggest that the monitoring frequency should be higher in the initial phases, e.g., biweekly, and could then be reduced to a monthly basis. For newly introduced individuals, it might be sufficient to quantify survival and growth rate. As the recruits reach a suitable size, other measurements should be added. Cover, density, morphological traits, and physiological metrics can be used to monitor their ability to adapt to the new environmental conditions generated from restoration. Monitoring measures are also important to develop the proper maintenance measures to control the abiotic and biotic factors that affect recruits at different life stages. An example is herbivory, producing negative effects in several subtidal transplantation experiments conducted in the Mediterranean. In addition, ecological quality should be evaluated after the implementation of the restoration measures and compared with the value previously determined. In the Cinque Terre MPA (particularly in Punta Mesco), ecological quality improved by one quality class after the restoration measures implemented by ROCPOP Life, from a good to a high ecological quality value (calculated with the CARLIT index). Long-term monitoring (at least until 3 - 5 years after the restoration action) aims to assess whether the introduced organisms are able to self-sustain. Fertility should be assessed at different times depending on the species restored. Indeed, different *Cystoseira* species reach the reproductive stage at different times: *E. amentacea* and *G. barbata*, for example, can become fertile after one year. Other monitoring activities should aim to assess changes in community structure (e.g., increase in number or type of species) or the appearance of new economically relevant species (fish or macroinvertebrates). Particular attention should be paid to detect new ecosystem services (e.g., water quality improvements, fisheries harvests, habitat provision, recreational activities such as snorkelling) that may contribute to human well-being.

If restoration has been successful, a new action can be planned. On the other hand, if an action was not successful, the results of monitoring can help to understand the causes of failure. If these are due to unexpected environmental factors, the recovery action should be completed. If the failure is due to methodological issues, efforts should be made to address them.

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Patrick ASTRUCH, SCHOHN T., BELLONI B., CASSETTI O., CABRAL M., RUITTON S., MICHEZ N., MASINSKI I., HARTMANN V., BOUDOURESQUE C.F.
GIS Posidonie, Aix-Marseille University, Marseille, France;
E-mail: patrick.astruch@univ-amu.fr

INVOLVING MANAGERS IN THE ECOSYSTEM-BASED ASSESSMENT OF MARINE HABITATS: A CASE STUDY IN FRENCH CATALONIA

Abstract

In the framework of the European Union LIFE 16IPE FR001 Marha project, Ecosystem-Based Quality Indices (EBQIs) provide a realistic assessment of the state and the functioning of ecosystems. The proposed EBQI was applied to four ecosystems of the north-western Mediterranean Sea: Posidonia oceanica Seagrass Meadows (POSM), Infralittoral Photophilous Reefs (IPR), Coralligenous Bioconcretions (CB), and Undersea Caves (UC). At over 29 sampling sites in the Natural Marine Park of the Gulf of Lion and in the Natural Marine Reserve of Cerbère-Banyuls (French Catalonia), results on POSM, IPR and CB have shown clear differences in ecosystem quality according to the management strategy and human activities. Results also highlighted the value of collaboration between scientists and managers, which provided useful data at both local MPA scale and regional scale (Monitoring Network). These data are useful for improving and adapting the initial protocols of the EBQI indices to enable their implementation in different ecological contexts and areas. Further investigations are obviously needed in other areas that are particularly influenced by global warming, different levels of anthropogenic pressures and related community shift issues.

Key-words: Ecosystem-based management, Ecosystem-Based Quality Index (EBQI), French Catalonia, Marine Protected Areas, ecosystem functioning

Introduction

Effective management of Marine Protected Areas (MPAs) calls for appropriate methods and descriptors to understand the functioning and the status of natural habitats and guide the decision-making process, particularly within the Mediterranean Sea. The Marine Strategy Framework Directive (MSFD, 2008/56/EC) aims to maintain or improve the quality of marine ecosystems. One of the pillars of the MSFD is the ecosystem-based approach (EBA), an integrated concept that considers ecosystems functioning and related services for management purposes. The EBA may be considered as a milestone in the progress of humans' approach to environmental issues (Boudouresque *et al.*, 2020). In that context, the aim of the European Union LIFE 16IPE FR001 Marha (Marine habitats) project is to improve the conservation status of marine habitats of European interest in France, according to the Habitats Directive (92/43/EC). To assess this conservation status, the EBA is a suitable tool for the purpose of properly characterising the structure and functioning of marine habitats. Several indicators were developed and have been applied in the frame of the Marha project: Ecosystem-Based Quality Indices (EBQI) developed for *Posidonia oceanica* Seagrass Meadows (POSM, Personnic *et al.*, 2014), Coralligenous Bioconcretions (CB, Ruitton *et al.*, 2014), Undersea Caves (Rastorgueff *et al.*, 2015), and Infralittoral Photophilous Reefs (IPR, Thibaut *et al.*, 2017). Managers at two local MPAs were trained to apply the EBQIs for that purpose: (i) the Natural Marine Reserve of Cerbère-Banyuls (NMRCB; French Catalonia, France), and (ii) The Natural Marine Park

of the Gulf of Lion (NMPGL, France). NMRCB was established in 1974 and has been managed by the *Pyrénées Orientales* local council since 1977. It corresponds to a 650-ha surface area including a 65-ha no-take zone (NTZ) added in 1981 (*Cap Rédéris*), where all fishing, mooring and diving activities are prohibited. The rest of the area is managed as an intermediate zone (IZ); spearfishing and trawling are prohibited while recreational and artisanal fishing are allowed but regulated. The main objective of the NMRCB is to protect the fauna and flora and to ensure that human activities are compatible with this protection. NMPGL was created in 2011; it encompasses a 400 000-ha surface area from the Spanish border to Cape Leucate. The park is managed by the French Biodiversity Office and is governed locally by a management board composed of 60 members representative of the territory. The Natural Marine Park ensures the comprehensive and non-fragmented management of an extensive maritime area. It integrates all public policies on biodiversity. The Park aims to meet three fundamental objectives: improving knowledge of the marine environment and its protection, and contributing to the sustainable development of maritime activities. To date, there are still no restricted areas within NMPGL (except for the NMRCB included in park). Here we present the results of the assessment of ecosystem quality in the two MPAs to highlight the potentiality of the EBA and its implementation in a context of collaboration between managers and scientists.

Material and methods

The training of NMRCB and NMPGL agents was undertaken in three stages. (i) Training for EBQIs was attended by two marine biologists in June 2019. After a presentation of the concept of the EBA and EBQI protocols, 7 dives were organised to train the agents according to their skills and experience. (ii) In October 2020, an online meeting was organised to present the database to the managers. (iii) A second training session took place in July 2021 focused on some specific descriptors and technical issues at the request of the managers (e.g., invertebrate identification for CB and IPR). After the training, agents from the two MPAs were able to sample data and measure an EBQI for three different ecosystems. Between May 2020 and August 2021, 29 sites were sampled: 8 for POSM, 10 for IPR and 11 for CB. The protocols presented in the related publications (Personnic *et al.*, 2014; Ruitton *et al.*, 2014; Thibaut *et al.*, 2017) and in a guide dedicated to managers (Ruitton *et al.*, 2017) were used to measure the three EBQIs and the Confidence Index (CI).

Results

Confidence Index (CI)

The CI for all sampling sites and for all the three ecosystems ranges between 98 and 100%.

***Posidonia oceanica* seagrass meadows**

EBQI values for POSM ranges from 3.90 to 7.47. Six out of the eight sampling sites had an Intermediate ecological state, and only one Poor and one Good. The Principal Component Analysis (PCA) (Fig. 1 and Fig. 4) projects the sites according to the influence of the compartment (= box) status. Most of the boxes are few or non-discriminant, such as sea birds (box 13a, 13b), *Pinna nobilis* density (5), meadow cover (2b), epibionta (3-4), other filter-feeders (6a, 6b). The most discriminating boxes between sites are carnivorous and piscivorous fish (10, 11, 10-11-12), litter (7) and detritus-feeders (8). The highest EBQI value was found in the no-take zone (H06 Pin Parasol).

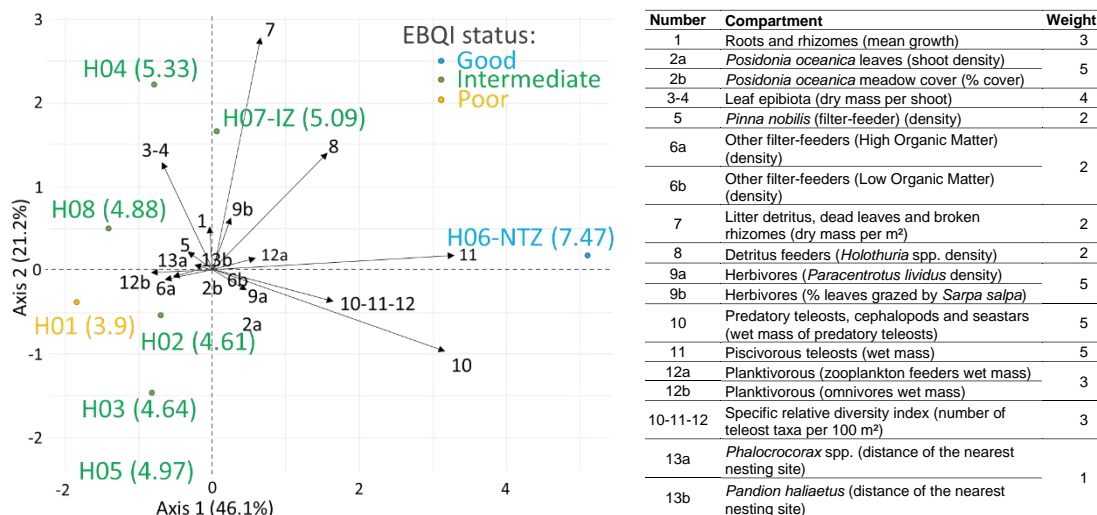


Fig. 1: Principal Component Analysis showing the influence of functional compartments on the ecological status of the sampling sites considering *Posidonia oceanica* seagrass meadows (compartments derived from Personnic *et al.*, 2014); the EBQI value is in brackets.

Infralittoral Photophilous Reef

EBQI values for IPR ranges between 4.00 and 7.88. Six out of the ten sites have an Intermediate status, while 2 are Poor. One site is in a Very Good status (I08 Cap R d ris, NTZ) and one is Good (I06, 3 Moines, IZ). The PCA (Fig. 2 and Fig. 4) shows that the highest values are strongly related with the fish assemblage boxes (6, 7-8, 9, 10). Sea birds (11a, 11b), invertebrate carnivorous *Hexaplex trunculus* (5b), filter-feeders (3) and macrophytes (1) explain few differences among sites.

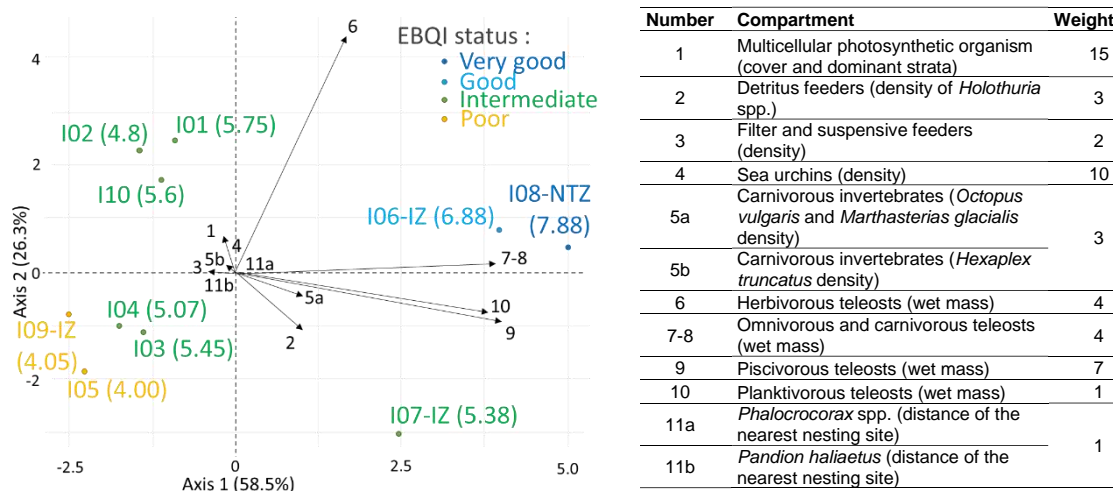


Fig. 2: Principal Component Analysis showing the influence of functional compartments on the ecological status of the sampling sites considering Infralittoral Photophilous Reefs (compartments derived from Thibaut *et al.*, 2017); the EBQI value is in brackets.

Coralligenous Bioconcretions

EBQI values for CB ranges from 3.75 to 6.18. One site has a Good status, while 6 sites an Intermediate status and 4 sites a Poor status. Teleost assemblage boxes (6, 7, 8, 6-7-8) explain the differences among sampling sites, such as Clionaidae bioeroders (4a), non-calcareous macroalgae (2a, 2b), and grazers (5). The highest value is found in the no-take zone (C09, Sec R d ris) (Fig. 3 and Fig. 4).

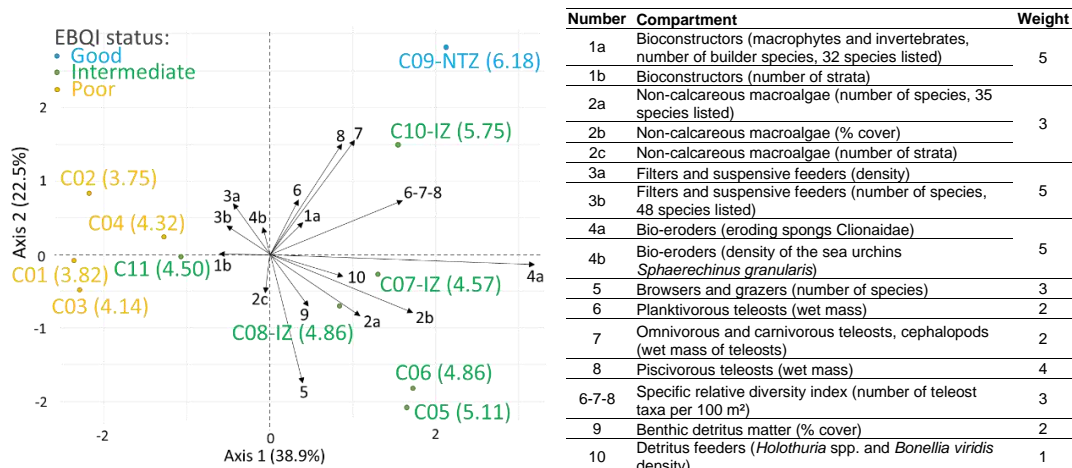


Fig. 3: Principal Component Analysis showing the influence of functional compartments on the ecological status of the sampling sites considering Coralligenous Bioconcretions (compartments derived from Ruitton *et al.*, 2014, 2017); the EBQI value is in brackets.

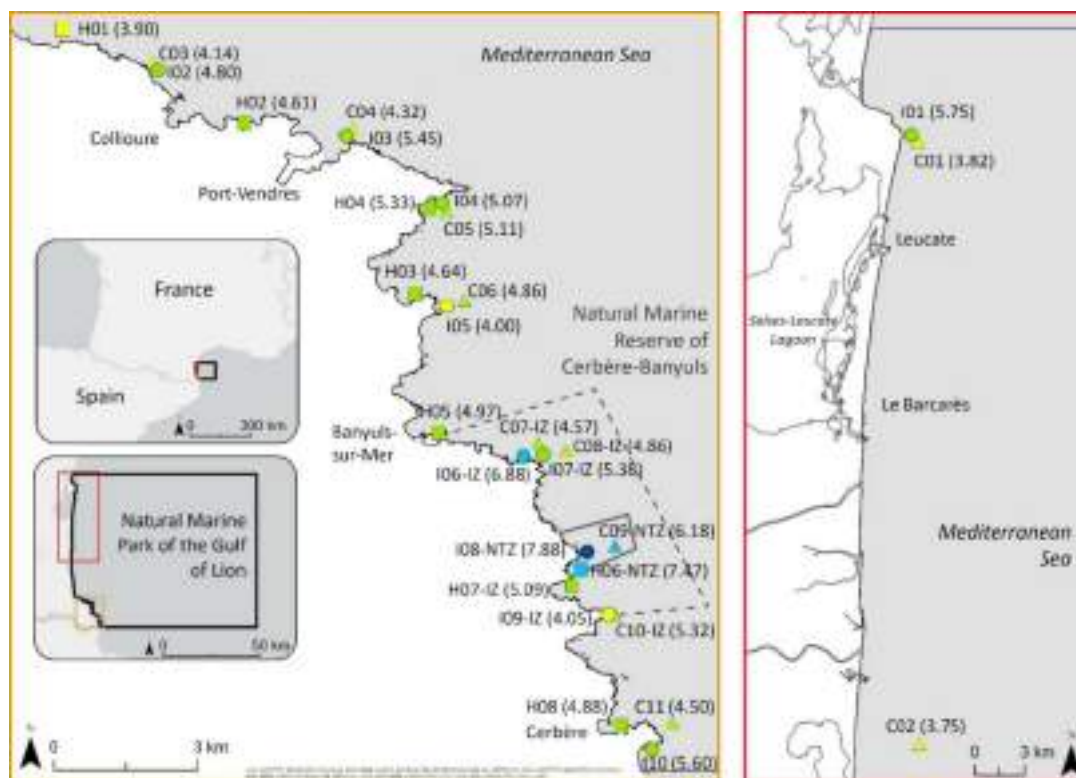


Fig. 4: EBQI values (in brackets) per site for the three ecosystems (square: POSM; circle: IPR; triangle: CB). Colours indicate the ecological status (dark blue: Very Good; blue: Good; green: Intermediate; yellow: Poor).

Discussion

EBQI appears to be a useful tool to better understand the ecosystem functioning at the MPA scale. It helps in the assessment of the ecological status of coastal ecosystems and of the MPA territory by MPA agents. The NMRCB and the NMPGL have been able to set up an effective local monitoring network. This new original dataset provides direct support for the managers in the current NMRCB extension project, arguing in favour of extending the no-take area and further conservation measures. In the frame of the Marha

project and the assessment of the conservation status of marine habitats of interest, these data are also useful at a larger scale (Habitats Directive and MSFD goals).

The application of EBQI in the Gulf of Lion has encountered specific biogeographical issues that must be taken into account. (i) POSM: the depth range of the seagrass meadows (about 3-15 m) does not allow the strict application of the recommended protocol (Personnic *et al.*, 2014). Adjustments have been proposed to make the sampling possible, e.g. the status of shoot density must take into account the current depth and use the classification by Pergent (2007). *Paracentrotus lividus* density was much higher than in the eastern part of the French Mediterranean coasts. This density could be considered either as the consequence of an input of organic matter linked with pollution, or it is probably a natural feature observed in this area. (ii) IPR: rocky reefs along the French Catalonian coast are characterised by a very low occurrence of canopy-forming perennial macrophytes, as a consequence of long-term decline (Thibaut *et al.*, 2005); this leads to a Poor or Intermediate value for box 1, which has the highest weighting, decreasing the overall status. (iii) CB: Bioconstructors show a low specific diversity and bioeroders (*Cliona* spp.) are abundant. Compared to the eastern sites (e.g., Provence, Corsica), macrophytes are less abundant in assemblages dominated by filter-feeders (e.g., Cnidaria, Porifera, and Bryozoans).

Possible changes in the descriptors or the weighting of the functional compartments for the 3 monitored ecosystems might be mandatory and should be discussed jointly between managers and the experts involved in the application of the EBQIs. The goal is to consider the biogeographical differences between areas; this step is challenging, we have to keep in mind that EBQIs assess the functioning of an ecosystem, which varies naturally between sites, and is not only driven by anthropogenic factors. Robustness analysis is needed to validate the efficiency of the indices with or without method adjustment. However, our results show a strong influence of the management level. The highest values of ecological quality are obtained in the *Rédéris* NTZ for all ecosystems. Compartments linked with fish assemblages are the most discriminant among the sampling sites. Fishing is known to be one of the most strongly impacting activities, affecting both the integrity of the seabeds (Quemmerais-Amice *et al.*, 2020) and fish stocks. The low status of teleost assemblages measured in our study, except within the NMRCB (NTZ and IR), is in line with this observation. We suggest applying EBQI to other Mediterranean areas with other biogeographical contexts influenced by anthropogenic activities, water warming and the spread of Non-Indigenous Species inducing phase shift with major consequences on the ecosystem functioning (Occhipinti-Ambrogi, 2007; Montefalcone *et al.*, 2015). In such areas, an EBA could be of help in the face of current and upcoming management issues.

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Thomas BOCKEL, BOISSERY P., DELARUELLE G., DETER J., GERVAISE C., HOLON F.

Andromède océanologie, 34130 Mauguio (France)

E-mail: gwenaelle.delaruelle@andromede-ocean.com

A NEW MAP ATLAS OF COASTAL MARINE BIODIVERSITY

Abstract

Since 2010, biological monitoring data on the marine environment have been acquired to contribute to the Water Framework Directive (WFD 2000/60/EC) and the Marine Strategy Framework Directive (MSFD 2008/56/EC).

*Published in 2020, the map atlas of the French Mediterranean coastal water quality proposes a new integrated mean to estimate the status of *Posidonia oceanica* meadows and coralligenous reefs and by extension the final status of water bodies hosting them (according to five categories). The final status is based on a triplet: 1) ecological status - indicators based on bibliography and expert proposals in the light of current knowledge; 2) functioning status- assessed by fish diversity through metabarcoding environmental DNA, by the biophony of fish, crustaceans and other benthic invertebrates and by the vitality of erect species; 3) the level of threats - based on 11 anthropogenic pressures. In total, 430 recent biological data (less than six years old) obtained from six monitoring programs are combined. All these data are interpreted and synthesised in 83 maps accessible on the MEDTRIX platform (Atlas project).*

*Based on current knowledge and available information, a good final ecological status is achieved in 74% of water bodies considering coralligenous reefs and 61% considering *Posidonia* meadows. Functioning is altered in most of the water bodies and downgrades the final ecological status (80% of the coralligenous reefs and 45% of the seagrass beds).*

This atlas makes it possible to better qualify the conservation status of seagrass beds and coralligenous areas by integrating information on ecological functioning. It helps to refine the identification of priority sectors, to highlight the results of the monitoring programs and to inform the public. This data can help coastal communities, government services and sea users to better understand and therefore better protect underwater biodiversity.

Key-words: atlas of marine biodiversity, *Posidonia oceanica*, coralligenous reefs, ecological status, functional status

Introduction

Knowledge efforts regarding the Mediterranean marine environment have been improved over the past 30 years strengthened in 2006 by the Water Framework Directive (WFD - 2000/60/EC) in the fields of chemistry, biology and pressures. Monitoring was even further expanded in 2016 in chemistry and biology (particularly ecosystem functioning) in order to contribute more actively to the Marine Strategy Framework Directive ((MSFD 2008/56/EC)). Thus, since 2010, several programs have been set up to collect a large amount of data and evaluate the health status of two key ecosystems in the French Mediterranean: *Posidonia* meadows (*Posidonia oceanica*) and coralligenous reefs (MEDTRIX, 2022).

Biological data obtained from TEMPO (*Posidonia* meadow) and RECOR (coralligenous reefs) monitoring networks form the basis for knowledge and understanding of these two ecosystems. These specific data collected every three years in each French region on 118 TEMPO's sites and 112 RECOR's sites located all along the French Mediterranean

coastline make it possible, *in fine*, to assess the general status of these habitats (according to the definition of the European directives concerned). This assessment takes into account the difference between the measured status and a reference status (within the framework of biological monitoring, a reference status is not assessed on a TEMPO or RECOR site in particular; each parameter measured is compared to a reference value fixed per region, i.e. a maximum value observed since the start of measurements, all sites combined, and which may therefore change during monitoring) and, if possible, a baseline ("pristine").

This data is supplemented by other monitoring networks operated simultaneously during the same field campaign: PISCIS (fish populations), CALOR (water temperature) and CALME (acoustics). Area-based data acquired over longer periods of time complement these biological data: the IMPACT (modelling of coastal anthropic pressures) and SURFSTAT (area-based analysis of marine habitats) networks.

These networks are operated during multi-network campaigns in order to optimise the costs of the monitoring programs but also to ensure better interpretation, particularly for the link between ecological status and ecological functioning. They are also constantly being optimised in terms of equipment and techniques (diving techniques, underwater data acquisition techniques, database and mapping tools).

In order to better understand all the information acquired during these monitoring programs (RECOR, TEMPO, PISCIS, CALME, SURFSTAT and IMPACT), the most recent data has been grouped together and synthesised in a map atlas (ANDROMEDE OCEANOLOGIE, 2020). Published in 2020, the map atlas of the French Mediterranean coastal water quality proposes a new integrated means to estimate the status of *Posidonia oceanica* meadows and coralligenous reefs and by extension the final status of water bodies hosting them.

Materials and methods

For the *Posidonia* meadows, the assessment of ecological status combines three indicators for *Posidonia* as a plant: 1) Vitality index based on the BiPo (Biotic Index using *Posidonia oceanica*) index (Lopez y Royo *et al.*, 2010); 2) Index of evolutionary dynamics at the lower limit, which takes into account the evolution of the shoots density (in %) and the microsurface dynamics of the meadow (% regression and/or progression) between each monitoring on the same site; 3) Surface indicator calculated by combining two landscape indices: decline and cohesion (Houngnandan *et al.*, 2020). The assessment of the ecological functioning of the meadow also combines three indicators: 1) the diversity index of ichthyological populations through metabarcoding environmental DNA; 2) the biophonic quality index of fish (Di Iorio *et al.*, 2018); 3) the biophonic quality index of invertebrates (Lossent *et al.*, 2017). The assessment of pressures considers the index of cumulative impacts on the seagrass based on 11 anthropogenic pressures (Holon, 2015; Holon *et al.*, 2015).

For coralligenous reefs, the assessment of ecological status combines three indicators based on the analysis of photographic quadrats acquired in the framework of the RECOR network (Deter *et al.*, 2012a, 2012b, Doxa *et al.*, 2016): 1) Sessile species vitality index based on the Coralligenous Assemblages Index (Deter *et al.*, 2012a, 2012b); 2) Filamentous algae bloom indicator, 3) Coralligenous dynamics indicator which corresponds to the percentage of necrosis of bioconstructive algae. The evaluation of the ecological functioning of the coralligenous also combines three indicators: 1) the diversity index of the ichthyological populations through metabarcoding environmental

DNA; 2) the biophonic quality index of the invertebrates; 3) the vitality index of erect species (gorgonians). The assessment of the level of pressures considers the index of cumulative impacts on coralligenous reefs based on 11 anthropogenic pressures.

For each water body considered under the WFD, some indicators are calculated on the entire scale of the water body, while the others are measured at specific stations, known as reference stations, during TEMPO or RECOR monitoring. The indicators are combined according to different weights depending on their level of representativeness of the entire water body. When several sites are monitored in the same water body, the most downgraded (i.e., the least favourable) value of the index is taken into account. The final status of water bodies is then assessed for each habitat according to five categories.

Results

In total, 430 recent biological data (less than six years old) obtained from six monitoring programs are combined. All these data are interpreted and synthesized in 83 maps accessible on the MEDTRIX platform (Atlas project), 39 for coralligenous reefs (Fig. 1) and 44 for *Posidonia* meadows (Fig. 2).

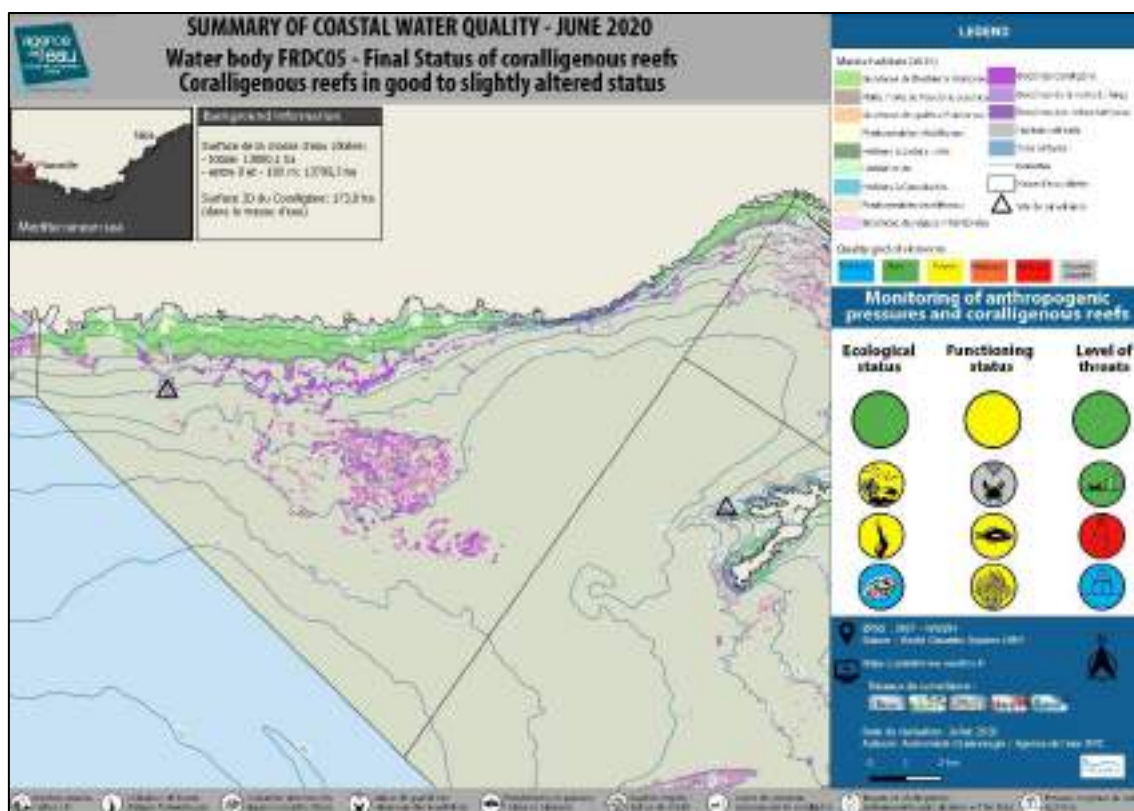


Fig. 1. Example of a map showing the final status of a waterbody for coralligenous reefs. The coloured dots in the inset on the right of the map indicate the quality of each indicator, allowing the assessment of the ecological status, the state of ecological functioning and the level of pressures for each ecosystem. The final status of each ecosystem is indicated in the title of the map. The location of the reference stations is indicated by a black triangle.

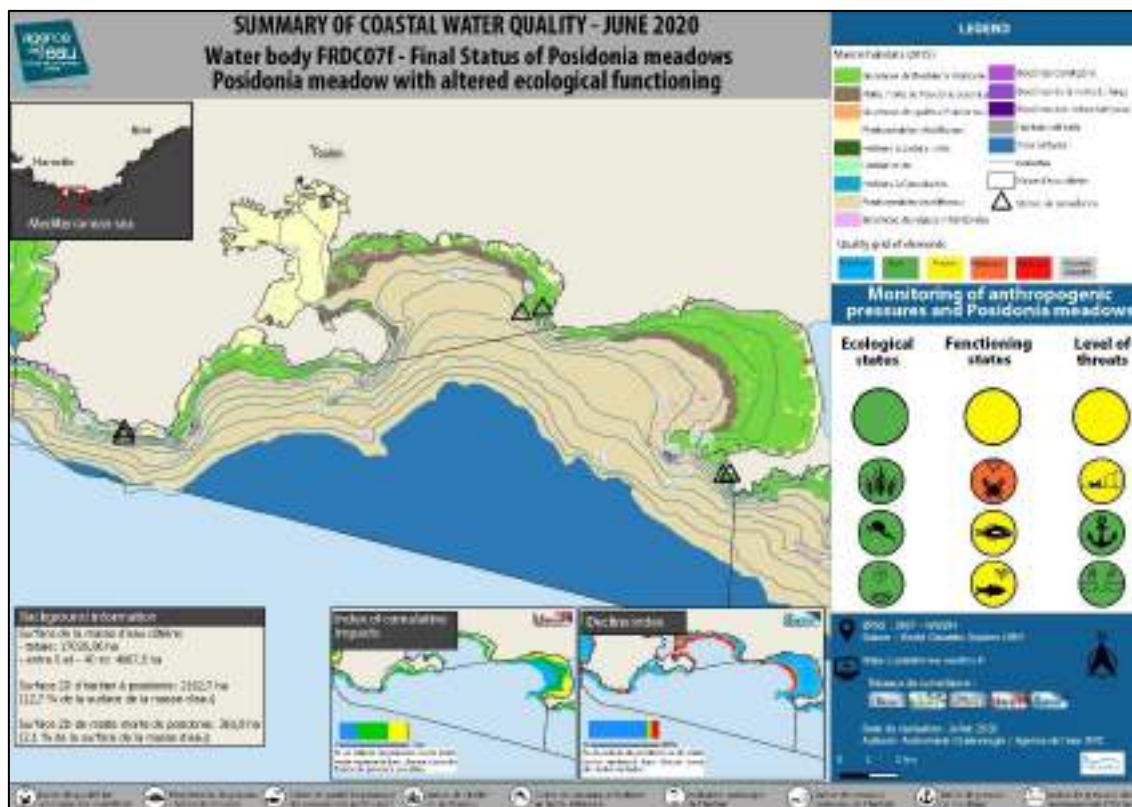


Fig. 2. Example of a map showing the final status of a waterbody for Posidonia meadows.

A good final ecological status is achieved in 74% of waterbodies considering coralligenous reefs and 61% considering Posidonia meadows. Functioning is altered in most of the water bodies and downgrades the final ecological status (80% of the coralligenous reefs and 45% of the seagrass beds).

Functioning is altered in most waterbodies (71% of water bodies for seagrass beds and 84% for coralligenous reefs) and degrades the final ecological status (80% of coralligenous reefs and 45% of seagrass beds). This significant alteration in the level of functioning of these habitats is not totally correlated with the levels of cumulative pressure, and future work will make it possible to better characterise this situation by adding "noise" and "professional and pleasure fishing" pressures to the IMPACT monitoring network.

Discussion and/or conclusions

This atlas makes it possible to better qualify the conservation status of seagrass beds and coralligenous areas by integrating information on ecological functioning. It helps to refine the identification of priority sectors, to highlight the results of the monitoring programs and to inform the public. This data can help coastal communities, government services and sea users to better understand and therefore better protect underwater biodiversity.

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Thomas BOCKEL, HOLON F., MARRE G, DELARUELLE G., BOISSERY P., DETER J.

Andromède océanologie, 34130 Mauguio (France)/ Université de Montpellier-UMR MARBEC

E-mail: thomas.bockel@andromede-ocean.com

AIS DATA HELP TO MANAGE ANCHORING AND ITS IMPACT ON FRENCH SEAGRASS MEADOWS

Abstract

*The Mediterranean Sea is a hotspot for worldwide tourism. Its waters also host a remarkable biodiversity including the endemic marine seagrass *Posidonia oceanica*, which procures many valuable ecological services to the area.*

The impact of boating activity through anchoring on the seagrass meadows has been documented and is considered particularly worrying for larger boats.

The anchoring behavior of large yachts equipped with Automatic Identification System (AIS) can be mapped precisely based on the analysis of the spatio-temporal distribution of boats positions.

*Based on more than 75000 anchoring positions derived from AIS data between 2010 and 2018 in the French Mediterranean region, we have shown a strong increase in the number of anchoring events (x4), that 25% of the anchors were dropped on *Posidonia* meadows, and a loss of more than 100 ha of seagrass in some sectors in this period.*

*This knowledge participated in raising awareness on this under evaluated threat, leading in 2019 to new regulations in the region with the interdiction of anchoring in seagrass *Posidonia oceanica* for large boats (more than 24 meters) (“arrêté n° 123/2019”).*

*Since the enforcement of the new regulation, AIS data analysis allowed us to show a decrease of approximately 20% of the number of large ships anchoring in *Posidonia* meadows between 2019 and 2021.*

Smaller boats, which represent a majority, also have an impact when anchoring in seagrass. They most of time are not equipped with AIS. The challenge is now to localize their positions and quantify their impact.

Key-words: *Impact, Anchoring, Seagrass, AIS, Monitoring*

Introduction

Marine traffic is a priority human activity in our globalized society, but its impact on the environment is high (Walker *et al.*, 2019), in particular on sensitive marine habitats. Quantifying human pressures and their impacts is a major challenge in the marine environment (Halpern *et al.*, 2008), even greater when it comes to regulation enforcement and evaluation. The Automatic Identification System (AIS, communicating information of position and identification between ships to avoid collisions) while initially developed as a security tool, reveals an interesting asset in addressing this challenge, and was previously used to map anchoring pressures and impact on French marine habitats including the protected *Posidonia* seagrass beds (Deter *et al.*, 2017). *Posidonia oceanica* forms large meadows between the surface and approximately 40 meters deep. *P. oceanica* is very sensitive to anchoring pressure (Abadie, 2016) and shows a very slow recolonization speed once damaged (Boudouresque *et al.*, 2006). Major regressions of *P. oceanica* were observed in the Mediterranean Sea in the last century (Marbà *et al.*, 2014).

A European and national legal protection framework exists for *P. oceanica* (DCE, DCSMM, Natura 2000) for many years. New strict regulations have been adopted in the past two years in the French Mediterranean (“arrêté n°155/2016”, “arreté 123/2019”), forbidding any anchoring in *P. oceanica* meadows for boats larger than 24 m.

Taking the study case of France, a hotspot for boating tourism in the world (UNEP/MAP, 2017), the objective of this paper is to show how AIS data can be used 1. to quantify the spatial distribution and the temporal evolution of anchoring pressure, and 2. to evaluate the effectiveness of the regulation.

Materials and methods

The study area is the Mediterranean French coast including Corsica (1800 km coastline) between 0 and –80 m.

- AIS data 2010-2018 (Marine traffic) and 2019-2021 (AISHUB):

AIS data was collected from two different sources. AIS data from 2010 to 2018 came from Marine traffic database (www.marinetraffic.com). Those AIS positions correspond to positions of declared anchoring activity, received by terrestrial AIS stations, with an hourly frequency. AIS data from 2019 to 2021 came from the AIShub network of receiving stations; those AIS data are raw positions that we collected with a frequency of one point every two minutes. AIS data contain information on boat identification, time of detection, position, direction, speed, dimensions, type, and destination.

The methodology used to obtain the anchoring positions from AIS positions (whatever the period) is derived from Deter et al. (2017). Originally, the authors filtered the AIS data with the following conditions: speed <1 knot, distance between the hourly AIS points ≤ 600 m, AIS points per vessel ≥ 4 , no anchoring within harbors or equipped mooring areas. They then fitted a regression circle to the AIS positions and its center was defined as the position of the anchor. In case of a calculated radius higher than 600 m, the centroid of AIS positions was considered as the position of the anchor. They finally defined the anchoring zone as a polygon including the anchoring position of a vessel and its AIS points and reduced it by one third in a concentric manner to obtain the anchoring impact area. We applied the same methodology in this study using R version 4.1.2. The computation challenges posed by the larger number of AIS positions, due to the use of raw AIS data, were addressed using PostgreSQL 9.6 and PostGIS 2.4 spatial database management tools.

All the anchoring positions are freely available for visualization in the cartographic platform Medtrix (www.medtrix.fr) on the “*suivi du mouillage*” project.

A major weakness of this method was highlighted in the literature (Deter *et al.*, 2017), showing that only 1/20 of the boats were detected with AIS compared to boats counted from land, mainly concerning small boats and medium boats (<50 meters). This weakness is partly addressed in this study by using raw AIS data instead of declared anchoring status only.

- Seabed habitat maps:

We used a 1:10,000 seabed habitat map (between 0 and –80 m) covering the entire study area. This continuous map was built in 2014 using a combination of aerial pictures, multi-beam echosounder data, side-scan sonar data and direct observations (“ground-truth points”) by divers (Andromède Océanologie 2014). This map is composed of eleven habitat classes: *Cymodocea nodosa* seagrass, *Posidonia oceanica* seagrass, dead matte association, infralittoral shingle association, infralittoral soft bottoms, photophilous algae association, coralligenous reefs (= biogenic reefs), circalittoral soft bottoms, artificial

habitats, offshore rocks and the bathyal zone. Dead matte association is the habitat resulting from the death of *P. oceanica* seagrass beds (Boudouresque et al., 2006). The seabed habitat map is freely available for visualization in the cartographic platform Medtrix (www.medtrix.fr) on the “DONIA expert” project.

- Mooring regulation:

The two recent prefectural decrees (n°155/2016 and in particular n°123/2019) were declined locally by 14 decrees between 2019 and 2021 with the definition of forbidden mooring areas.

A regulation database was created regrouping all polygons of anchoring interdiction. This regulation database is available freely for visualization on the cartographic platform Medtrix (www.medtrix.fr) on the “Donia” project.

- Data analysis:

The first analysis looked at the evolution of the number of anchoring events per year and size of the boat. This part of the analysis was performed separately for 2010-2018 period and the 2019-2021 period because the data source changed in 2019 making the absolute numbers not comparable. The positions before 2018 include only declared anchored positions while all boats positions are considered after 2018. A comparative analysis was performed for 2018 (both data were collected for this year) on the number of anchoring positions.

The ratio between the number of anchoring points, determined from declared anchored positions, and all boat positions is calculated for the year 2018 for which both datasets are available. This ratio equals 2.22 on average, ranging from close to 1 for boats larger than 40 m to almost 50 for boats smaller than 24 m (the low average is explained by a very small number of declared anchored positions for boats smaller than 24°m).

The proportion of anchoring events per habitat was then analyzed to investigate the effect of the regulation. This proportion is first plotted for the boats longer than 80 meters between 2014 and 2021, then for the boats longer than 24 meters between 2016 and 2021. This part of the analysis was focusing on proportions, we hence were able to analyze together the 2010-2018 and the 2019-2021 period. The habitat data were here grouped in four classes: *P. oceanica* seagrass, *P. oceanica* dead matte, soft bottom and other. For each class of boat size (+80 m and +24 m (+24°m includes +80°m)), the relationship between the variables habitat and year was tested with a Chi2 test in order to investigate the change of anchoring behavior with time before and after the regulation change (2016 for boats larger than 80 m and 2019 for boats larger than 24 m).

Results

Between 2010 and 2021, the dataset used corresponds to more than 170000 anchoring positions (more than 50000 between 2010 and 2018 and more than 130000 between 2019 and 2021).

The number of anchoring positions constantly increased between 2010 and 2018. This trend appears visually consistent in every category of boat size. The number of anchoring positions with an unknown boat size is lower after 2015 than before.

The number of anchoring positions per marine habitat highlights contrasting results for the different categories of size of boats.

For boats longer than 80 meters, the number of anchoring positions on *P. oceanica beds* decreased from 2015 (99 positions, 19 %) to 2021 (34 positions, 5 %) (Fig. 1). The relationship between the variables habitat and year is significant ($\text{Chi}^2=145, p<10^{-15}$).

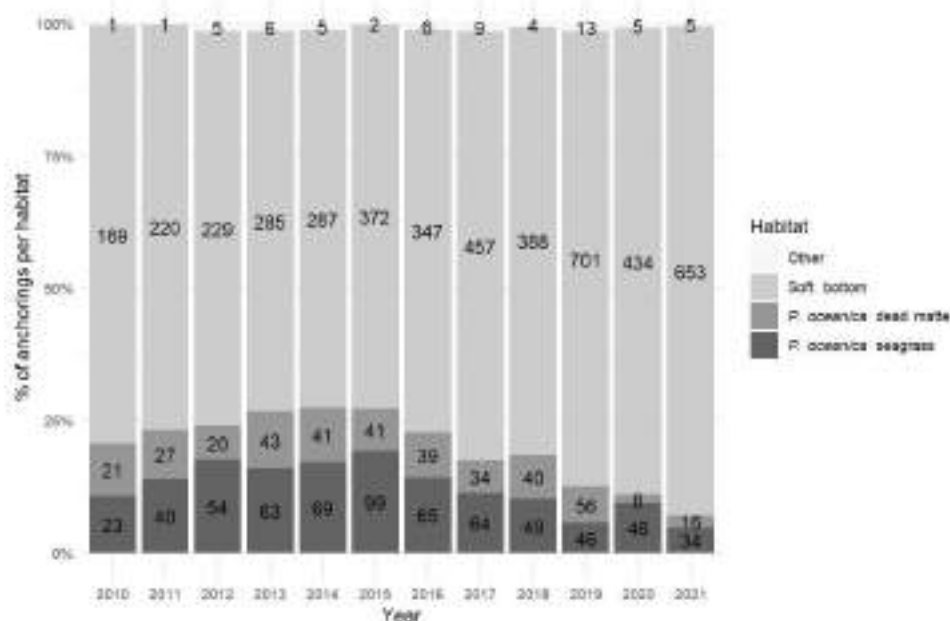


Fig. 1: Percentage of anchoring positions per year and habitat during the summer between 2010 and 2021 for boats with size higher than 80 m (number of anchoring positions inside bars).

For boats longer than 24 meters, the number of anchoring positions on the seagrass *P. oceanica* appears to be decreasing between 2020 (1665 positions, 35 %) and 2021 (443 positions, 9 %), when looking at the water bodies concerned by the local declinations of the prefectural decree n°123/2019 (Fig. 2). The relationship between the variables habitat and year is significant ($\text{Chi}^2=1043$, $p<10^{-15}$).

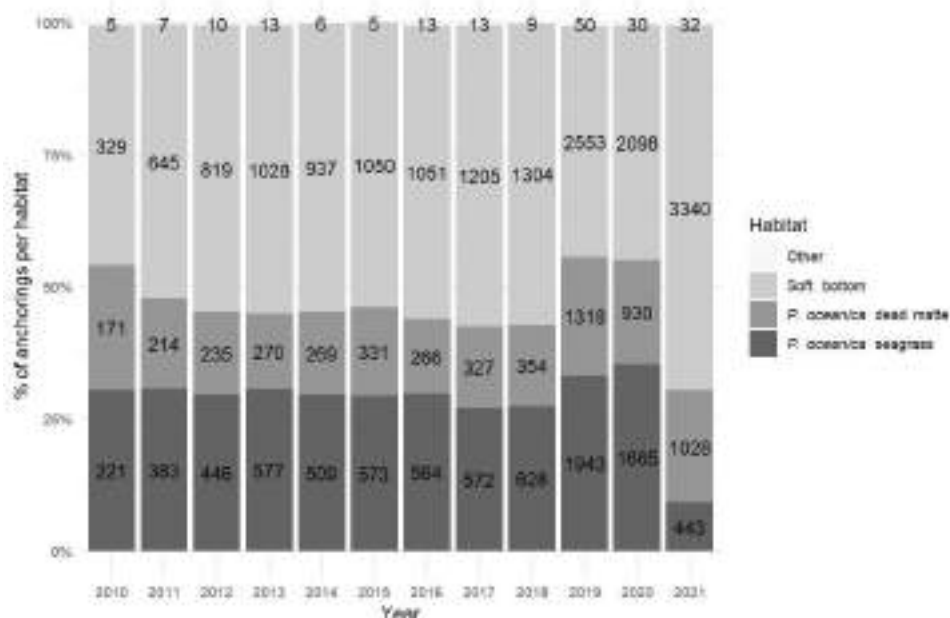


Fig. 2 Percentage of anchoring positions per year and habitat during the summer between 2010 and 2021 for pleasure boats with size higher than 24 m, on water bodies concerned by the local declinations of the prefectural decree n°123/2019 (number of anchoring positions inside bars).

Discussion and conclusions

This work shows that AIS data can be used efficiently to monitor anchoring pressure distribution and its evolution in time.

It highlights the efficiency of the new regulations regarding anchoring on *P. oceanica* in the French Mediterranean Sea. The evolution of anchoring events detected from AIS data well reflects the restrictions enforced by decree n°155/2016 and n°123/2019 regarding boats larger than 80 meters and boats larger than 24 meters respectively.

While a slight decrease is also observed for boats smaller than 24 meters, maybe due to an increasing communication on *P. oceanica* ecological importance and sensitivity to anchoring in the recent years, the anchoring behavior of small boats (smaller than 24 meters) remains almost unchanged with more than 1/3 of the anchoring events on *P. oceanica* in 2021.

AIS data therefore plays a very important role in the management of boating pressures and impacts, in particular those due to anchoring, in the French Mediterranean Sea. It effectively allows quantifying and mapping anchoring pressure but also evaluating the effectiveness of management measures.

Smaller boats are, however, mostly not equipped with AIS and knowledge is still needed on small boats anchoring pressures and impacts.

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**Charles-François BOUDOURESQUE, MARINIER M., CAVALIER M.,
DRIANCOURT T., BLANFUNÉ A., PERRET-BOUDOURESQUE M., THIBAUT T.**
Aix Marseille University, Mediterranean Institute of Oceanography (MIO), CNRS, IRD,
Campus of Luminy, 12009 Marseille, France
E-mail: charles.boudouresque@mio.osupytheas.fr

THE PERCEPTION OF *POSIDONIA OCEANICA* BANQUETTES BY BEACHGOERS IN THE FRENCH RIVIERA

Abstract

*In much of the Mediterranean, banquettes of dead leaves of the seagrass *Posidonia oceanica* washed up on beaches are removed by local authorities, who claim to do so to satisfy the wishes of beachgoers. This practice has serious consequences: the erosion of beaches, sand-replenishment and the further destruction of seagrass beds constitute a kind of vicious circle. But are beachgoers really demanding beaches without banquettes? A survey of the perception of beachgoers in the French Riviera shows that the people's dislike of the banquettes is much less strong than might be expected. A majority of beachgoers, whether frequent or occasional beach users, are not strongly opposed to beaches with banquettes (100% and 94% respectively), do not consider banquettes to be a nuisance (68% and 52%) and are aware of the importance of banquettes for the protection of beaches against erosion (66%). Once beachgoers are duly informed, this majority becomes overwhelming. By removing the banquettes of *P. oceanica*, the local authorities are therefore acting on the basis of their own perception rather than that of the real users.*

Key-words: Beaches; *Banquettes*; French Riviera; *Posidonia oceanica*.

Introduction

The *Posidonia oceanica* seagrass, endemic to the Mediterranean Sea, forms extensive meadows from sea level down to 30-40 m depth (Boudouresque *et al.*, 2012). The consumption of leaves by herbivorous sea urchins (*Paracentrotus lividus*) and fish (*Sarpa salpa*) is usually low (Pergent *et al.*, 1994). As a result, after shedding, dead leaves constitute a litter within the *P. oceanica* meadow. Thereafter, according to currents and storms, these leaves are exported to all benthic ecosystems, from the infralittoral to the bathyal stages, where they constitute a significant contribution to the local food webs, *via* detritus-feeders (Boudouresque *et al.*, 2016).

Some of these dead leaves are washed up on the beaches. There, they form *banquettes*, the thickness of which can exceed 2 m. These *banquettes* constitute an important compartment of the Dune-Beach-Banquette (DBB) ecosystem, which characterizes the Mediterranean (Boudouresque *et al.*, 2017). They play a major role in the protection of beaches against erosion: where they are removed (see below), the beaches recede (Vacchi *et al.*, 2017).

All around the Mediterranean, local authorities are removing the *banquettes*. They claim to do so to satisfy the wishes of beachgoers (Simeone & De Falco, 2013; Otero *et al.*, 2018). Dead leaves and the accompanying sand are often pushed with backhoe loaders to the back of the beach, or to one side of the beach. In the worst cases, they are subsequently evacuated to a landfill. More rarely, they are pushed towards the sea or transported

offshore, in a barge, to be dumped there. This last treatment is the least damaging (Conseil Scientifique des Îles de Lérins & CREOCEAN, 2011; Otero *et al.*, 2018).

The removal of *P. oceanica banquettes* is an ecological and economic disaster (Chessa *et al.*, 2000; Boudouresque *et al.*, 2017; Otero *et al.*, 2018; Fontaine *et al.* (2020); Boudouresque, 2021). (i) A large part of the fauna and flora of the DBB ecosystem is extirpated. (ii) The flow of dead leaves towards the dune, which contributes to both its construction and its supply in nutrients, is compromised. (iii) Sand is an important component of the *banquette* (up to 294 kg/m³); removing the *banquette* deprives the beach of large quantities of sand. (iv) The beach, which is no longer protected, recedes; the local authorities then carry out costly and ineffective sand-replenishment operations which, in a sort of vicious circle, accelerate the withdrawal of the beach. (v) Finally, the *banquette* is a rapidly changing structure: from one storm to another, and depending on the season, leaves arrive, are washed offshore, are washed back and/or change to another beach. Overall, the fate of most of the leaves of the *banquette* is to return to benthic ecosystems, where they constitute a major source of organic carbon and nutrients (Fig. 1). One cubic meter of *banquette*, once processed *via* food webs, represents ~35 kg fish (Boudouresque, 2021). For these reasons, several authors have developed the concept of 'ecological beach', based on the non-removal of *banquettes* (Serantoni, 2015; Boudouresque *et al.*, 2017; Otero *et al.*, 2018; Borrello *et al.*, 2019; Scarpato *et al.*, 2020).

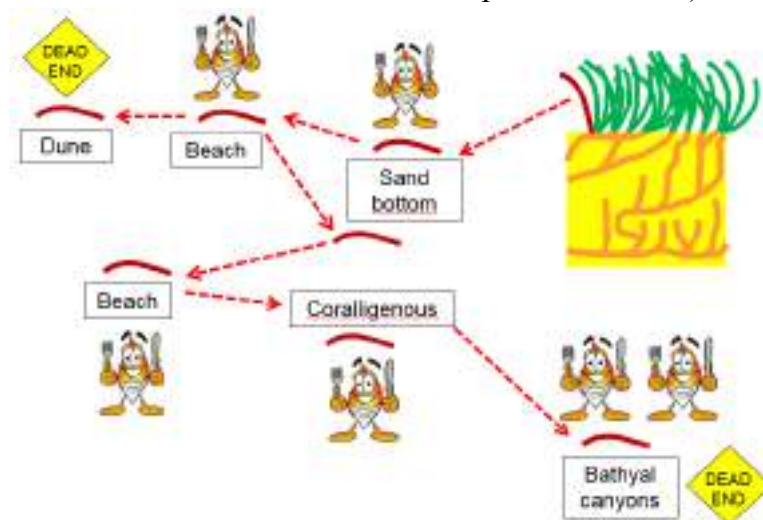


Fig. 1: The possible voyage of a *Posidonia oceanica* dead leaf, from shedding (top right) to total consumption. In the final stage, *via* food webs, symbolized by a crustacean with a knife and fork, dead leaves are converted into fish to be caught by artisanal fishermen.

Local authorities claim that they remove the *banquettes* from the beaches at the request of tourists and beachgoers. In doing so, they trigger a vicious circle, a kind of chain reaction, with serious ecological consequences, which may not matter to them, but which have disastrous economic consequences. But are beachgoers really demanding beaches without *banquettes*? Here, on the basis of a questionnaire for beachgoers from one of the most concerned regions, the French Riviera, we have tried to answer this question.

Material and methods

The questionnaire was drawn up by the authors. We chose to limit it to a small number of questions, formulated in simple and clear language, so that the response time (~10 min) would not be a dissuasive factor in selecting respondents (Tab. 1). The questionnaire was

distributed on social networks (Snapchat, Instagram and Facebook) between October and November 2021 and reached about 550 people, 185 of whom responded, which is an acceptable rate in this type of survey. For questions Q7 and Q12, several answers could be given.

After a first round of the survey not accompanied by scientific explanations (questions Q1 through Q9), a brief scientific text was provided to the respondents. This explained: **(i)** The *banquettes* are part of a unique ecosystem housing many species often endemic to the Mediterranean. **(ii)** The *banquettes* are not a source of significant nuisance for users. **(iii)** *Banquettes* protect beaches against erosion. **(iv)** The dead leaves returning to the sea are a major source of carbon and nutrients for coastal ecosystems. **(v)** Their removal is costly and triggers a vicious cycle of ecological and economic damage. A second round was then organized for Question 8, with three supplementary questions (Q10 through Q12).

Tab. 1: The questionnaire (the original questionnaire was in French).

No of the question	Question
Q1	Have you ever seen the banquettes of <i>Posidonia oceanica</i> ?
Q2	If there is a beach with <i>banquettes</i> , do you settle there or do you look for another beach?
Q3	Do you consider the <i>banquettes</i> of <i>P.oceanica</i> as a nuisance or an inconvenience?
Q4	In your opinion, do the banquettes house a lot of species?
Q5	In your opinion, how important are <i>banquettes</i> in protecting beaches against erosion?
Q6	Are the <i>banquettes</i> an unnatural phenomenon that negatively impacts biodiversity?
Q7	What types of nuisances can the <i>banquettes</i> represent?
Q8	Is the removal of the <i>banquettes</i> of <i>P. oceanica</i> beneficial for the coast or for the users?
Q9	Assess the economic cost of removing the <i>banquettes</i>
Q10	Assess the importance of <i>P. oceanica</i> with regards to the species diversity of coastal ecosystems
Q11	Would you be ready to change your habits by going to beaches with <i>banquettes</i> ?
Q12	Would you be ready to get involved in the protection of the <i>banquettes</i> ?

Additional questions made it possible to draw up a portrait of the respondents: age, profession, region of origin and frequency of beach visits. This latter information enabled us to divide respondents into two groups of unequal size: frequent users (more than 20 visits/year: n=57) and occasional users (less than 20: n=128). For each question, χ^2 tests were performed to assess the responses of the users to the questionnaire.

Results

About 60% of respondents are 18 to 25 years old and 30 % are 40-60. Most of them live in the French *Région Sud* (French Riviera, southern Alps and Provence), but about one fifth comes from other regions or countries. A wide spectrum of professions are represented among the respondents, but the majority are students (56%), which is due to the mode of distribution of the questionnaire. This bias must be taken into account when interpreting the results. It is important to note that age, profession and origin do not differ significantly between the two groups of users (frequent and occasional) (results not presented).

As expected, occasional users know the *banquettes* less well (Q1) and avoid them more (Q2) than frequent users; the ratio of occasional users who find them a nuisance (Q3 and Q7) is higher, but not significantly. A majority of beachgoers, whether frequent or occasional beach users, are not strongly opposed to beaches with *banquettes* (Q2: 100% and 94% respectively), do not consider *banquettes* to be a nuisance (Q3: 68% and 52%) and are aware of the importance of *banquettes* in the protection of beaches against erosion (Q5: 66% and 65%). Once beachgoers are duly informed, this majority becomes

overwhelming (Q8); in addition, they are ready to change their habits (Q11) and even to invest time in the protection of the *banquette* (Q12) (Tab. 2 and 3).

Tab. 2: Answers to questions (first round). χ^2 tests: comparison between frequent and occasional users.

No of the question	Frequent users (n = 57)		Occasional users (n = 228)		χ^2
Q1	Yes: 95% - No: 5%		Yes: 81% - No: 19%		$\chi^2 = 5.10$; $p > 0.05$ Proportion of users who have already seen a <i>banquette</i> significantly different
Q2	I settle there: 55% I look for another beach if not too distant: 45% I look for another beach, even if very distant: 0%		I settle there: 31% I look for another beach if not too distant: 63% I look for another beach, even if very distant: 6%		$\chi^2 = 11.56$; $p > 0.05$. Proportion of users who decide to settle on a beach with or without <i>banquettes</i> significantly different
Q3	1 (not inconvenient): 17% 2: 41%. 3: 19%. 4: 21% 5 (very inconvenient): 2%	No: 68% Yes: 32%	1 (not inconvenient): 14% 2: 22%. 3: 31%. 4: 28% 5 (very inconvenient): 5%	No: 52% Yes: 48%	$\chi^2 = 3.49$; $p < 0.05$. Proportion of users inconvenienced (yes) not significantly different
Q4	1 (few species): 2% 2: 12%. 3: 33%. 4: 24% 5 (many species): 29%	Few: 30% Many: 70%	1 (few species): 9% 2: 10%. 3: 32%. 4: 31% 5 (many species): 18%	Few: 35% Many: 65%	$\chi^2 = 0.24$; $p < 0.05$. Proportion of users who think that a <i>banquette</i> hosts many species not significantly different
Q5	1 (not important): 7% 2: 14%. 3: 27%. 4: 31% 5 (very important): 21%	No: 34% Yes: 66%	1 (not important): 9% 2: 10%. 3: 32%. 4: 31% 5 (very important): 18%	No: 35% Yes: 65%	$\chi^2 = 0.001$; $p < 0.05$. Proportion of users who think that a <i>banquette</i> is important in protecting beaches against erosion not significantly different
Q6	1. natural (no impact on biodiversity): 81% 2. Unnatural without negative impact: 7% 3. Unnatural with negative impact: 12%		1. natural (no impact on biodiversity): 76% 2. Unnatural without negative impact: 8% 3. Unnatural with negative impact: 16%		$\chi^2 = 0.60$; $p < 0.05$. Proportion of users who think that a <i>banquette</i> is an unnatural phenomenon that impact biodiversity not significantly different
Q7	No nuisance: 26% Bad smell: 50% Human toxicity: 10% Negative impact on flora and fauna: 14%		No nuisance: 20% Bad smell: 47% Human toxicity: 11% Negative impact on flora and fauna: 22%		$\chi^2 = 2.05$; $p < 0.05$. Proportion of users who think that a <i>banquette</i> is a nuisance not significantly different
Q8	1 (no beneficial): 31% 2: 17%. 3: 28%. 4: 14% 5 (very beneficial): 10%	No: 62% Yes: 38%	1 (no beneficial): 20% 2: 20%. 3: 35%. 4: 16% 5 (very beneficial): 9%	No: 57% Yes: 43%	$\chi^2 = 0.22$; $p < 0.05$. Proportion of users who think that the removal of a <i>banquette</i> is a benefit not significantly different
Q9	1 (low cost): 0% 2: 7%. 3: 33%. 4: 34% 5 (high cost): 26%	Low: 23% High: 77%	1 (low cost): 3% 2: 8%. 3: 25%. 4: 29% 5 (high cost): 35%	Low: 24% High: 76%	$\chi^2 = 0.01$; $p < 0.05$. Proportion of users who think that removing a <i>banquette</i> is cheap not significantly different

Tab. 3: Answers to questions (second round). χ^2 tests: comparison between frequent and occasional users.

No of the question	Frequent users (n = 57)		Occasional users (n = 128)		χ^2
Q8	Not beneficial: 88%. Beneficial: 12%		Not beneficial: 92%. Beneficial: 8%		$\chi^2 = 0.35$; $p < 0.05$. Proportion of users who think that the removal of a <i>banquette</i> is a benefit not significantly different
Q10	1 (not important): 0% 2: 2%. 3: 2%. 4: 17% 5 (very important): 79%	Low: 3% High: 97%	1 (not important): 0% 2: 1%. 3: 2%. 4: 19% 5 (very important): 78%	Low: 2% High: 98%	$\chi^2 = 0.01$; $p < 0.05$. Proportion of users who think that a <i>banquette</i> is important in hosting high species diversity not significantly different
Q11	Never: 2% From time to time: 5% Most of the time: 62% All the time: 31%		Never: 5% From time to time: 28% Most of the time: 39% All the time: 28%		$\chi^2 = 15.3$; $p > 0.05$. Proportion of users who are ready to change their habits significantly different
Q12	I do not want to get involved: 6% Yes, by raising awareness around me: 42% Yes, by seeking to know more: 22% Yes, by choosing beaches with <i>banquettes</i> : 30%		I do not want to get involved: 7% Yes, by raising awareness around me: 43% Yes, by seeking to know more: 21% Yes, by choosing beaches with <i>banquettes</i> : 29%		$\chi^2 = 0.144$; $p < 0.05$. Proportion of users who are ready to invest time in protecting <i>banquettes</i> not significantly different

Discussion and conclusions

Local authorities who, legally or not, destroy the *banquettes* of *P. oceanica*, sometimes to send them to landfill, have always claimed to do so at the request of the public. This study shows that the people's dislike of *banquettes* is neither as clear nor as massive as local authorities claim. This study is not the first to draw attention to this paradox: is the personal dislike of a handful of managers prevailing over the ecological and economic evidence and the opinion of the users?

So far, studies on the perception of the *banquettes* are unfortunately only available in the gray literature. A study based on users (n = 201) of 17 beaches, from Provence to the French Riviera (France), shows that negative or somewhat negative opinions are given by 44% of respondents, but that 56% have a neutral to positive opinion (Berghthold, 2017). Another study, carried out in five EU countries (n = 1 200), illustrates the divergence between the perception of users and that of tour operators and especially local authorities (Otero *et al.*, 2018; Mossone *et al.*, 2019) (Tab. 4). All in all, would the ecological and economic disaster of the removal of *banquettes* on the beaches be mainly decided on by few technocrats and local government officials, ill-informed and reading neither gray literature nor scientific publications (as already suggested by Boudouresque *et al.*, 2017; Boudouresque, 2021)? Furthermore, our survey shows that informing beachgoers regarding the importance of *banquettes* could be an effective solution for managers.

Tab. 4: Divergent perceptions of the *banquettes* of *P. oceanica*, between beachgoers, tour operators and local authorities, in five EU countries (Cyprus, France, Greece, Italy and Spain). Based upon data from Otero *et al.* (2018) and Mossone *et al.* (2019).

		Beachgoers	Tour operators	Local authorities
Perception of the presence of <i>banquette</i> ?	Negative	41%	75%	74%
	Neutral	33%	18%	22%
	Positive	26%	7%	5%
Does the presence of the <i>banquette</i> affect tourism?	Yes, negatively	43%	75%	80%
	No	48%	12%	6%
	Yes, positively	9%	13%	20%

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Julie DETER, HOUNGNANDAN F., BENKHABCHECHE F., HARDY P.-Y., GUILBERT A., TRIBOT A.-S., POURTAU M., SUTTON G., BARROIL A., HOLON F., BOISSERY P.

Andromède océanologie, 34130 Mauguio (France)/ Université de Montpellier-UMR MARBEC

E-mail: julie.deter@andromede-ocean.com

PARTICIPATIVE MAPPING AND AERIAL PHOTOGRAPHS HELP TO SHOW THE STRONG DECLINE OF NORTH EASTERN *POSIDONIA OCEANICA* SEAGRASS BEDS (FRENCH OCCITANIE REGION)

Abstract

Since the 1960's the French region Occitanie has undergone profound development in order to create a tourism economy. This development have strongly impacted the coastline and consequently the marine ecosystems. However, no monitoring work had been done to measure the impact of human activities on the seagrass beds in this region presently known for its almost total absence of Posidonia meadow. Using bibliographic data, aerial photographs and a survey of local people, we were able to map the historical Posidonia oceanica beds in Occitanie.

Our results confirm that some areas never harboured Posidonia meadows because the local conditions are not favorable (i.e. lagoon outlets, fine sandy bottoms, hydrodynamics), such as the sandy coast from Argelès-sur-mer to Vias. On the other hand, observations have been made on the disappearance of Posidonia meadows in certain areas, such as off Frontignan, where a whole area of meadows has disappeared to make way for dead matte, visible or buried. Meadows of Posidonia oceanica experienced an overall regression of 65% (409.8 ha) in the last 70 years. The main causes cited during the surveys are coastal development, pollution and anchoring. At the same time, all the respondents noted a decrease in fish populations over the last 70 years, the causes of which, according to them, were: overfishing (35%), water quality reduction (31%), and the decline of the P. oceanica meadow (16%).

This work shows the importance of local knowledge and the interest of mixing different methods to establish a baseline in a region. This works brings also a baseline for Posidonia meadows in their French northwestern limit of distribution.

Key-words: Participative mapping, *Posidonia oceanica*, Spatial analysis, Local ecological knowledge (LEK)

Introduction

Historical data and long time series are important for assessing the magnitude and changes in the long-term evolution of ecosystems (Ellingsen et al., 2017) and for planning and adapting the ecological management strategy (Gatti et al., 2015). The use of historical information is also recommended to establish baselines used for ecological restoration and/or define the good environmental status of marine ecosystems under the European Community Marine Strategy Framework Directive (Borja et al., 2013). In ecology, scientists have understood the importance of indigenous knowledge in assessing the state of biodiversity degradation (Skroblin et al., 2020). This knowledge is also now being used to set up marine protected areas (Strickland-Munro et al., 2016), manage fisheries (Hall-Arber et al., 2009) and for marine spatial planning (Trouillet, 2019).

Assessments of long-term changes were recently used to show that one third of European seagrass area has been lost during the last 150 years, due to disease, deteriorated water quality, and coastal development (de los Santos et al., 2019). Losses peaked in the 1970s and 1980s but some reversal seems to be possible since 2000's even for the slow growing seagrass *Posidonia oceanica* (de los Santos et al., 2019). *Posidonia oceanica* (L.) Delile is the most common seagrass species in the Mediterranean. This protected and endemic species forms extensive marine grasslands, an habitat of community interest (92/43 / EEC Habitats Directive, habitat codes 1120: Posidonia). It establishes on the major part of the Mediterranean coastline, between 0 and -40 m deep and occupies 1-2% of the seabed (Pasqualini et al., 1998), i.e. a total area of 1,224,707 ha (Telesca et al., 2015). Here we use a multidisciplinary approach combining local ecological knowledge (LEK), and the analysis of old aerial pictures in order to investigate the baseline of *P. oceanica* seagrass beds in a region that has known a high development during the last half century. The comparison with the current cover permits to estimate lost areas.

Materials and methods

The study is interested in the French Occitanie region (ex Languedoc-Roussillon region, 225 km coastline) which is directly influenced by the Rhone river and therefore has fairly turbid waters. This coastline is artificialized to the extent of 17.06 % (MEDAM). Most of this artificialization is the result of an interministerial mission known as "Mission RACINE" aiming to shift the regional economy from cheap wine growing to tourism. In 20 years (1963-1983), mosquito-infested areas were drained and six tourist units were built on almost virgin land, this led in return to the highest demographic growth in France (between 1982 and 2012) through the development of nearby cities (Montpellier, Béziers, Narbonne and Perpignan) (Sagnes, 2001).

The past distribution of *P. oceanica* meadows was estimated mixing bibliographic sources (Pergent et al., 1985), surveys targeting old people with a good knowledge of local seabed (fishermen, divers) and old aerial photographs (the oldest was from 1935) analyzed by the method described by (Holon et al., 2015) and (Pasqualini et al., 1998) for seagrass beds above -20 m. Old aerial photographs were obtained from remonterletemps (2021) and Sextant (2021). The questionnaire consisted of 34 semi-structured questions, 19 open questions and several SHOM charts allowing respondents to draw the former distribution areas of the meadows. Respondents were also asked for personal data concerning the age, past and current activity, and hobbies, their knowledge degree about *P. oceanica* and other seagrass beds within the study area, the major changes that have affected the coastline and their impacts on the seagrass and underwater fauna, and finally their proposals to reduce the observed impacts. The *P. oceanica* distribution map obtained from old aerial pictures was compared to the participatory maps obtained from the surveys to analyze concordances and discordances. All maps were then combined to build a most complete as possible single map of areas former covered by *P. oceanica*. This latter was compared to the current distribution map (Donia expert 2021) of *P. oceanica* and dead matte (what remains after the death of *P. oceanica*) to analyze the spatial changes that occurred over the last 20 to 70 years. Maps were built and compared using ArcGIS software version 10.3.

Five metrics were chosen for the analysis of the current and old seascapes patchiness: the mean patch area, the area-weighted perimeter-area ratio, the landscape division index, the number of patches (Sleeman et al., 2005) and the cohesion index (Houngnandan et al.,

2020). These landscapes indices were calculated using The FRAGSTATS software version 4.2, first developed by (McGarigal and Marks, 1995), at class and seascape level using an eight cells neighborhood rule coupled with no specific sampling strategy to consider the whole part of meadow selected (Abadie et al., 2015).

Results

In total 22 responses were obtained in spring 2018 and 14 in spring 2021. The respondents were between 44 and 91 years old but in majority older than 60.

Our results confirm that some areas never harboured *Posidonia* meadows because the local conditions are not favorable (i.e. lagoon outlets, fine sandy bottoms, hydrodynamics), such as the sandy coast from Argelès-sur-mer to Vias. On the other hand, observations have been made on the disappearance of *Posidonia* meadows in certain areas, such as off Frontignan, where an entire part of the meadows has disappeared to make way for dead matte, visible or buried (Fig. 1). In Carnon 99 % of the known meadows have disappeared (Fig. 1). In the last 70 years *Posidonia oceanica* meadows experienced an overall regression of 65% (409.8 ha) in the region.

For all areas, the landscape indices showed that *P. oceanica* seagrass knew a process of fragmentation between 1954 and 2014: reduction in the number of patches (-5 to -64%), in the average patch area (-5 to -96%) and in the cohesion between patches has (0 to -36%). On the other hand, the fragmentation of the seagrass beds was linked to an increase in the complexity of patch's shape (0 to 85%) and a higher fragmentation of the landscape (0 to 63%). The main causes for the lost areas and fragmentation cited during the surveys were coastal development, trawling, pollution and anchoring. At the same time, all the respondents noted a decrease in fish populations over the last 70 years, the causes of which, according to them, were: overfishing (35%), water quality reduction (31%), and the decline of the *P. oceanica* meadow (16%).

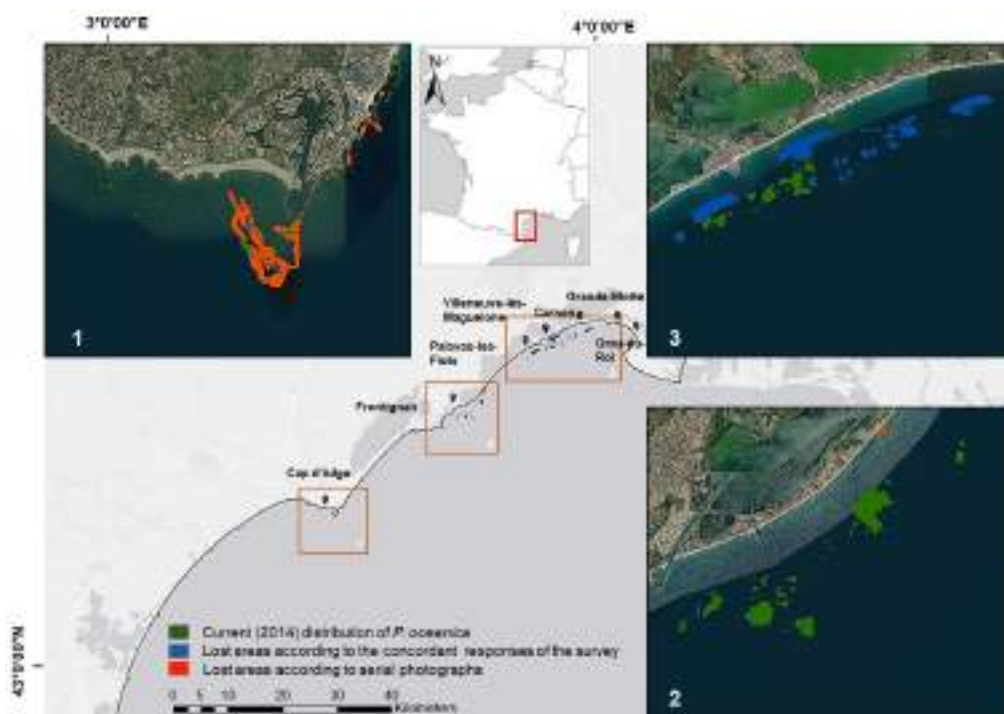


Fig 1. Map showing the entire study area and the distribution of *P. oceanica* according aerial photos, survey data and current distribution. Data concerning the current distribution were obtained from the MEDTRIX platform (<https://plateforme.medtrix.fr>, Project DONIA expert)

Discussion and/or conclusions

While the use of aerial photographs was already demonstrated to assess the decline and degradation status of seagrass beds (Holon et al., 2015; Nahirnick et al., 2019), the present study showed the interest to combine it with a complementary survey approach. Although the old photographs were too patchy to allow complete mapping because their acquisition program focused on lagoons and coastlines and not on seagrass beds, they confirmed and/or filled in the gaps in the participatory maps. Moreover, the cartographic recollections of the respondents were surprisingly accurate and consistent. Given the age and experience of the people, it is important to have this kind of information that could have been lost. The low number of people in the sample showed the rarity of the data and the importance of having this participatory mapping.

We showed for the first time that *P. oceanica* was anciently well developed in the region. Our study made it possible to quantify its distribution along 225 km of the western French Mediterranean coastline. The decline in covered area was huge: 65 % in mean (0.03 to 99 % depending on the sites) in 70 years for the upper part of the meadows i.e. 409.8 ha. This percent is similar to that estimated by Holon et al. (2015), i.e. 73 % in 85 years, estimated in the eastern French Mediterranean coastline for the upper part of the meadows. However, it is more than the global decline (13-38%) found by a comparative study in Mediterranean (Marbà et al., 2014).

In addition to the reduction of the covered area, this process of degradation, was also manifested in terms of configuration by a decrease in the number of patches, cohesion between the patches, changes in shape and an increased fragmentation. This confirms the usefulness of habitat fragmentation indices to detect the decline of habitats (Abadie et al., 2015; Houngnandan et al., 2020; Sleeman et al., 2005).

Such a fragmentation and huge lost in covered area should have produced important ecological and economic consequences given the essential roles of *P. oceanica*. The 22 respondents of this present study all agreed to affirm that according to their personal experience, fish abundance (number, size and weight) had decreased as confirmed by (Chaboud et al., 2015). In addition, the decline of *P. oceanica* beds could explain the increased coastal erosion observed over the last decades in the region (Boudouresque et al., 2012) like for example in the bay of Aigues-Mortes where a significant attenuation of the waves was measured where *P. oceanica* seagrass was still present (Leredde et al., 2016).

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Fabrice HOUNGNANDAN, FERY C., BOCKEL T., DELARUELLE G., BOISSERY P., KEFI S., DETER J.
MARBEC, Univ Montpellier, CNRS, IFREMER, IRD, Montpellier, France
E-mail: houngnandan@gmail.com

A HIERARCHICAL SCALE-BASED APPROACH FOR INFERRING THE ECOLOGICAL QUALITY OF A MARINE ECOSYSTEM FROM ITS SPATIAL COMPOSITION AND CONFIGURATION

Abstract

*Numerous indicators exist to measure changes in marine water quality and evaluate whether progress towards these objectives has been made; some focus on seagrass meadows, which are key marine ecosystems. This study aimed to analyse the relationships between different indicators used to evaluate the environmental status of marine waters. We used data from *Posidonia oceanica*, which conservation status was monitored by biotic indicators (BiPo “Biotic index based on *P. oceanica*” and PREI “*P. oceanica* rapid easy index”), and its spatial distribution was described through landscape indicators: one composition index (the decline) and three configuration indexes (patch cohesion, landscape shape and landscape division). We found a strong and significant linear relationship between the two biotic indicators. These results suggest a way of optimising the acquisition efforts and sampling strategy for the evaluation of the environmental status of marine waters using data from seagrass meadows*

Key-words: Ecological indicators, habitat condition, decline index, conservation status, good environmental status

Introduction

Human activity is the primary driver of current biodiversity loss worldwide (Jungblut, 2020). A growing world population (expected to reach 9.7 billion in 2050 and could peak at nearly 11 billion around 2100, United Nations, 2019, <https://www.un.org/>) and increasingly destructive lifestyles threaten all oceans and seas (Halpern *et al*, 2019; 2015) and lead to a decline in global marine life. It is therefore urgent to effectively track the environmental status of ecosystems, in order to detect, stop and, if possible, reverse these losses by appropriate management actions (Martínez-Crego *et al*, 2010). Monitoring programmes aiming to assess the conservation status of seagrass meadows are increasing worldwide (Martínez-Crego *et al*, 2008; Orth *et al*, 2006; Short *et al*, 2006). They use a wide diversity of indicators representing different structural, functional and trophic levels and different spatial scales (Marbà *et al*, 2013). Because of their abundance, wide distribution along the Mediterranean coast and sensitivity to anthropogenic pressures (Balestri *et al*, 2003), *P. oceanica* meadows are a biological quality element used to assess coastal water quality in the European Union Water Framework Directive (WFD; EU, 2000).

Therefore, a first approach to assessing coastal water quality consists of using a range of European indicators based on the condition of the *P. oceanica* habitat, such as, for example, the *P. oceanica* multivariate index (POMI, Romero *et al*, 2007), the biotic index based on *P. oceanica* (BiPo, Lopez y Royo *et al*, 2010), the *P. oceanica* rapid easy index (PREI, Gobert *et al*, 2009) or the PosWare (cited as PoSte in some publications) (Buia *et*

al., 2004). A second approach consists of measuring the areas covered by seagrass meadows, based on maps on which landscapes indicators, i.e. configuration and composition indicators, can also be calculated: the decline index (Holon *et al.*, 2018; Houngnandan *et al.*, 2020; Moreno *et al.*, 2001), the cohesion index (Moore *et al.*, 2011; Prada *et al.*, 2008), the landscape shape index (Teixidó *et al.*, 2007) and the landscape division index (McGarigal, 1994; Sleeman *et al.*, 2005). The indicators of the first approach provide a punctual synthetic measure of ecological quality, based on a combination of local metrics. In contrast, landscape indicators allow assessment on a large spatial scale. They can be calculated on the complete spatial distribution of the species if maps are available.

Both approaches present advantages and inconveniences, although each one requires significant implementation efforts. However, the abundance of existing indicators makes it difficult for managers and stakeholders to choose the best indicators according to the objectives to assess the ecological quality of water bodies in Mediterranean Sea. Logically, the cost–efficiency ratio is considered in the context of limited means in terms of people and/or money.

The objective of this work was to analyse the relationships between biotic indicators and landscape indicators used to assess the environmental status of marine waters. To achieve this, we studied *P. oceanica*, a species and marine habitat for which various biotic indicators have been surveyed along the French coastline since 2014 and for which an entire 1:10000 distribution map is available for the calculation of landscape indicators. Landscape indicators were calculated at different scales around 47 study stations to identify the scale at which the best correlations could be obtained. We aimed to contribute to a better understanding of the relationships between the different indicators, in order to optimise the acquisition efforts and sampling strategies for evaluation of the environmental status of marine waters. Indeed, if landscape indicators accurately reflect biotic indicators, they could be used at the scale of the entire distribution of the habitat and increase what is known about the habitat studied.

Materials and Methods

Study area

This study was carried out along the 1700 kilometres of French Mediterranean coastal areas including Corsica (see Fig. 1). Three regions composing the French *P. oceanica* distribution area were surveyed: the (i) Occitanie, (ii) Provence-Alpes-Côte d’Azur (PACA) and (iii) Corse regions, between a depth of 0 and 40 m, which is the bathymetric growth limit of *P. oceanica* seagrass in France (Boudouresque *et al.*, 2012).

Sampling design and data acquisition

Biotic indicators based on P. oceanica vitality parameters

As part of the TEMPO program (data available at <https://plateforme.medtrix.fr>), PREI and BiPo monitoring are conducted during June every year in one of the three regions outlined in section 2.1. Three consecutive years are thus needed to obtain comparable data for the entire monitored area. We used data from 2014 to 2019, which corresponded to one year of survey per region. During this period, a total of 47 stations were surveyed at a depth of -15 m along the French Mediterranean coast.

Landscape indicators based on marine habitat distribution

The spatial distribution data of *P. oceanica* beds and dead matte (which is what remains of the plant after its death) come from the 1:10 000 map of 11 marine habitats available

on the MEDTRIX platform (<https://plateforme.medtrix.fr>, Project DONIA expert, see Holon *et al.* (2015) for details concerning data and map building).

Around each sampling station, we defined joined cells of identical sizes representing the different scales using the ArcGIS software version 10.07. At all six different scales (i.e., cell dimensions) were used for the analyses: 100 m × 100 m, 200 m × 200 m, 400 m × 400 m, 600 m × 600 m, 800 m × 800 m, and 1000 m × 1000 m. Four landscape indicators were calculated for each scale, from the rasters of the marine habitats maps to characterise the landscape configuration of *P. oceanica*: i) the landscape division index, ii) the landscape shape index, iii) the cohesion index and iv) the decline index. These indicators were computed with the SDMtools package (Vanderwal *et al.*, 2019) using the “R” statistical software (R Core Team, 2018). For each index, values are expected to vary according to the conservation status of the habitat.

Statistical analyses

Correlations between landscape and biotic indicators at different scales

A Pearson correlation was used to assess the relationship between the different biotic indicators because the data fitted a Gaussian distribution, while a Spearman correlation was used to assess the relationship between landscape indicators calculated at different scales and biotic indicators, because the distribution between them did not fit a particular distribution and were not Gaussian. The strengths of the correlations (Pearson and Spearman) were evaluated as recommended by Akoglu (2018) and Schober and Schwarte (2018). The cell dimension for which we identified the strongest correlation between two different indicators, and for which this correlation was the most significant in terms of P-value (<0.05), was used to explore the nature of the relationship (see below). Correlations with values inferior to 0.4 were not considered further.

Use of the landscape indicators to predict the biotic indicators of the meadow

The relationships between biotic indicators (variables to be predicted = BiPo, PREI) and landscape indicators were explored using simple linear regression “LR” models and non-linear models (i.e., polynomial regressions “PR” and generalized additive models’ «GAM”). The choice of the best model was based on the AIC (Akaike Information Criterion) estimated by the maximum likelihood method. The AIC is defined as: $AIC = -2 \log(L) + 2K$; where L is the maximised likelihood and K is the number of free parameters in the model. With this criterion, the model deviation is penalised by twice the number of free parameters. The best model is therefore the one with the lowest AIC (Parzen *et al.*, 1998).

Results

Correlation between the biotic indicators

The Pearson correlation between PREI and BiPo was strong and highly significant ($r=0.76$; $P=2.1 \times 10^{-4}$). The relationship was best described by a polynomial regression model (F11).

Correlation between biotic and landscape indicators

The analysis of the correlations between biotic and landscape indicators for all the scales tested showed that only PREI correlated significantly with all landscape indicators. Moreover, these correlations were all found at the scale of 1000 m × 1000 m, except for the decline index, where the highest and most significant correlation was found at 200 m × 200 m. In addition, the BiPo presented moderate and significant correlations with all landscape indicators except the landscape shape index; these correlations were found at the 600 m × 600 m scale and the 800 m × 800 m scale.

The values of the biotic indicators both increased with decreasing decline index and landscape division index values. In contrast, the values of the patch cohesion index and the landscape shape index increased with the values of the biotic indicators. The strongest relationships observed were between PREI and the decline index at a scale of 200 m × 200 m (adjusted $R^2=0.82$) and between the PREI and the patch cohesion index at a 1000 × 1000 m scale (adjusted $R^2=0.48$). The lowest explained variance ($R^2=0.0006$) was between BiPo and the patch cohesion index at a scale of 600 m × 600 m.

Discussion

Landscape indicators accurately reflect habitat condition

The objective of this study was to quantify the strength and nature of the relationships between landscape indicators estimated at different scales and biotic indicators calculated for a key species, *P. oceanica* seagrass. To our knowledge, no study had studied these relationships despite the relevance of the question.

Using a hierarchical approach based on the estimation of landscape indicators at different scales (6) our results showed that there is a strong and significant correlation between landscape indicators (4 indicators calculated at 6 different scales) and biotic indicators firstly and secondly between biotic indicators: PREI and BiPo. The strong and significant relationship found between PREI and BIPO is consistent with what Gerakaris *et al.* (2017) showed and is explained in particular by the fact that the vitality parameters used in their estimation are almost the same at the lower limit. The same author had also shown that PREI and BiPo accurately measure the cumulative pressures impact (Gerakaris *et al.*, 2017) on the *P. oceanica* meadow as previously shown by Gobert *et al.*, (2009) for the PREI and Lopez y Royo *et al.* (2010) for the BiPo. More recently, Güreşen *et al.*, (2020) have also confirmed the effectiveness of BiPo in large-scale assessment of *P. oceanica* meadows conservation status. We also showed that PREI was the biotic index having a strong and significant relationship with all landscape indicators, and all these strong correlations were observed for large scales. More specifically, composition (decline index) and configuration (patch cohesion index, landscape shape index, landscape division index) accurately reflected the *P. oceanica* habitat conditions estimated by PREI and BiPo, mostly using relatively large scales (800 m × 800 m to 1000 m × 1000 m). Our results confirm the observations of previous studies (Abadie *et al.*, 2018; Güreşen *et al.*, 2020a) on landscape indicators relevance in the monitoring of seagrass meadows. This means that we could calculate these landscape indicators over large study areas – in fact everywhere where a distribution map is available – and thereby obtain information on habitat conditions. For calculation of these landscape indicators the scale and map resolution are critical, because they influence ecological processes and statistical relationships (McGarigal *et al.*, 2002).

Because of the multitude of indicators used to assess the status of coastal marine ecosystems and their possible redundancy, in this study we quantified the strength and nature of the relationships between biotic and landscape indicators using a data set from *P. oceanica* seabeds in the Mediterranean Sea. Our results, obtained at around 15 m (the sampling depth for biotic indicators) along the entire French coastline, need to be confirmed in other countries and at other depths. This study further underlines the complementary nature of data acquired within the framework of the WFD and the multiple descriptors and criteria targeted by the MSFD to assess coastal water quality and environmental status of marine waters.

The simultaneous use in *P. oceanica* studies of several indicators that provide complementary information still seems to be the best strategy to meet different management objectives.

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Sara KALEB, DESCOURVIÈRES E., BANDELJ V., CIRIACO S., ORLANDO-BONACA M., FALACE A.

DSV – University of Trieste, Italy

E-mail: sara.kaleb@gmail.com

EX SITU PILOT EXPERIMENT TO ASSESS THE FEASIBILITY OF ECOLOGICAL RESTORATION OF *FUCUS VIRSOIDES* IN THE GULF OF TRIESTE (NORTHERN ADRIATIC)

Abstract

Fucus virsoides J. Agardh, which is endemic to the Adriatic Sea, has experienced dramatic declines or local extinctions. The lack of adult plants producing recruits hinders the natural recovery of impacted areas. Therefore, active restoration is recommended to restore populations within a reasonable time frame.

In this study, an *ex situ* pilot experiment was carried out by outplanting cultivated recruits at two sites (Miramare MPA in Italy and Piran in Slovenia) to test the efficiency of seedling culture in the laboratory, the effectiveness of an intermediate step in the field in hanging structures, and subsequent anchorage on the rocky substrate by quantifying germlings cover and growth.

This study extends the biological knowledge of the early life and juvenile stages of *F. virsoides* and provides some practical considerations for implementing restoration measures.

Key-words: *Fucus virsoides*, *ex situ* restoration, fucoid, macroalgae, marine forests

Introduction

Canopy-forming algae of the order Fucales and Laminariales (Phaeophyceae) are among the most ecologically and socio-economically valuable marine species in temperate waters. They provide a structural and trophic framework that supports rich biodiversity (Fabbrizzi *et al.*, 2021). *Fucus* species are brown macroalgae that live in the intertidal of rocky coasts and provide a number of important ecosystem services, including high primary productivity and carbon sequestration, shelter and nursery for fauna, direct sources for many traditional and commercial uses, aesthetic beauty and intrinsic evolutionary value (van den Hoek *et al.* 1995, Rindi & Guiri 2004; Frederiksen *et al.*, 2005; Watt & Scrosati, 2013). They also play a central role in the growth of understory assemblages by influencing light, desiccation, water movement and the rate of transport and deposition of suspended sediments.

Fucus virsoides J. Agardh (Fucales, Phaeophyta), endemic to the Adriatic Sea (Linardić, 1949; Giaccone & Pignatti, 1967; Munda, 1972), is a glacial relict that has survived in the Adriatic Sea thanks to the specificities of this basin (Munda, 1972, Orlando-Bonaca *et al.*, 2013). In the past, its range extended from the Venice Lagoon to the Boka Kotorska (Mačić, 2006).

Recently, it has experienced dramatic declines or local extinctions (e.g. Battelli, 2016; Falace *et al.*, 2010; Mačić, 2006; Orlando-Bonaca *et al.*, 2013). Due to its endangered status and ecological role, it has been included in the "List of Threatened or Endangered Species" of the Barcelona Convention (amended Annex II of the Protocol on Specially Protected Areas and Biological Diversity in the Mediterranean, UNEP; 2019).

This study aimed to improve the knowledge of the early life stages of *F. virsoides* by studying embryonic development to develop an *ex situ* protocol to investigate the

possibility of reintroducing this species on the Italian and Slovenian coasts of the Gulf of Trieste through a pilot project.

Material and methods

Mature receptacles of *F. virsoides* were harvested in winter 2022 at low tide in the tidal marsh of Marina Julia (northern Adriatic - eastern Mediterranean) (45°46'37.7 "N 13°32'02.6 "E), which represents the last known population in the Gulf of Trieste (Fig. 1).



Fig. 1: Map showing the geographical location of the donor site, the laboratory culture site, the lantern net culture site, and the receiving sites in northern Adriatic.

As this population is threatened with extinction, only a limited number of receptacles were collected for these preliminary trials.

The receptacles were taken to the laboratory of the University of Trieste, where they were rinsed with seawater to remove epibionts and stored in the dark at 4°C within 1 hour after collection.

A first culture was carried out to test the best conditions (i.e., temperature and culture medium) for seedling growth. According to Munda & Kremer (1977), the optimum temperature for *F. virsoides* growth is probably higher than 15°C, so two temperatures were tested: 15°C (T-) and 22°C (T+). Two culture media were also tested: von Stosch's enriched filtered seawater (VS) (Falace *et al.*, 2018) and Provasoli Enriched Seawater (PES) (Munda, 1977). The photoperiod was set to a 15:9 hour light-dark cycle and the light irradiance to 125 $\mu\text{mol photons m}^{-2}\text{s}^{-1}$. Light was supplied by LED lamps (AM366 Sicce USA Inc., Knoxville, USA) and irradiance was measured using a LI-COR LI - 190/R photometer (LICOR-Biosciences, Lincoln, NE, USA).

For each condition, two mature receptacles were placed in 5 Petri dishes. After 24 hours, the zygotes were released, and the receptacles removed. Ten days after release (AR), the number of seedlings at different growth stages (Fig. 1) was counted by processing photographic data. For each Petri dish, 5 randomly selected areas (1278 mm^2) were photographed using a Canon Powershot G9 on an inverted microscope (Leica, DM IL LED). To analyze the differences among conditions, a PERMutational multivariate ANalysis Of VAriance (PERMANOVA) (Anderson, 2001) was applied using the percentage of individuals at each stage as a response variable and conditions as factors.

A second culture was then established based on the results of the first experiment. Ten clay tiles were covered with two receptacles each and cultivated in 4L aquaria in triplicate. The aquaria were filled with VS and the culture medium was renewed every three days to minimise nutrient limitation. The culture medium was oxygenated with pumps (Mimouse Circulation Pump SICCE) and aerators (Airlight Silent Air Pump 3300 SICCE). The laboratory culture lasted 14 days. The tiles were then transported to the C-

zone of the Miramare MPA, where they were attached to hanging lantern nets (Savonitto *et al.*, 2021) in the intertidal zone for 4 months. Tile coverage and thalli length were measured after 16 days (T1), 1 month (T2), 3 months (T3) and 4 months (T4). Subsequently, half of the tiles were placed on the rocky shores in the A zone of Miramare MPA (Italy) and the other half in Piran (Slovenia) (Fig. 1).

Tile cover and thalli length were measured after 1 month (T5) and 2 months (T6). The tile cover has been calculated with the tile where *F. virsoides* was still present ($n = 14$ at T5 and $n = 11$ at T6). Germlings were observed with a stereo microscope (Leica, MZ 6) and photographed with a Nikon Coolpix 4500 camera. The juvenile length and cover measurements were carried out with the software ImageJ (Schneider *et al.*, 2012).

Results

Laboratory cultivation

The percentage of individuals was calculated for each of the four developmental stages observed in the Petri dishes (Fig. 2). PERMANOVA confirmed significant differences in embryonic development between the two temperatures tested (p-value < 0.001), with the T- condition characterised by a higher number of more developed individuals (stage IV) than T+. Conversely, no significant differences were found between the two culture media (VS, PES) or within the interaction terms.

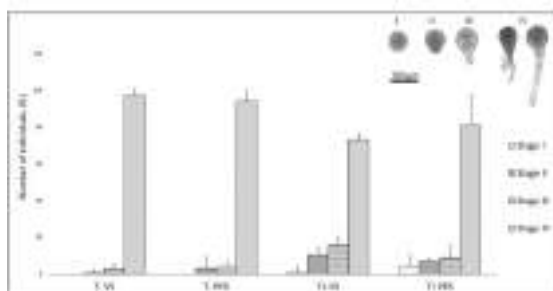


Fig. 2: Percentage of embryo developmental stages after 10 days under the different conditions.

Lantern net and outplanting in the field

After two weeks in the lantern net (T1), thalli were $0.39 \text{ mm} \pm 0.06 \text{ SD}$ long and $0.12 \text{ mm} \pm 0.01 \text{ SD}$ wide, while after four months of suspended culture (T4), they reached an average length of $4.94 \text{ mm} \pm 0.97 \text{ SD}$ and a width of $1.63 \text{ mm} \pm 0.42 \text{ SD}$, with a maximum of 6.53 mm (Fig. 3). The average cover at T4 was $44.26 \% (\pm 17.14 \text{ SD})$ (Fig. 4). One month after outplanting on the rocky intertidal in Piran (T5), thalli increased rapidly in size, reaching a maximum length of 11.47 mm and a width of 4.87 mm , with a mean length of $6.11 \text{ mm} \pm 2.07 \text{ SD}$ and a mean cover of $50.44 \% (\pm 22.78 \text{ SD})$. At T6, a reduction in cover was observed ($44.49\% \pm 23.23 \text{ SD}$), probably due to grazing. Indeed, signs of grazing were visible on many juveniles (Fig. 5). Nevertheless, the average length was $7.81 \text{ mm} \pm 2.46 \text{ SD}$ with a width of $2.49 \text{ mm} \pm 0.81 \text{ SD}$. The frequency of clay tiles with *F. virsoides* was $92,86\%$ at T5 and $78,57\%$ at T6.

In contrast, no thalli were found in Miramare MPA at T5.

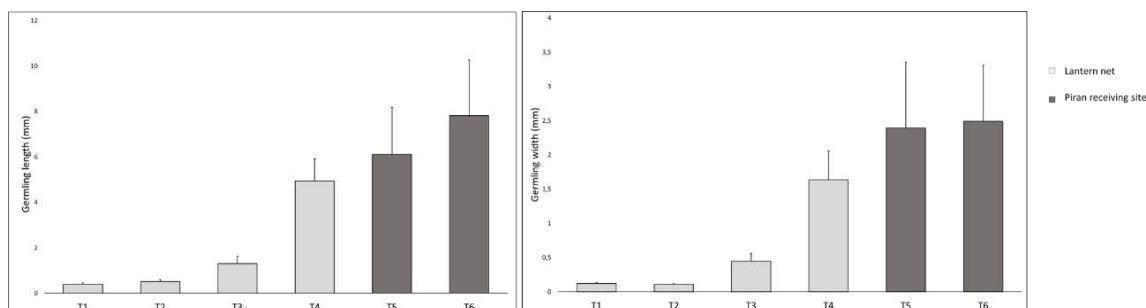


Fig. 3: Thalli length (mm) in the lantern net (light gray) and attached on the intertidal rocky shore at Piran (dark gray). Juveniles were moved to the receiving site after 4 months of suspended culture in lantern net.

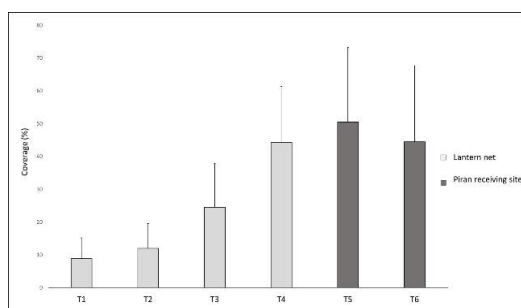


Fig. 4: Percent coverage of germlings in lantern net and at the Piran receiving site.

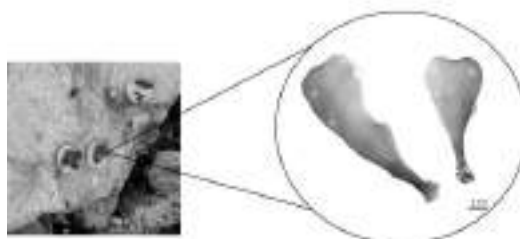


Fig. 5: Juveniles of *F. virsoides* at the Piran receiving site two month after outplanting (T6).

Discussion

F. virsoides is a glacial relict (Giaccone, 1991) and evolutionarily closely related to *F. spiralis* (Coyer *et al.*, 2006). Although tolerant to variations in many environmental factors (such as salinity, wave exposure, etc.) and once one of the most abundant intertidal species (Orlando-Bonaca *et al.*, 2013), it has completely disappeared from Slovenian coasts (Battelli, 2016). It has drastically declined in many areas of its range due to dystrophic conditions, grazing, emerging pollutants, and probably also climate change (e.g., Sfriso & La Rocca, 2005; Mačić, 2006; Munda, 2008; Falace *et al.*, 2010; Battelli, 2016; Felling *et al.*, 2019). Therefore, the inclusion of this species in the IUCN Red List, which classifies it as "vulnerable", as proposed by Verlaque *et al.* (2019), could allow its conservation and management to be prioritized to prevent the extinction of this endemic species.

The development of early life stages of *F. virsoides* was favoured by the lowest temperature (*i.e.*, 15°C), at which 96.85% (VS) and 94.05% (PES) of individuals were well developed (stage IV).

The suspended culture in the field allowed to reduce the culture time required for the seedlings to reach a "refuge size" (Savonitto *et al.*, 2021), thus avoiding bacterial or

microalgal outbreaks common to prolonged culture in mesocosms (Orlando-Bonaca *et al.*, 2021; Lardi *et al.*, 2022).

At the Miramare site, no thalli survived one month after outplanting. As observed in previous restoration experiments here, the sharp decline in macroalgae has not only affected the Fucales, as the benthic system has shifted into an alternative state dominated by turf algae and other grazing-resistant species (Falace *et al.*, 2010; Bevilacqua *et al.*, 2022).

In the Piran receiving site, a decrease in the frequency (14,29 %) and in the coverage (5,95 %) of *F. virsoides* probably due to mollusc feeding was observed in July. The anomalously high temperatures recorded throughout the Adriatic basin during the summer of 2022 may have affected the cold-affinity *F. virsoides* germlings making them more prone to grazing by limpets.

Control of grazers with protective cages is not feasible in the intertidal zone, but removal of grazers before planting and regular maintenance could be a suitable solution.

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Onur KARAYALI, AKÇALI B., BİZSEL K.C., KABOĞLU G.

Department of Marine-Inland Water Sciences and Technology, Institute of Natural and Applied Science Ege University, Faculty of Fisheries, Erzene Mah., 35100 Bornova Izmir, TURKIYE

E-mail: onurkarayali@gmail.com

NEED OF REFERENCE CONDITIONS FOR COMPARABLE MONITORING OF *POSIDONIA OCEANICA* AT ECO-REGIONAL SCALE INCLUDING THE SLIDING BASELINE SYNDROME: A DISCUSSION OVER FOCA SEPA

Abstract

Posidonia oceanica meadows are one of the most important habitats to be protected in the Mediterranean Sea. Several studies and monitoring programs have been carried out over the last decades. As a result, i) a comprehensive guideline and protocol on monitoring has been developed, ii) the progressive performance for implementing them in the Eastern Mediterranean could not match those in the Western Mediterranean, and iii) the problems in determining the baseline (reference) conditions could not be solved yet. The purpose of this study is to highlight and discuss the challenges in assessing reference conditions of *P. oceanica* meadows in the Foca Special Environmental Protected Area (SEPA, Turkiye) by comparing the variations in the upper limits of the meadow between 2005 and 2019. While studying the detected regression, uncertainties in determining or even using baseline (reference) conditions have been observed. This regression seems to indicate the sliding baseline syndrome (SBS) even though the area has been a SEPA since 1990. Considering that the uncertainties are driven by the SBS together with the discrepancies and failures in monitoring, it is safe to assert that the issue is not unique to this particular site. Therefore, this study underlines the significance of spatial representability, which should be based on an appropriately reliable scale, i.e., eco-regional scale. In this way, the baseline (reference) conditions can be determined by using the more realistic and reliable variations driven by natural and anthropogenic impacts within the same scale. A strategic approach for determination of eco-regions within the Mediterranean Sea by using recently emerged technologies has been emphasized in order to obtain and implement reliable and comparable baseline conditions for improving efficiency in monitoring and management.

Key-words: Mediterranean Sea, seagrass regression, GIS, MPA, habitat loss, reference conditions

Introduction

Among marine ecosystems, seagrass meadows are thought to provide more than half of the total value of the world's natural capital and services (Costanza *et al.*, 1997; Holon *et al.*, 2015). *Posidonia oceanica* L. (Delile) meadows are among the prominent, as well as vulnerable habitats under stress, mainly due to critical functional roles in coastal ecosystems such as protection against erosion, turbidity, de-oxygenation, whilst creating and maintaining spawning and nursery grounds for diverse biological communities, in spite of their slow growth properties (Boudouresque *et al.*, 2012; 2016). Any change that has a negative impact on the environment cause a stress on the meadow health and hence on its distribution patterns and area (Marba *et al.*, 2006; Madden *et al.*, 2009; Conte *et al.*, 2021). Being distributed over the upper shelf area along coasts, *P. oceanica* meadows

have been exposed to intensive human exploitation, especially after the second half of the 20th century (Boudouresque *et al.*, 2012; Pergent-Martini *et al.*, 2021), such as eutrophication, turbidity, pollution, and coastal erosion resulting in severe loss of coastal habitats and biodiversity (Benoit & Comeau, 2005; Marba *et al.*, 2014). Currently *P. oceanica* is classified as a ‘Least Concern’ species on the IUCN Red List (IUCN, 2021), and as recognized and cited in several international (*e.g.*, Habitats Directive, Bern, and Barcelona Conventions), as well as national legislations.

The monitoring studies on *P. oceanica* meadows started in 1980 at the French Riviera (Boudouresque *et al.*, 2000; Lopez y Royo *et al.*, 2010) and then promptly spread in almost all the Mediterranean countries, particularly along the Western Mediterranean coasts. On the contrary, in the Eastern Mediterranean coasts these efforts did not reach a similar momentum in their monitoring performances. In addition, the analysis by Lopez y Royo *et al.* (2010) has revealed a number of issues concerning data quality, reliability, and comparability in the Western Mediterranean efforts, which still remains unsolved. Particularly, the information about historical reference points and consequences of ongoing pressures are essentially important in pursuing a sustainable path. Recently, the problem of the “sliding or shifting baseline syndrome (SBS)” (Montefalcone *et al.*, 2015), which affects the monitoring efforts, has demonstrated the deadlock state of the challenges towards a successful monitoring and management for protection and conservation of *Posidonia oceanica* meadows. This study is to explore these challenges over our experiences in Foca SEPA and discuss on the possibilities for overcoming them by benefiting from the recent technological advancements on observation systems.

Materials and Methods

Foca, located in the mid-eastern Aegean coasts of Turkiye (Fig. 1), is a touristic destination and one of the major fishing towns in the region. At the north-east edge of the Izmir Bay, it has complex marine morphological characteristics due to the formation of nearshore islands and rocks (Fig. 1). It was declared as one of the coastal/marine SEPAs of Turkiye in 1990 (Anonymous, 1990), and its boundaries were enlarged to its present borders in 2007 (Anonymous, 2007). The upper limits of the *P. oceanica* meadows in Foca have been mapped in 2005 and in 2019 and they have been compared by overlay analysis on a Geographic Information System (GIS). The 2005 distribution was mapped using high resolution satellite imagery, whereas the 2019 distribution was mapped using high resolution side scan sonar imagery, both with ground-truth data. Details on the mapping methodologies can be found in Akçalı *et al.* (2019) and in SPA/RAC–UN Environment/MAP (2020), respectively. The meadow regression in three selected locations (distinguished according to the exposure and to the occurrence of some prominent human impacts) was calculated in GIS by means of linear distances and as the total lost area (Fig. 1).

Results and Discussion

In Location 1, the disappearance of some existing patches and the regressed upper limit corresponds to a total loss of 13790 m². In Location 2, the disappearance of some existing local patches corresponds to a total loss of 11426 m². In Location 3, the regressed upper limit and increased patchiness in the meadows corresponds to a total loss of 12166 m². In Location 1, the loss seems to be attributed to the intensive recreational activities along the beach. In Location 2, the reason seems to be the coastal structures built, while the main impact is likely to be the anchoring in the Location 3, since the zone is one of the

most preferred anchoring spots for the touristic boats during tourist season. However, there are also some regressed areas that cannot be attributed to any of such human impacts. For instance, shores of the Foca SEPA have some segments that are exposed to waves with varying long fetch distances, while some others are quite enclosed and sheltered at varying degrees by the capes, islands, and rocks. Such environmental features jeopardize efforts for inferring how much of the observed regression and deterioration in the upper limits of the meadows is due to the human impacts. The resulted uncertainties on the reference status and depth of the upper limit of *P. oceanica* meadows have important consequences for coastal zone management, since they are fundamental in the current protocols for seagrass monitoring activities (Montefalcone, 2009; Boudouresque *et al.*, 2012). Then, reliable reference conditions to track their evolution under natural and anthropogenic influences are needed. This is much more crucial when the monitoring is carried out in a protected area that potentially and expectedly plays a role as a potential reference site providing baseline conditions for local and regional or even for the Mediterranean wide assessments. All these uncertainties have undoubtedly suggested a need for a more efficient sampling design based on the appropriate spatial scale representing spatial variation, well selected set of descriptors, and well-defined objectives, *i.e.*, conservation, water quality and mapping, satisfying the targeted ecological and managing expectancies.

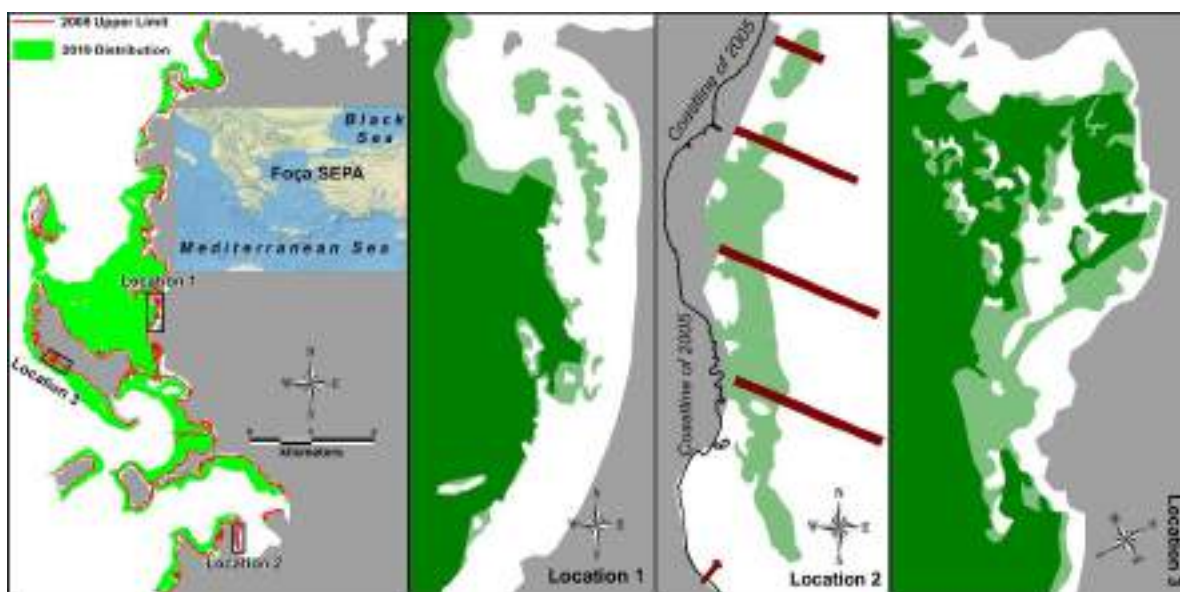


Fig. 1: Foca SEPA in Türkiye and the three selected locations for comparing the upper limits observed in 2005 and 2019.

As Lopez y Royo *et al.* (2010) identified, there are a number of issues concerning data quality, reliability and comparability in monitoring efforts. Each may have varying effects by introducing errors at their own relevance and this is particularly important in assignment of the baseline reference condition for descriptors and hence in data comparison. The risks caused by such issues inevitably magnify when they have been applied to larger areas as management measures. Moreover, the SBS appeared due to the climate change driven stresses, and this describes the incremental lowering of ecological standards. Assessing long-term changes become a dilemma and thus a major concern because an already-degraded-environment status could be accepted as a reference, as emphasized by Montefalcone *et al.* (2015). Those related with spatial representability of

the data and results seem the most essential and critical one for achieving efficient management results for securing the future of the meadows. In comprehensive discussions in Lopez y Royo *et al.* (2010), all those issues were addressed and for the potential importance of data analysis at eco-regional scale and hence monitoring efficiency were emphasized. However, the adoption of different sampling designs, spatial scales and sampling methods in different monitoring programs or efforts, as a fact, can particularly increase data variability and affect data reliability and interpretation, within and between monitoring programs. Moreover, their conclusive remarks on the difficulties in assessing the magnitude of the errors introduced at this stage, so that the quality assurance issues may limit the potential of *P. oceanica* as an ecological indicator at eco-regional level, was very appropriate and inspiring the way forward. Our main concern is exactly at this important and accurate inference since the difficulty to cope with the implementation of protection. Conservation measures based on well-developed scientific expertise into the managerial decision-making processes is the ultimate target that has still been challenging. Such processes by their nature require an area with defined boundaries. Considering the continuing debates and discussions on the definition of the boundaries of a protected zone, defining an eco-region sounds more challenging. In spite of the perceived magnitude of this challenge, the recent Global Ocean Observation Systems (GOOS) and related technologies have made possible the provision of up-to-date data offers new merits to deal with those challenges. Information on Mediterranean water masses via several platforms and models, *e.g.*, Copernicus, Poseidon, DieCast, offer the possibility to produce more resolute information and knowledge on formation and circulation patterns of the Mediterranean waters (Milot & Taupier-Letage, 2005). This is a promising progress towards defining eco-regions within the Mediterranean. Such information and knowledge, which have been continuously improving, have important potential in determining the eco-regions within the basins of the whole Mediterranean Sea, which enables the determination of reference (= threshold) values or ranges of descriptors that represent each eco-region reliably and comparably. Thus, the efforts for the protection and conservation of *Posidonia oceanica* meadows in the Mediterranean can improve their potential contribution to the management plans at national and international scales. Nevertheless, this should not lead us to overlook the other issues identified in Lopez y Royo *et al.* (2010), in particular for those on the sampling design, the selection of the set of descriptors, and the monitoring efficiency. The latter is highly critical since the many monitoring performance thresholds have usually been expressed as management objectives rather than ecological condition thresholds, *i.e.*, baselines (Hilton & Cook, 2022).

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Marie LA RIVIERE, DELAVENNE J., DE BETTIGNIES T., MICHEZ N., JANSON A.-L., ANDRES S., GAUDILLAT V.

PatriNat, OFB/MNHN/CNRS, France.

E-mail: mlariviere@mnhn.fr

BENTHIC HABITATS ID AND POLICY CROSS-LINKS TO IMPROVE MARINE CONSERVATION

Abstract

Identifying benthic habitat and providing clear definition of management units is central to the effective conservation of the marine environment. After the creation of a French marine habitat classification providing a comprehensive list of benthic habitats possibly occurring in national waters, we described each habitat unit (biocenosis and associated sub-units). At the same time, in order to facilitate the implementation of the Habitat Directive in the marine environment and clearly establish which habitat units should be considered as natural Habitat types of Community Interest (HCI), a national marine HCI definition was specified, as long as correspondences with habitats units of the national typology. These tools based on the best scientific knowledge available allowed to highlight knowledge gaps and will ease the assessment, management and reporting tasks both at local and national scales. By sharing these units' definitions with other Member States, and sharing the outputs of these projects internationally, we hope to facilitate the identification of common benthic habitat units and help to achieve a harmonised interpretation of HCI to be managed in the framework of the Habitat Directive.

Key-words: benthic habitats, classification, habitats directive, habitat units, description

Introduction

Identifying benthic habitat is central to the effective conservation of the marine environment. The use of habitats classifications as common language amongst decision-makers, managers and researchers is necessary to:

- define homogenous working units at local, national or regional scales;
- collect and share habitats occurrence and distribution trends data;
- identify which habitats should be monitored, assessed and managed under national or European regulations.

European directives and international conventions have developed their own list of habitats (European Commission, 2013; European Commission, 2017; OSPAR, 2008; PNUE *et al.*, 2006), but they do not encompass the variability of all habitats at the national level. A national marine habitat classification was thus created in order to provide a comprehensive and hierarchical list of benthic habitats possibly occurring in French waters for both seafronts: Mediterranean (Michez *et al.*, 2014) and North Sea-English Channel-Atlantic (Michez *et al.*, 2019). To ensure an appropriate identification of those habitats, we undertook the description of each habitat unit (biocenosis and associated sub-units) of the Mediterranean classification (La Rivière *et al.*, 2021). A similar work is in progress for the Atlantic classification.

The Habitats Directive (HD, 92/43/CEE) defines natural Habitat types of Community Interest (HCI) whose conservation requires the designation of special areas of conservation, but the definitions of the marine HCI are broad and do not represent appropriate management and conservations units for Natura 2000 site managers and

policy makers to work with. In order to clearly establish which habitat units should be considered as HCI, we specified national marine HCI definitions for the nine marine HCI present in national waters (de Bettignies *et al.*, 2021) and made crosswalks with the French national marine habitat classification.

All published information on French habitats will be available online through the HabRef repository (Gaudillat *et al.*, 2021). These tools based on the best scientific knowledge available allow to highlight knowledge gaps and will ease the assessment, management and reporting tasks both at local and national scales, and hopefully also at an international scale.

Materials and methods

The French national benthic habitats classification uses a hierarchical structure of 4 levels (biological depth zone-level 1, substrate type and/or salinity-level 2, biocenosis-level 3, facies/association-level 4). Habitat units were described at the levels 3 and 4.

The description of the national habitat units has been undertaken in two phases, first in 2016-2017 and then in 2020. After the creation of a template, the description for each habitat unit of the national Mediterranean classification was based on existing information (Bellan-Santini *et al.*, 1994; Bensettiti *et al.*, 2004; Pérès & Picard, 1964; PNUE *et al.*, 2006), updated or completed by French benthic scientists (Universities, CNRS, Ifremer, MNHN, etc.). The taxonomy has been updated in 2020.

The national reinterpretation of HCI types was undertaken in a second stage, involving Natura 2000 site managers and national benthic habitat experts. Existing approaches for interpreting HCI types from other EU member States were reviewed to serve as a basis (BFN, 2021; European Commission, 2007; 2013; JNCC, 2021; VV.AAAA., 2009).

Nine marine HCI types are present in the French waters (EU Codes 1110, 1120*, 1130, 1140, 1150*, 1160, 1170, 1180, 8330), two of those being considered as priority habitat types (*). All the HCI are defined by the European Commission in a unique document referred to as “EUR28” (European Commission, 2013). The reinterpretation work consisted in the description of each HCI type based on the EUR28 definition, and the identification of possible overlays between different types, following a defined template.

Results

All descriptions of the national classification habitat units were published in a common PDF catalogue (La Rivière *et al.*, 2021), as long as on each habitat webpage on the INPN (National Inventory of Natural Heritage) website through the HabRef repository (v6, Gaudillat *et al.* 2021) <https://inpn.mnhn.fr/programme/referentiel-habitats?lg=en>.

All level 3 and 4 habitats units of the French classification were described except for one association (level 4) due to the lack of knowledge and qualified available experts to provide a description. A total of 152 habitat units were published, with each index card providing pictures and information for each identification categories (abiotic factors, distinctive features, variability, characteristic species, associated species, risks of habitat confusion, natural dynamics and geographical distribution) when available (Fig. 1).



Fig. 1: Extract of the descriptive index card of the Association with *Cymodocea nodosa*, *Zostera noltei*, *Caulerpa prolifera* and *Caulerpa ollivieri* (III.2.3.a) within the Biocenosis of Mediterranean superficial muddy sands in sheltered waters (III.2.3) – from La Rivière *et al.*, 2021

When no information was available in one of the descriptive categories, whether due to a lack of knowledge or a lack of relevant information to provide, the section was not displayed. It should be noted that the descriptive content is not homogenous amongst all habitat units because of the heterogeneity of knowledge.

This work allowed to update the classification with taxonomical evolutions since its original publication in 2014, and the deletion of one unit for which no proof of existence could be found.

The national reinterpretation for each HCI type occurring in French waters was published in a common PDF catalogue (de Bettignies *et al.*, 2021), and will be available on each HCI type webpage on the INPN website through the HabRef repository by the end of 2022 (v7, in prep.). Each marine HCI type is endowed with an index card providing the following information: code and official name from EUR28, alternative French name (when the official French translation is not explicit), reminder of the EUR28 definition, definition of the habitat type according to the French interpretation and details (identification and distinctive criteria, possible overlays, national specificities, reference to other Member States definitions), references used and pictures illustrating the variability of the habitat type that can be encountered in French waters (Fig. 2-3). Unless stated otherwise, all identification criteria should be met in order to qualify a habitat as the considered HCI type.

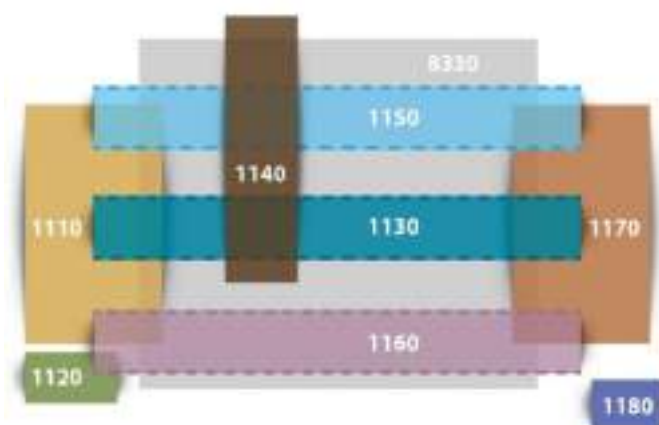


Fig. 2: Overlay possibilities between the 9 HCI types encountered in French national waters. Dotted lines indicate geomorphological types that can never overlap with each other – from de Bettignies *et al.*, 2021



Fig. 3: Extract of the French interpretation index card of the HCI type 1110 “Sandbanks which are slightly covered by sea water all the time”– from de Bettignies *et al.*, 2021

Discussion and/or conclusions

These benthic habitats definition projects provided a description for all habitat units of the French Mediterranean national classification and a clarification of the definition for each Habitats Directive’s HCI type based on a national interpretation. Level 3 (biocenosis) of the national classification is generally suitable for conservation and management issues, notably for mapping exercises. Level 4 (dominant lifeforms/communities) is more difficult to map at a large scale but can provide important details and knowledge of ecosystem functioning and environmental status and at a more local scale.

Following these two projects, we were able to establish 550 correspondences between the national classification and the HCI list in order to clearly identify which biocenosis should be managed within the framework of the HD and improve consistency in the management of Natura 2000 sites and reporting at the biogeographical scale.

Although only published in French for now, we hope this work can also be helpful to other Mediterranean countries that share similar benthic habitats to better identify habitats units in their waters. The interpretation of HCI types and the correspondences with the national classification could also improve the comparison of conservation status and/or management measures efficiency assessments across Member States by clarifying which biocenosis are encompassed into each HCI type.

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Alice OPRANDI, MANCINI I., BIANCHI C.N., MORRI C., AZZOLA A., MONTEFALCONE M.

SEL (Seascape Ecology Lab), DiSTAV (Department of Earth, Environment and Life Sciences), University of Genoa, C. so Europa 26, 16132, Genova, Italy

Email: alice.oprandi@edu.unige.it

INDICES FROM THE PAST: RELEVANCE IN THE STATUS ASSESSMENT OF *POSIDONIA OCEANICA* MEADOWS

Abstract

*Biotic indices are major tools recommended by European Directives for assessing the ecological status of marine coastal ecosystems. Although they have a relatively recent history, ecological indices have been widely used since their appearance in the 2000s under the requirements of the Water Framework Directive. The synergistic effect of anthropogenic pressures and global change in recent years caused an accelerated rate of change in marine ecosystems such as *Posidonia oceanica* meadows, which is detectable through the comparison with past data. Nevertheless, historical information is often scarce and fragmentary, then hardly to find. In this work, the availability of historical data on three *P. oceanica* meadows along the Ligurian coast (NW Mediterranean Sea) made it possible to evaluate the change over time in their environmental status. A number of indicators and ecological indices at different levels of ecological complexity, including the multimetric PREI (*Posidonia Rapid Easy Index*) were employed and compared through a novel approach named RESQUE (*RESilience and QUality of seagrass Ecosystem*). RESQUE is a graphical approach that allows obtaining a measure of the overall environmental quality and of consistency among indices. Indices resulted inconsistent when considered individually, leaving little scope for a proper interpretation of meadows' ecological status, whereas an effective measure of the ecosystem complexity was possible applying RESQUE. All the meadows showed an improvement over time. This study confirmed the need of using multiple descriptors for the status assessment of *P. oceanica* meadows and the potential of the RESQUE approach that is here applied for the first time to historical data.*

Key-words: Biotic indices, historical data, ecological quality, *Posidonia oceanica*, Mediterranean Sea

Introduction

Posidonia oceanica meadows represent one of the most threatened marine ecosystems of the Mediterranean Sea due to anthropogenic pressures. Coastal development has resulted in a generalized regression of seagrass beds over the last century that has been particularly severe in some areas of the basin, such as the Ligurian Sea (Burgos *et al.*, 2017). Although rare, first information regarding the status of seagrass beds in Liguria dates back to the late 1980s. But it was only with the Habitats Directive (92/43/CEE) that, starting in the 1990s, monitoring of *P. oceanica* meadows became a routine activity, allowing even more data to be collected.

Subsequent European directives, such as the Water Framework Directive, WFD (2000/60/EC) and the Marine Strategy Framework Directive MSFD (2008/56/EC) emphasized the importance of biological indicators to determine the ecological quality of coastal waters and advocated for the use of biotic indices to pursue the seafloor integrity (Borja *et al.*, 2000).

Seeing that *P. oceanica* is deemed an excellent indicator of water quality, several indices and descriptors have been developed basing on the features of the plant or on the structure of its meadows (Gobert *et al.*, 2009; Montefalcone, 2009). Despite the large availability of biotic indices, getting reliable information about status of seagrass can be a challenge because metrics are often not very comparable. Previous works where more indices were applied together displayed difficulties in assembling too differing responses and in drawing conclusions about the status of the habitat (Mancini *et al.*, 2020; Rigo *et al.*, 2020; Rigo *et al.*, 2021).

Although scarce and fragmentary, historical data are valuable because their comparison with the current conditions of the habitats allows change over time to be detected (Bianchi *et al.*, 2022). Of course, no historical data about *P. oceanica* biotic indices exist before 2000 but it is still possible to compute them when past information on seagrass meadows are available.

In this work, the availability of historical information on three *P. oceanica* meadows of Liguria, made it possible to calculate a set of 9 indices and descriptors on the past status of *P. oceanica*, and to evaluate habitat change over time by comparing them with current data through a novel graphical approach named RESQUE (RESilience and QUality of seagrass Ecosystem) (Oprandi *et al.*, 2021). RESQUE is able to incorporate all the different indices' responses allowing the integration of relevant meadow information into an overall expression of meadow health.

Material and methods

The investigated *P. oceanica* meadows were located along the coast of Liguria, an administrative Italian region facing the northernmost part of the western Mediterranean Sea. The three study sites were selected basing on the availability of historical information on *P. oceanica*, which specifically date back to 1992 for Bergeggi (Sandulli *et al.*, 1994), 1994 for Monterosso (Peirano *et al.*, 2001; Peirano *et al.*, 2011), and 2003 for Prelo (Lasagna *et al.*, 2006a, b). In 2017, following previous field methods, data were collected along depth transects (Bianchi *et al.*, 2004), laid between the lower and the upper limit of the meadows. Additionally at 15 m depth, shoots density was estimated within a sampling area of 40 cm × 40 cm (0.16 m²) in nine replicates and 18 *P. oceanica* shoots were sampled. Field and laboratory activities allowed to obtain a set of 9 indices and descriptors working at different level of ecological complexity and commonly used for the monitoring of *P. oceanica* meadows. Leaf surface LS (cm²·shoot⁻¹) was considered as a descriptor at individual level; shoot density SD (number of shoots·m⁻²), lower limit depth LLD (m), and the percent cover at the lower limit LLC (%) were considered as descriptors at population level; epiphyte biomass EB, expressed as the ratio between the mean epiphyte weight per shoot and the mean leaf surface per shoot (mg_{dw}·cm²) was considered as a descriptor at community level; Conservation Index CI, Substitution Index SI and Phase Shift Index PSI were considered as seascape indices (Montefalcone, 2009) and finally, PREI (Posidonia Rapid and Easy Index), due to its formula combining more metrics, was considered as a multilevel descriptor (Gobert *et al.*, 2009).

The 9 indices and descriptors were compared among themselves and over time through the RESQUE approach (Oprandi *et al.*, 2021). All data were normalized to get comparable values in the range between 0 and 1 and then graphically depicted on radar charts. According to the RESQUE approach, the total area inscribed in the resulting polygons was considered as a measure of the overall environmental quality, while circularity of the polygon perimeter was considered as an expression of consistency

between indices and as a measure of the meadow resilience. Relative Area Index (RAI) and Relative Circularity Index (RCI) were calculated by dividing the area and circularity of each meadow' polygon for those of a regular nonagon representing an ideal polygon whose scores were all equal to 1 (maximum quality and consistency combination).

Results

Indices showed very variable responses in the three meadows in both the time periods, as confirmed by the common edge profile of the polygons (Fig. 1) and by the general low values of circularity (Tab.1). Radar charts resulted from the plotted EQR scores revealed, however, an overall improvement over time in all the meadows according to the size and shape of the polygons (Fig. 1). The most irregular ones, corresponding to the lowest values of circularity were Bergeggi 2017 and Prelo 2003 (respectively, 0.500 and 0.517) (Tab. 1). Monterosso, whether past or current, had the maximum values of both, area (20554 in 1994, 28509 in 2017) and circularity (0.696 in 1994, 0.721 in 2017) (Tab. 1). Area of the polygons always increased through time by approximately 35 %. Similarly, circularity increased in Monterosso and Prelo by about 5% but showed a negative trend in Bergeggi with a slight decrease (6%). Finally, even when considered together, RAI and RCI, confirmed an improvement over time in the three meadows (Tab. 1), that was definitely more pronounced in Prelo (+22%) followed by Monterosso (+14%) and Bergeggi (+8%).

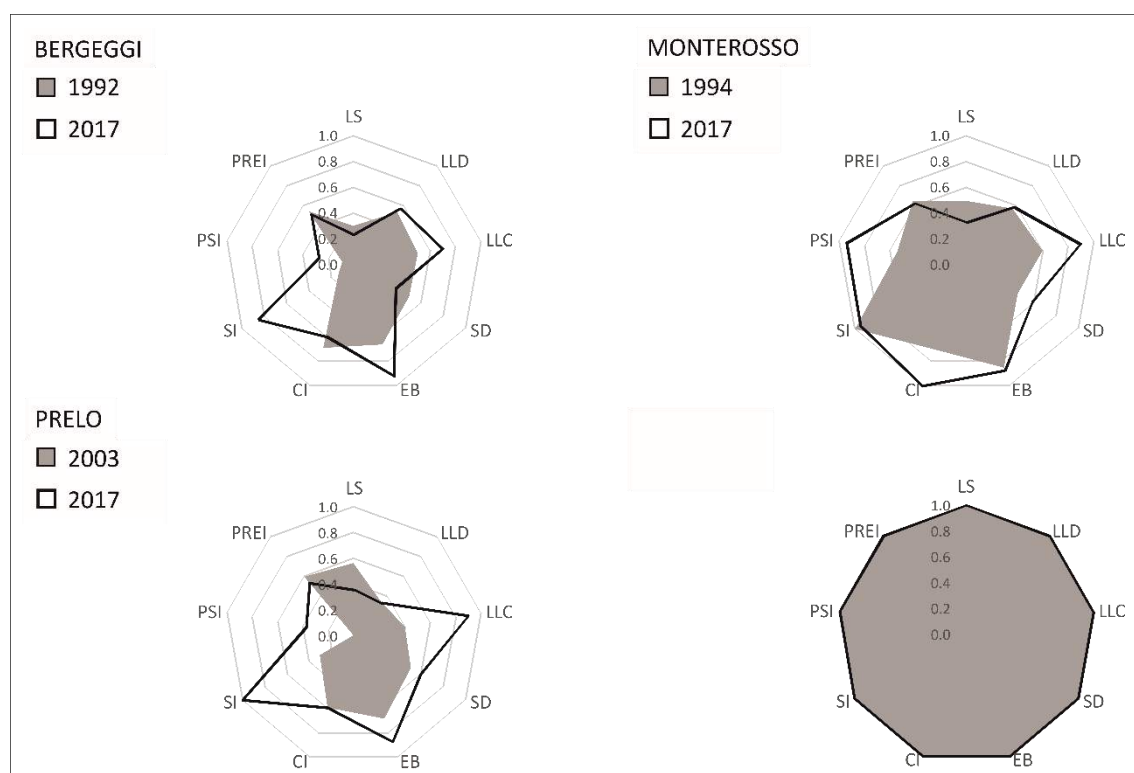


Fig. 1: Polygons resulting from the EQR scores of all the indices in the 3 meadows for the two time periods. An ideal nonagon is represented in the last diagram as a reference

Tab. 1 – Area and circularity of polygons and their corresponding indices. Reference values of area and circularity correspond to those of the regular nonagon whose 9 EQR scores are all equal to 1.

	AREA (pixels)	RELATIVE AREA INDEX (RAI)	CIRCULARITY	RELATIVE CIRCULARITY INDEX (RCI)	AVERAGE RAI - RCI
BERGEGGI 1992	9428	0.193	0.533	0.615	0.404
BERGEGGI 2017	14585	0.299	0.500	0.577	0.438
MONTEROSSO 1994	20554	0.421	0.696	0.804	0.612
MONTEROSSO 2017	28509	0.584	0.721	0.833	0.708
PRELO 2003	9650	0.198	0.517	0.597	0.397
PRELO 2017	18243	0.374	0.555	0.641	0.507
Reference Area: 48815			Reference Circularity: 0.866		

Discussion and Conclusions

Among the three *P. oceanica* meadows, Monterosso was found to be, on the whole, the best-preserved ones according to both historical and recent data. Its good conditions were consistent with previous studies (Peirano *et al.*, 2011), as it has been extensively investigated in the past years, being the largest *P. oceanica* meadow in the eastern Liguria included in the Cinque Terre Marine Protected Area (MPA). Additionally, Cinque Terre is the first Ligurian MPA, and since its establishment in 1997, *P. oceanica* has benefited from more than 20 years of protection regime that surely encouraged the improvement of Monterosso meadow. Bergeggi and Prelo, while similarly showed an improvement over time, had a significantly lower health status than Monterosso. Although the meadows suffered from various local impacts in recent decades, their reduced resilience, confirmed by the low circularity values, probably has the same origin. Indeed, both meadows in the past experienced substitution by the ruderal seagrass *Cymodocea nodosa* and/or by the invasive algal species of the genera *Caulerpa* that caused a moderate phase-shift in the ecosystem (Montefalcone *et al.*, 2007; Oprandi *et al.*, 2014). Regression of *P. oceanica* meadows in Liguria, indeed, started long before we bothered to measure it. Bergeggi MPA has a comparatively recent establishment than Cinque Terre MPA but the meadow has been heavily affected by the construction of a large artificial beach in the 1970s and by yearly beach replenishments (Montefalcone, 2009). Prelo is the only meadow that is not located within a MPA. Although it has been affected in the past by anchoring and by the construction of a jetty that altered the local hydrodynamics with negative consequences on the seagrass (Lasagna *et al.*, 2006b), to date, it seems to be the meadow that proportionally recovered more.

However, the overall recovery trend resulted in all sites is consistent with other evidences in the Mediterranean Sea (De los Santos *et al.*, 2019). If in Monterosso, the recovery is a natural consequence of the enforcement of conservation policies that in last decades made ceased or slowed down the decline of *P. oceanica* meadows, in Prelo and Bergeggi, the recovery is definitely linked to the, recently observed, reduction in the spatial distribution of substitute species.

Due to the complexity of seagrass habitat, the use of more indices and metrics has already been recognized as a pivotal method for the status assessment of *P. oceanica* meadows (Bianchi *et al.*, 2022). Nevertheless, differently from any other approach before, RESQUE provide an easy way to compare and interpret the different indices responses through a real portrayal of the habitat quality. The graphical approach confirmed the

potential of RESQUE, here applied for the first time to historical data, in returning a concise but comprehensive measure of ecosystem complexity, which is even more appreciated in the comparison over time.

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Christine PERGENT-MARTINI, ACUNTO S., ANDRÉ S., BARRALON E., CALVO S., CASTEJÓN-SILVO I., CULIOLI J.M., LEHMANN L., MOLENAAR H., MONNIER B., OBERTI P., PEY A., PIAZZI L., SANTONI M.C., TERRADOS J., TOMASELLO A., PERGENT G.

EqEL, UMR CNRS SPE 6134, Université de Corse, Campus Grimaldi, 20250 Corte, France.

E-mail: pmartini@univ-corse.fr

POSIDONIA OCEANICA RESTORATION, A RELEVANT STRATEGY AFTER BOAT ANCHORING DEGRADATION?

Abstract

*The anchoring of large pleasure boats constitutes one of the main threats in shallow marine habitats and particularly for seagrass beds. In the Mediterranean, this activity has seen constant development during the last decades, causing major physical disturbances in *Posidonia oceanica* meadows and associated ecosystem services, notably in terms of climate change mitigation (i.e. carbon fixation and sequestration capacities). In this context, the aims of the present study are to estimate the impact of these anchoring activities on *P. oceanica* meadows in a particularly highly-frequented area (Sant'Amanza gulf, SE Corsica Island) and to set up a strategy to restore this major carbon sink. Since the last decade, time-series of marine habitat maps revealed an important regression of *P. oceanica* meadows, with a loss of 72.9 ha, corresponding to 11% of the meadow surface and 9% decline in the total carbon fixation and sequestration performed each year. Moreover, in the most impacted part of the bay (Balistra bay), a loss of 16.6 ha (28%) has been recorded between 2011 and 2022. Following recent enforcement of anchoring regulation, prohibiting the anchoring of large units (greater than 24 m), and the lockdown linked to the COVID-19 pandemic, anchorages in the seagrass reduced by 92%, between 2018 and 2022. Natural recovery was observed at the edge of the meadow (plagiotropic rhizomes) but this growth is slow and the areas to recover are large. As a result, a transplant experiment, from cuttings harvested from the adjacent meadows, was initiated in the spring 2021. Four restoration techniques are being tested in the some pilot sites and the development of these transplants will be monitored and compared to the natural recovery.*

Key-words: Anchoring, seagrass, regression, transplantation, Corsica

Introduction

In the Mediterranean Sea, the endemic seagrass *Posidonia oceanica* (L.) Delile forms extensive meadows from the surface to more than 40 m depth. The *P. oceanica* meadows play a major role in coastal areas by providing ecosystem functions and services notably in climate change mitigation associated with the formation of an outstanding organic deposit, the matte. This belowground structure, composed of intertwined rhizomes, roots and leaf sheaths embedded in the sediment, exhibits a very low decay rate in relation with the highly refractory nature of the organic matter and the anoxic conditions. The accretion of organic material in coastal sediments beneath the *P. oceanica* meadows constitutes one of the largest carbon sinks in coastal areas worldwide and can reach several meters in height and remain over millennia (Monnier *et al.*, 2022).

Although this species is one of the main targets of conservation actions, the regression of *P. oceanica* meadows is well-documented over the whole Mediterranean basin. As for other seagrasses worldwide, mechanical impacts, particularly related to anchoring by large boats, are of increasing importance. Despite the relatively small size of the

Mediterranean Sea, more than half of the world fleet of large pleasure boats (>24 m in length) frequent Mediterranean waters for at least eight months per year (Carreño and Lloret, 2021), mainly in the Western Mediterranean basin. In Corsica, an increase of this activity has been recorded during the last decade. Out of nearly 2,000 of these large pleasure boats counted in 2018 around the coasts of Corsica, 44% of them anchored in the southern part of the island, in Bonifacio strait, and 77% of them on the *P. oceanica* meadows (Fontaine *et al.*, 2019). The aims of this study are to estimate the impact of this anchoring on *P. oceanica* meadows in a particularly highly-frequented area (Sant'Amanza gulf, Corsica Island) and to set up a strategy to restore this carbon sink.

Materials and Methods

The study site is located in the south-eastern part of Corsica in Sant'Amanza gulf (41.436°N, 9.246°E), an area included in the Specially Protected Area of Mediterranean Importance (SPAMI) Natural Reserve of the Strait of Bonifacio. The site covers a surface area of 1,466 ha and is notably frequented, during the summer season, by recreational boats due to its orientation, which makes it a sheltered site, especially for Balistra bay.

The information concerning the identification, movements and location of boats was based on Automatic Identification Systems (AIS), from 2010 to 2021. Eight fields of the dataset were used: Maritime Mobile Service Identity (MMSI), ship name, length, status (anchored = 1; moored = 5), speed, latitude, longitude, date, and time. Incorrect data (*e.g.*, anchored with moving speed) were removed according to Deter *et al.* (2017) and classification was made according to vessel length (20-40 m, 41-60 m, 61-80 m, and >80 m). The location of each boat was reported on a Geographical Information System (GIS; ArcGIS 10.8® software) to perform a spatial analysis. The anchoring pressure of recreational boats, in the gulf of Sant'Amanza and the bay of Balistra, was estimated from AIS data covering the period between 1st April and 30th September, corresponding to the main period of frequentation (more than 95% of the year) of the site.

The mapping of the gulf of Sant'Amanza was achieved between 0 and 50 m depth. The identification of the main marine benthic biocoenoses was based on the typology from Michez *et al.* (2011). The shallow area (<15 m depth; 627 ha) was mapped using a complete photographic coverage (BD ORTHO® 2016 – French National Geographic Institute (IGN)) with a 50 cm pixel resolution. The deeper area (>15 m depth, 839 ha) was mapped using exhaustive acoustic coverage acquired by side-scan sonar Klein 3000™ during the Carbonsink oceanographic survey (Ifremer) in August 2018. Ground-truth data (n = 560) were acquired using various tools according to depth (bathyscope, Remotely Operated Vehicle, underwater photo quadrat, scuba diving, Van Veen grab). These *in situ* observations were reported on ArcGIS 10.8®.

The mapping of the bay of Balistra was achieved using the BD ORTHO® 2011 (IGN, 50 cm pixel resolution), and four drone images carried out annually (12/06/2019; 16/02/2020; 24/02/2021, and 18/02/2022). These drone images, covering about 117.5 ha of the shallow waters (0-20 m depth), were acquired with a DJI Phantom 4 Pro 2.0 unmanned aerial vehicle (UAV) flying at an altitude of 30 m above sea level. The data were georeferenced with a GNSS RTK Spectra® SP 60 system (Leopont Instruments), offering centimeter level accuracy positioning, and compiled to obtain an orthomosaic with a 5 cm pixel resolution (Agisoft Metashape Professional 1.7 software). All the aerial imagery was processed using Envi 4.7® software following the method described by Bonacorsi *et al.* (2013) and integrated into ArcGIS 10.8® software. The estimation of the

surface areas occupied by the different marine benthic habitats was determined using the RGF 1993 – Lambert-1993 projection system (EPSG: 2154).

Carbon fixation and sequestration estimates were performed in 12 stations located along two transects oriented perpendicular to the coast at the central part (T1) and entrance part of the bay (T2). At the following depths (5 m, 10 m, 15 m, 20 m, 25 m, and 30 m), shoot density was measured using a 40 cm × 40 cm quadrat (5 replicates randomly distributed within the seagrass meadow) and orthotropic shoots ($n = 10$) were collected in June 2020. Phenological and a lepidochronological analyses were carried out. Net primary production was estimated using the lepidochronological cross method (Vela *et al.*, 2006) taking into consideration the biomass of foliar tissues produced during a one-year period. Rhizome production was evaluated from segments of the rhizome, with roots inserted, between two dead sheaths with a minimum thickness. The carbon content (% C) was determined for each sample using elemental analysis (Elementar Vario Micro Cube®). The global estimates of carbon fixation (blades, sheaths and rhizomes) and sequestration (sheaths and rhizomes) were achieved by fitting logarithmic regression curves using the mean carbon fixation and sequestration values, determined at each depth interval along the two transects. The corresponding equations were integrated into a morpho-bathymetric Digital Terrain Model in ArcGIS 10.8®.

In the spring of 2021, four *P. oceanica* transplant experiments were being carried out in Balistra bay, on a site occupied by dead matte (at 15 m depth) with anchoring prohibited. The cuttings used come from a seagrass bed located at an equivalent depth in the bay (Pergent *et al.*, 2021). In total, more than 3,600 cuttings, or nearly 11,500 shoots, were transplanted over an area of 900 m², using four different protocols (Molenaar and Meinesz, 1995; Calvo *et al.*, 2020, 2022; Castejón-Silvo and Terrados 2021; Piazzini *et al.*, 2021).

Results

The attendance of large pleasure boats (>20 m) in the bay of Balistra experienced a very significant increase in the early 2010s (Fig. 1). In total, 371 boats over 20 m were anchored in the *P. oceanica* meadow in this area, with boats between 40 m and 60 m length being the most numerous. However, over the past three years, their number has dropped significantly from 63 boats in 2018 to 5 boats in 2021. The peak in attendance is also increasingly late with a peak in the first week of August in 2019 and in the third week of August in 2020 and 2021 (data not shown).

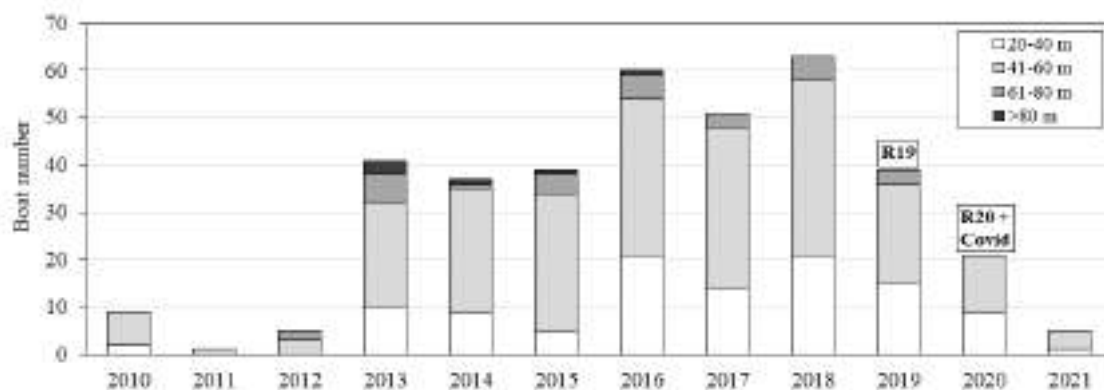


Fig. 1: Number and length of boats anchoring in the *P. oceanica* meadow in the bay of Balistra. R19: Decree N°123/2019; R20: Decree N°206/2020; Covid: COVID-19 pandemic.

The cartography of biocoenoses of the gulf of Sant'Amanza reveals a *P. oceanica* meadow that develops preferentially between 5 m and 30 m depth, that is the 45% of the surface area of the bay (649.7 ha), but also large areas of dead matte in the central area of the bay of Balistra, associated with anchoring (72.9 ha, 11%). The time series map of Balistra bay, between 2011 and 2022, shows an important regression of the seagrass meadows, which reached 16.6 ha, *i.e.*, nearly -28% of the original surface covered by *P. oceanica* (Fig. 2). This regression is significant (Mann-Kendall trend test) between 2011 and 2020 (-15.9 ha; -1.8 ha yr⁻¹) and then stabilizes between 2020 and 2022 (-0.7 ha; -0.3 ha yr⁻¹). At the same time, the surfaces of dead matte increased by 13.7 ha and the soft bottom (*e.g.*, well calibrated fine sand covering some dead matte areas) by 3.4 ha.

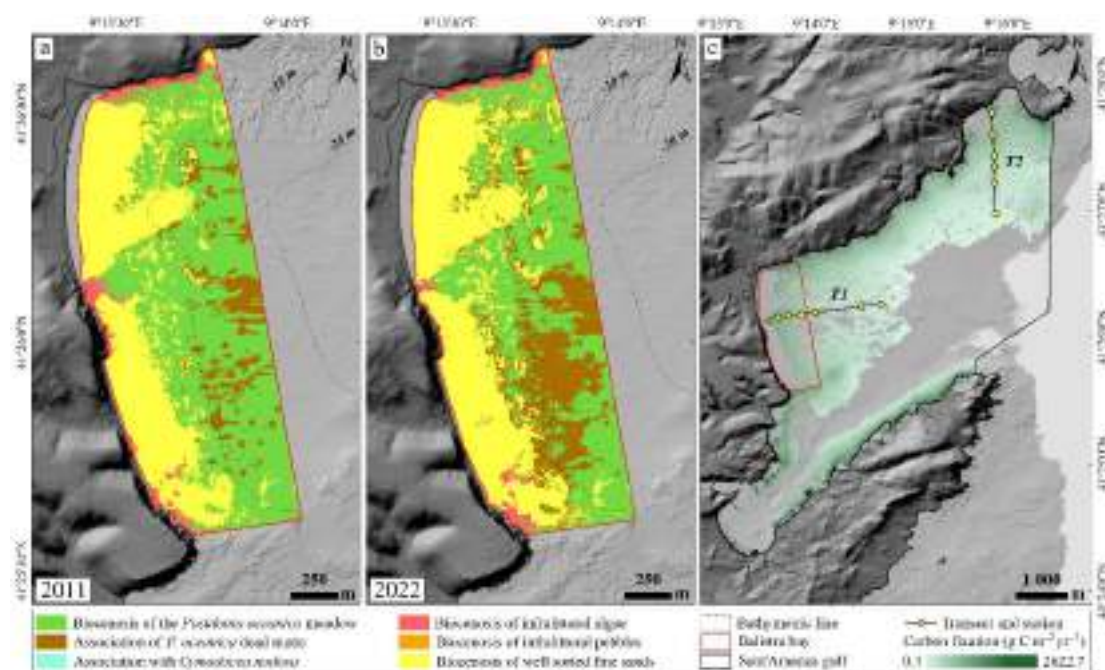


Fig. 2: Distribution of main biocoenoses and associations in 2011 (a), 2022 (b) in Balistra bay and carbon fixation in the gulf of Sant'Amanza (c).

Carbon fixation by the leaves of *P. oceanica* decreases with depth and varies between 1,035.2 mg C shoot⁻¹ yr⁻¹ (T2, 5 m) and 382.9 mg C shoot⁻¹ yr⁻¹ (T1, 30 m). The mean density of *P. oceanica* meadows classically decreases with depth and it is, on average (\pm S.E.), always lower along T1 (351.0 ± 194.4 shoots m⁻²) than along T2 (422.5 ± 186.4 shoots m⁻²). The mean annual carbon fixation by the meadow varies between 726.2 g C m⁻² (5 m) and 95.4 g C m⁻² (30 m), reaching in average (\pm S.E.) 325.8 ± 95.8 g C m⁻². The annual carbon sequestration in the matte varies between 202.3 g C m⁻² (5 m) and 16.4 g C m⁻² (30 m) and is on average (\pm S.E.) 84.4 ± 29.1 g C m⁻² (23.6% of total carbon fixation). The integration of the carbon fixation and sequestration equations over the entire bathymetric range of the *P. oceanica* meadow at the study site provides a basis for estimation of the total fixation rate at $1,531.4 \pm 349.3$ t C yr⁻¹ for the entire gulf in 2021 ($5,620.2 \pm 1,281.9$ t CO_{2e}; Fig. 2). The quantity of carbon sequestered reaches 373.2 ± 104.1 t C yr⁻¹ ($1,369.6 \pm 382.2$ t CO_{2e}).

The monitoring of the transplants, after one year, shows a survival rate of the cuttings that varies between 86% and 100% (Tab. 1). The greatest increase in shoot density (+22.3%)

is observed at the level of protocol 1, only protocol 3 experiences a regression in the density of the shoots (-9.4%).

Tab. 1. Evolution of cutting survival and shoot density, between May 2021 and May 2022.

Cutting survival	Protocol 1	Protocol 2	Protocol 3	Protocol 4
- July 2021	89%	100%	90%	81%
- May 2022	89%	100%	87%	70%
Shoot density				
- July 2021	79%	99%	122%	91%
- May 2022	97%	108%	111%	97%

Discussion

After an increase in the attendance of large recreational boats in the bay of Balistra, from 2010 to 2018, the number of boats declined from 2019 and started later in the summer season in 2020 and 2021 (Fig. 1). This pattern could be explained (i) by the recent regulations, relative to the anchoring of the largest boats (>24 m), in force since June 2019 (e.g. French Naval Prefecture, Decrees N°123/2019 and N°206/2020), and (ii) the COVID-19 pandemic and the corresponding spring lockdowns, as observed in both the Mediterranean basin and worldwide. Anchoring regulations appear to be mostly respected and constitute an effective measure to limit the impact of large recreational boating on Sant'Amanza *P. oceanica* meadow. However, such regulations are still scarce at the scale of the Mediterranean with the exception of France, Croatia and Spain – Balearic Islands (e.g., Official Bulletin of the Balearic Islands, Decree N° 25/2018, 28 July 2018).

As regards more specifically the loss of the carbon sink provided by *P. oceanica* meadows, the consequences can be subdivided into two main categories: (i) a reduction in living meadow areas, which directly leads to a loss of carbon fixation and sequestration capacity, estimated respectively at 141.3 ± 37.1 t C and 33.0 ± 9.8 t C, or 9% of the total fixation and sequestration of the meadows from the gulf of Sant'Amanza; and (ii) a destruction of the mat, under the combined action of anchors and chains, which results in a remineralization of carbon accumulated for thousands of years in the sediments of seagrass meadows (Monnier *et al.*, 2022). In addition, the measurements carried out within the anchor tracks, showed a furrowing of the mat of 20 to 30 cm, over a width of 1 to 2 m, which is a volume of lost mat estimated on average at 0.37 m³ per linear meter. Considering the average amount of carbon stored in the first 30 centimeters of *P. oceanica* mat (107 ± 5 t C ha⁻¹; Monnier *et al.*, 2022) and the surface areas destroyed within the bay (72.9 ha), boat anchoring would be responsible for the loss of more than $7,800 \pm 365$ t C ($28,627 \pm 1,338$ t CO_{2e}), i.e. all of the carbon sequestered by the *P. oceanica* meadow of the gulf of Sant'Amanza over the past 20 years.

Even if the protection of the *P. oceanica* meadow in the gulf of Sant'Amanza is likely to allow the natural recolonization of the seabed covered today by dead mat (measurements are in progress), the low growth rate of the plant (a few centimeters per year) and the large areas of dead mat to colonize (several tens of hectares) will require a colonization period of several decades to several centuries (Pergent-Martini *et al.*, 1995). So, the use of transplantation of cuttings of *P. oceanica* could be considered to restore this habitat and the associated ecosystem services more quickly. The preliminary results from the experiments set up in 2021 in the bay of Balistra are very encouraging

with very high survival rates (>95% in average) and mostly a multiplication of the shoots in three of the four protocols. Of course, a regular monitoring over at least three years is essential to assess the success and the interest of this approach and above all to compare it to the natural colonization. Finally, full compliance with the decree is expected, to achieve zero boat anchoring in the *P. oceanica* meadow in this protected area.

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Dimitris POURSANIDIS, IOSSFIDIS S., VASSILOPOULOU V.

terraSolutions Marine Environment Research, Archimidous 63 Heraklion, Crete, Greece

E-mail: dpoursanidis@gmail.com

DETERIORATION OF PRECIOUS BLUE CARBON ECOSYSTEMS IN A NATURA 2000 SITE: THE IMPACT OF ANCHORAGE IN THE SEAGRASS MEADOWS OF THE NATIONAL MARINE PARK OF ALONISSOS NORTHERN SPORADES

Abstract

Climate, oceanographic and seascape factors define where, how and to what extent, seagrass meadows in the Mediterranean Sea grow and thrive. The same characteristics define where touristic boats find shelter during windy and stormy weather conditions all year around when they sail in the Aegean Sea. There, islands, and islets form complex archipelago, where human and nature coexist for millennia. Among them, Sporades Archipelagos is a unique system, where the National Marine Park of Alonnisos Northern Sporades has been established in 1992. There, extended seagrass meadows cover almost all sites with low wind effects and developing on soft sediment bottoms up to 35 m depth, while in deep waters, where the dim light reach, elusive coralligenous formations with gorgonian forests thrive. During summer period, the Park is among the most famous destinations for sailing and short-term holidays using boats and sailings. In absence of eco mooring systems, the impacts of the anchorage to the seagrass meadows are expected to be concerning; however, until now no information was available. During the period 2018-2021, under the project INTERREG MED AMAR and a funded initiative by the Management Body, a non-destructive protocol has been used by means of scientific scuba diving to collect data for the characterization of the ecological status of the meadows in selected sites of the two zones of the Marine Park, based on the pressure exerted by vessels. In almost all sites, at depths shallower than 15 m, the meadows showed a bad ecological status with destroyed parts and invasion by alien algae, while in the deep portions the situation was better than in shallow waters, showing clear impacts of the anchorage to the critical blue carbon ecosystem. The need for monitoring is imperative in order to catch upon early signs of habitat degradation and plan mitigation measures.

Key-words: anchorage, ecosystem services, human activity, scientific diving, seagrass.

Introduction

One of the most effective means of protecting marine and coastal biodiversity is through the establishment and effective management of marine protected areas (MPAs). According to IUCN's definition, a protected area "is a clearly defined geographical space, recognized, dedicated and managed, through legal or other effective means, to achieve the long-term conservation of nature with associated ecosystem services and cultural values". In Greece, two areas have been declared as MPAs, the Zakynthos Marine Park putting conservation emphasis on the loggerhead turtle *Caretta caretta*, and the National Marine Park of Alonnisos Northern Sporades (NMPANS) established for the preservation of the monk seal *Monachus monachus*. The latter is the largest coastal marine park in the Mediterranean and has been declared as a marine park under a Presidential Degree in 1992, while the final declaration was endorsed by the official paper of the government 621/19.6.203. The NMPANS has been selected as the Greek case study

of the INTERREG MED project AMAre, followed by a back-to-back project from the Management Body of the Marine Park in 2021 with the aim to provide concrete scientific results, useful towards drafting an effective management plan for the MPA. Emphasis has been given to the priority habitat 1120* (*Posidonia oceanica*) as it is the dominant seagrass species in the Marine Park with a wide distribution in the area and early signs of pressure have been already identified from the Management Body. The aim of this work is to provide insights on the spatial distribution of the seagrass ecosystem status using non-destructive methods that are widely used in the Mediterranean Sea, which can be handled over by non-experts as citizen scientists to provide robust results and quantitative and replicable data on the ecosystem status of the precious seagrass meadows (Boudouresque et al. 2012; Vassallo et al. 2013). The results are intended to be used for local scale maritime spatial planning and future steps towards the installation of eco mooring systems. All in all, the work sets the baseline for the current status during the period 2018-2021 aiming at the continuous monitoring of the priority habitat and setting a policy-society discussion panel on the needs for the conservation of seagrass meadows.

Materials and Methods

Field work took place during the summer of 2021, in June and July (postponed due to the pandemic, as it was initially planned for 2020). Based on the previous expeditions during the AMAre project (Poursanidis and Vassilopoulou, 2019), field work has been designed in order to survey old stations and to also include new ones in sites that have not been investigated in the previous work due to logistics and weather constrains. At each station at 10, 15, and 20 m depth, the following data have been collected: a) shoot density using a frame of 20 cm × 20 cm in five replicates per depth; b) 25 m long transects, using a topographic tape, for collecting data for the estimation of the Conservation Index (Moreno et al., 2001) only at 10 m depth in three replicates; c) lower limit depth and typology; d) occurrence of invasive alien species; e) occurrence of marine litter; and f) signs of human destruction such as anchorage by boats or other bottom contact that caused significant and severe impacts in seagrass meadows. Data interpretation follows the recommendation by UNEP/MAP on the Common Indicators 1 & 2 for the Ecological Objective 1 (EO1) for the *Posidonia oceanica* meadows (UNEP/MAP, 2019). The selection of these metrics was based on the non-destructive nature of them, the potential use by non-experts after minimum training and the absence of wet laboratory requirements for the drawing of insights. The developed indicators that link the seagrass ecological status with the EQR scale would provide additional value, but these analyses require more sampling effort, logistics and fundings. When availability of time and laboratory infrastructures are limited, the metrics here adopted can anyway provide fast and efficient insights on seagrass status for each sampling site.

Results

Fourteen stations have been visited during the latest campaign in 2021. Three of them are in the Zone A and eleven in the Zone B. The unbalanced distribution among the two zones of the MPA is due to the distance from the main harbor (only one in the south part of the Zone B), the operational capacity of the available inflatable boat and the weather conditions during fieldworks. Almost all the stations showed a good to excellent status in the deep portions of the meadows (at 20 m depth) and gradually changed to moderate and bad in the shallower portions (15 m and 10 m depth) (Fig. 1; Tab. 1).

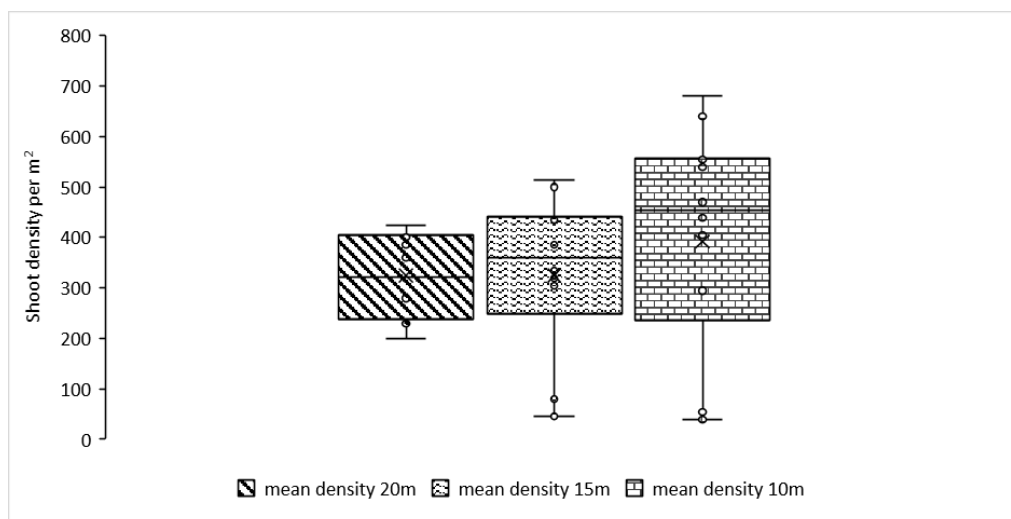


Fig. 1: Boxplots of the mean (\pm sd) shoot density per depth in all the stations.

The Conservation Index showed a moderate variability among depths and stations (Fig. 2). The majority of the stations showed signs of anchorage, dead noble shells, invasive alien macroalgae and seagrass along with marine litter. The invasive seagrass *Halophila stipulacea* was present in situations where *P. oceanica* meadow has been dredged by the repeated anchorage and the integrity has been lost. The green alien macroalgae *Caulerpa cylindracea* was also present mostly in sites that were protected by winds. Marine litter was present mostly in shallow waters of all stations, with origins either from the oceanographic circulation or the tourism activities from yachting and boating.

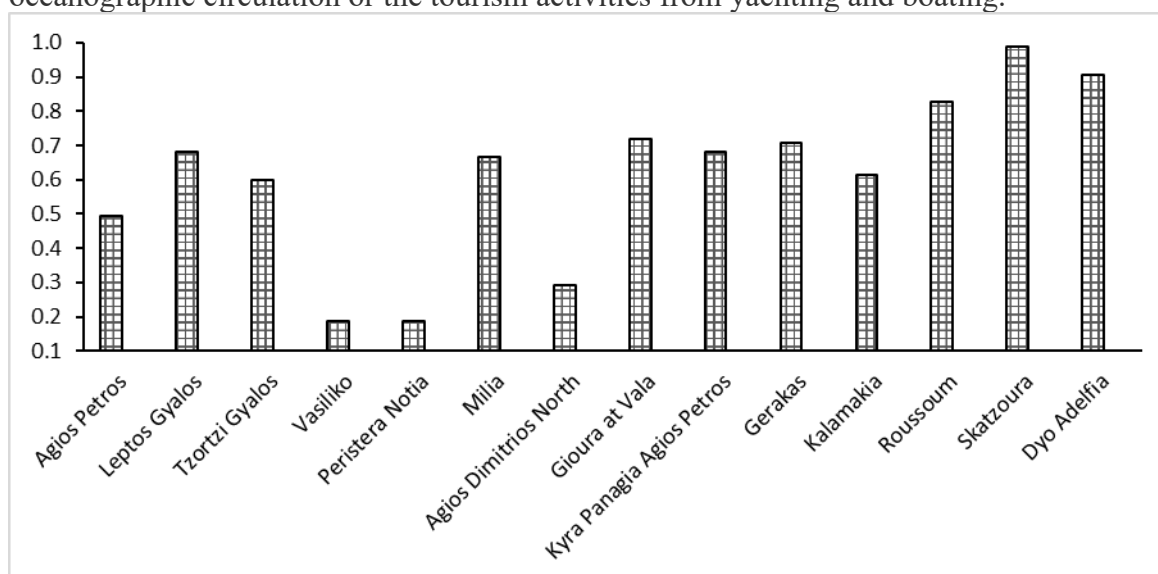


Fig. 2: Conservation Index per station.

The noble shell (*Pinna nobilis*) was dead in all sites of the Marine Park. The first signs of its decline have been recorded in 2019 when the mortality has been spread in the Aegean Sea (Zotou et al., 2020). The shallow portions of the majority of the stations have scars of anchorage or even anchored boats. These are of variable size and extent, while in some locations, deep trenches from repeated anchorage and large yachts have caused severe dredges and destructions. In few stations, the meadow until 15 m depth has been almost disappeared and the sediments have been pulverized from the heavy repeated anchorage.

Tab. 1: Shoot density, lower limit depth and typology, and pressures/threats at each depth of the study stations. With *italic* and underlined are the stations with a bad status, in **bold** are with poor status, in *italic* are that with a moderate status, with **bold and underlined** are of good and excellent status.

Station	mean shoot density per m ²					lower limit depth	lower limit typology	pressures/threats
	20m	15m	10m	31	32			
Agios Petros	<u>400</u>	305	470	31		Natural Shaded	Dead noble shells, marine litter, signs of anchorage	
Leftos Gyalos	<u>410</u>	<u>500</u>	680	31		Natural Shaded	Dead noble shells, marine litter, signs of anchorage, <i>Halophila stipulacea</i>	
Tzortzi Gyalos	<u>425</u>	335	295	32		Natural Shaded	Dead noble shells, signs of anchorage, marine litter	
Vasiliko	200	<u>45</u>	<u>40</u>	31		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage, destroyed meadow, marine litter	
Peristera South	235	<u>45</u>	<u>40</u>	31		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage, destroyed meadow, marine litter	
Milia	<u>405</u>	390	440	33		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage	
Agios Dimitrios North	<u>230</u>	<u>80</u>	<u>55</u>	31		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage, destroyed meadow, marine litter	
Vala at Gioura island	<u>280</u>	305	555	32		Natural Shaded	Signs of anchorage	
Agios Petros at Kyra Panagia Island	<u>280</u>	445	640	33		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage, destroyed meadow, marine litter	
Gerakas	<u>360</u>	385	480	32		Natural Shaded	Dead noble shells, signs of anchorage	
Kalamakia	<u>240</u>	320	295	31		Natural Shaded	Dead noble shells, <i>Halophila stipulacea</i> , <i>Caulerpa cylindracea</i> , signs of anchorage	
Roussoum	<u>285</u>	<u>515</u>	405	33		Natural Shaded	Dead noble shells, signs of anchorage	
Skatzoura	<u>405</u>	<u>440</u>	560	32		Natural Shaded	No pressures	
Dyo Adelfia	<u>385</u>	<u>435</u>	540	33		Natural Shaded	No pressures	

This situation already appeared from the expedition of 2018, during the Interreg MED AMAre project and initiated the needs for a thorough investigation of the conditions, using scientific diving and protocols but also Earth Observation data, which allowed us to count boats at the study locations using daily Planet SuperDove data at 3 m pixel size.

Discussion

In view of the NMPAS, from the perspective of a visitor looking at the map, the largest coastal marine protected area of the European waters appears as an attractive destination, far away from the anthropocentric crowded islands. There are expectations of “pristine” seas and elusive habitats occur, allowing a dream of a seascape full of life. However, when zooming into the coves and bays and counting the amount of the boats that everyday cross over the marine area, the enemies of Poseidon appear and show they dark faces. Repeated anchorage for years, without any regulation or rules, have the power to destroy the shallow portions of seagrass meadows, a habitat of priority importance for the European Union that quite recently has taken the place in the position of the blue carbon as among the most important habitats to compensate for carbon offsets. However, given the slow growth of this plant, any anchorage activity that destroys an area of 20 m² each time will take at least some decades to recover the seagrass appearance, also including the banning of anchorage and restoration activities. No robust information is available up to now on the time needed to recover again the stored carbon in the rhizomes and the matte. By that, there is an urgent need for harmonized action among the Mediterranean countries, to halt this behavior and promote the sustainable use of permanent mooring systems in any cove and bay of our sea.

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Francesca ROSSI, ROMERO G., PEY A., VENTURA P., LOQUES F., DERLIARD B., COTTALORDA J.M., DI FRANCO E., GUIDETTI P., BUSSOTTI S.

Université Côte d'Azur, CNRS, UMR 7035 ECOSEAS, Nice, France

E-mail: francesca.rossi@cnrs.fr

THE TROPHIC ROLE OF THE NECROMASS OF *POSIDONIA OCEANICA* (L) DELILE IN THE SURF ZONE OF SANDY SHORES

Abstract

*In the Mediterranean, the detritus (necromass) of the endemic seagrass *Posidonia oceanica* (L) Delile accumulates on the seafloor of sheltered and embayed sandy shores, where it forms organically-enriched patches interspersed within an unvegetated bare sandy matrix. This habitat is often considered important as food and refuge for both invertebrates and fishes, yet it is little studied. We present here results from an ongoing study where we have sampled patches of seagrass detritus and sandy substrate across 6 beaches with a different degree of urbanisation in the area of Cote d'Azur, North-Western Mediterranean, France. We compared assemblages of fishes (with in situ non-destructive visual censuses) associated with bare sand to those found in *P. oceanica* wrack beds and we examined the trophic relationships between detritus, invertebrates and two dominant invertivorous fishes, through stomach content (fish) and stable isotopes (not shown). We have found, so far, more fishes on the detrital patches. Stomach contents of these fish taxa were often composed of invertebrates found in the detritus. Stable isotope samples are in progress. We will discuss the ecological role of this habitat derived from the natural cycle of the iconic Mediterranean seagrass *P. oceanica* and the importance of extending conservation initiatives to its necromass.*

Key-words: macrophyte, wrack, sandy shores, UVC, Mediterranean Sea

Introduction

Seagrass primary production often exceeds herbivore consumption and the often large amount of seagrass detritus contribute to carbon burial and to foodweb subsidies locally or in other ecosystems, where detritus is drifted by water current (Mateo & Romero, 1997; Polis *et al.*, 1997; Heck *et al.*, 2008). On Mediterranean sheltered and embayed sandy shores, the detritus of the iconic and endemic seagrass *Posidonia oceanica* (L) Delile derived from local meadows may accumulate on both the supralittoral and the surf zone (Colombini & Chelazzi, 2003). On the supralittoral beach, they form berm-like structures, sometimes called banquettes, whereas in the surf zone they form patches of an organically-enriched brown bed interspersed within an unvegetated bare sandy matrix (Vacchi *et al.*, 2017). These accumulations are recognised to be important for coastal carbon cycling, for habitat and food provision to many marine and terrestrial invertebrates (Gallmaetzer *et al.*, 2005; Costa *et al.*, 2019) and for limiting beach erosion (Vacchi *et al.*, 2017). Moreover, it is generally recognised that fish species might use them as a habitat providing food and refuge (Personnic *et al.*, 2014; Boudouresque *et al.*, 2016), as for other seagrass detritus (Baring *et al.*, 2018). But scientific evidence for *P. oceanica* wrack beds is limited and, only recently, Bussotti *et al.* (2022) observed in the surf zone of Sardinian waters more omnivore and invertivore fishes on *Posidonia* patches than on bare sediment, often acting like feeding on the detritus. The study however did not investigate invertebrate distribution and fish diet. In this study, we investigate

invertebrate and fish assemblage distribution and the diet of most abundant fishes across sandy embayed beaches of the Côte d'Azur. We also plan to use stable isotopes to investigate fish and invertebrate diet, but these results will not be presented here. Here, in particular, we expect more fishes associated with the *Posidonia* detritus than the sand and that the invertebrate assemblage associated with detritus resembles that found in the fish gut content.

Materials and methods

During June - September 2021 we sampled 6 sandy shores (hereafter Sites), along the Côte d'Azur, between Cap d'Antibes (43° 32' 59" N, 7° 07' 48" E) and Cap Ferrat (43° 41' 0" N, 7° 20' 0" E) (Western Mediterranean Sea, France). The sites were all pocket beaches, embedded around artificial groynes or natural rocky shores, where seagrass accumulation on the emerging beach was removed during spring-summer. They had, however, a different level of artificialization, being the sites of Galice and Passable the most artificial and the sites of Paloma and Ondes the least ones (Fig. 1).



Fig. 1: Study sites

At each site, we selected 3 similar *Posidonia* detritus patches and 3 sandy patches, by snorkeling. Fish assemblages were quantitatively assessed 3 times by means of the “stationary point counts” of 3 m radius (Bohnsack & Bannerot, 1986). Abundance, size and behavior (swimming/feeding and hiding/camouflage behaviors) of fishes was estimated. Here we only analyze fish density at the second sampling campaign, when we captured the 2 most abundant and widespread invertivores fish species (up to 5 fishes per site) using a fishing harpoon in and sampled for seagrass detritus using a 20 cm diameter fishnet. We collected 3 replicate fishnets at each of 3 patches per site. In the laboratory, we analysed fish stomach content, sorted and identified invertebrate species for both the fish stomach and the detritus. We also prepared all the samples for stable isotope analyses, which are still in progress and they will not be presented here.

We assessed the distribution patterns of both invertebrate and fish assemblages by means of generalized linear models for the multivariate density data with the function ‘manyglm’ from the mvabund R package (Wang *et al.*, 2012). As predictor variables, we considered “Habitat” (2 levels: detritus and sand) and “Site” (6 levels) as crossed factors with 3 replicated transects for fish assemblages; “Site” and “patches” (nested in Site for the invertebrates found in the detritus and “patches” for the gut content for each of the 2 fish

species). Non-metric multidimensional scaling (nMDS, Bray–Curtis distances) was used to visualize the results of the multivariate generalized linear models (r package vegan).

Results

We counted 18 fish taxa across sites and habitats (sand vs detritus) and there were differences in the interaction Habitat x Site in the density distribution of fishes (Deviance_{5,23} = 64.17, $p = 0.01$). The distribution of fishes differed between habitats at all sites except one (Galice beach). The differences among habitats were mainly due to the fact that more species and individuals were more associated with the detritus than the sand (Fig. 2). *Mullus surmuletus* and *Symphodus tinca*, together with *Diplodus sargus* were among the most abundant (20, 9 and 24% of the total density, respectively) and most spread species across sites (42, 40 and 69% of total transects). We examined the two invertivorous species for stomach content.

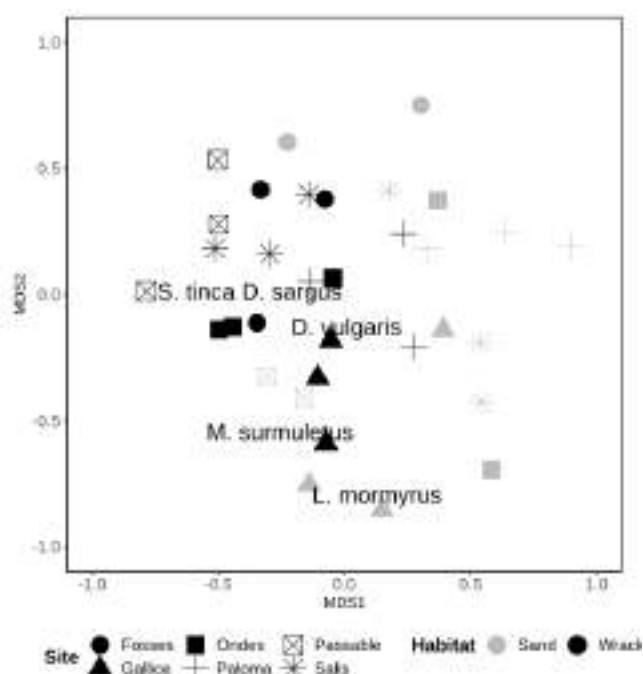


Fig. 2: nMDS for fish density. Points indicate the sites and the type of patches, the names are the most important fishes.

There were differences in prey items abundance and taxa among species and sites (Deviance_{5,27} = 30.64, $p = 0.046$). Both species showed similar prey items, except in the Salis beach (Deviance_{1,7} = 11.08, $p = 0.05$) and there were differences among sites only for *M. surmuletus* (Deviance_{5,9} = 67.2 $p = 0.01$). Some detritus leaves were also collected in the gut, especially for *S. tinca* (90% of specimens captured). Overall, Gammaridae (especially *Gammarus* sp) and Isopoda (especially *Idotea* sp.) characterised the gut of both species, especially for 3 out of the 6 sites, whereas Ophiuroidea and Polychaeta were found in fishes fished in the Galice beach. Gastropods (*Bittium* sp) characterised more often the gut of the fishes collected in the Passable and Fosses Beaches (Fig. 3a). We collected 47 species and 2338 individuals of macrofauna invertebrates in the detritus patches. There was a lot of variation among patches, but there were clear differences among sites in the invertebrate distribution associated with the presence of detritus of *Posidonia* (Deviance_{5,41} = 324.0 $p = 0.01$, Fig. 3b). The differences among sites were

mainly due to the abundance of the bivalve *Loripes lacteus* at the Galice site and to the abundance of Gammaridae (*Gammarus* sp and *Gammarella fucicola*) and Isopoda (*Idotea balthica*) at the 3 sites of Ondes, Paloma and Salis. The gasteropoda *Bittium* sp and *Jujubinus* sp characterized some of the patches at the remaining sites (Fig 3b).

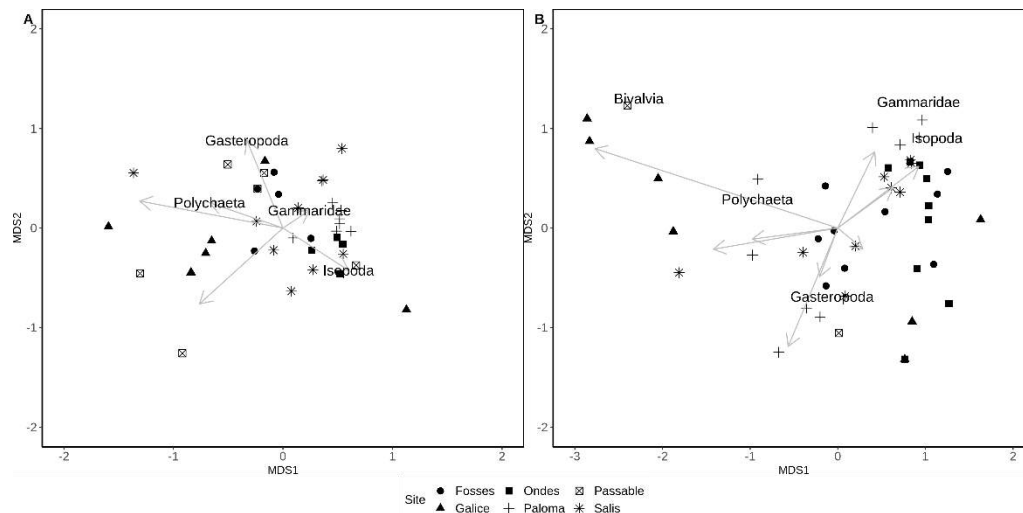


Fig. 3: nMDS (Bray-Curtis dissimilarity index after log transformation) for (a) the gut content of the two fish species across sites and (b) the invertebrate assemblage associated with the seagrass detritus. Analyses for the invertebrate assemblage were done with the matrix of species. Here we indicate high level taxa for analogy with the gut content

Discussion and conclusions

Fish associations to macrophyte detritus have been little examined (Baring *et al.*, 2018), especially for the Mediterranean Sea (Bussotti *et al.*, 2022), where *P. oceanica* detritus is recognized as an important habitat (Boudouresque *et al.*, 2016; Vacchi *et al.*, 2017). The surf zone, in particular, represents an important habitat to forage or hide in for coastal fishes during a part of their life cycle (Esposito *et al.*, 2015). In this study, most species, which included the numerically dominant *Diplodus sargus*, *Symphodus tinca*, and *Mullus surmuletus*, were more abundant on the detritus. This result is consistent with data reported by Bussotti *et al.* (2022) suggesting as a general pattern that detritus is an important habitat component for these species in the surf zone. These taxa are well known to mostly inhabit rocky reefs and *P. oceanica* meadows (e.g. Guidetti, 2000) and they might move between adjacent rocky reefs and *P. oceanica* meadows using detritus patches as shelter, as observed by Barings *et al.* (2018). Bussotti *et al.* (2022) observed that along Sardinian coasts several fishes showed more frequent feeding acts on detritus than on sand, including the invertivorous fishes *M. surmuletus*, *S. tinca*, omnivorous sparids (ex *Diplodus* spp.) and the piscivorous *Serranus scriba*. We observed a large number of invertebrate species, especially crustaceans, inhabiting the *Posidonia* detritus. Even if similar studies are lacking for the Mediterranean Sea, some papers report significant abundances of prey for invertivorous fishes (e.g., crustaceans, mollusks, meiofauna species) within the detritus accumulating within *P. oceanica* meadows or within bottom depressions, between stones or boulders (Gallmetzer *et al.*, 2005; Remy *et al.*, 2018, Costa *et al.*, 2019). The same invertebrates found within the detritus were found in the stomachs of the fishes studied. Studies conducted in southwestern Australia reported particularly high densities of crustacean amphipods in wrack accumulations and in the stomachs of fish feeding on them (Baring *et al.*, 2018).

Vanderklift and Jacoby (2003), moreover, reported a significant relationship between species number, abundance or biomass of invertivorous fishes and the amount of drifting plant material in sandy substrates adjacent to seagrass beds. In addition, in our study some detritus was found in the stomach of both fish species, further indicating that detritus and invertebrates living in there constitute feeding items. Very likely, the detritus supported invertebrate assemblages and provided invertebrate preys to invertivorous fishes. In turn, this might increase the availability of small fishes for piscivorous predators. Further evidence on the feeding role of the detritus for fishes will be provided using stable isotopes, which may help estimating the carbon contribution of detritus to fish diet, especially for commercially interesting fishes. In conclusion, this study highlights the importance of detritus as a foraging area for even commercially important fishes. The current decline of several vegetated habitats could thus have negative consequences far beyond the areas where seagrasses grow and conservation efforts should consider seagrass detritus. We should also notice that the wrack in the surf zone does not come only directly from the *Posidonia* meadows, but also from the accumulations on the supralittoral beach (Vacchi *et al.*, 2017). In touristic regions like the Mediterranean, beach wracks are often perceived as unpleasant and as an ecosystem 'disservice', driving beach grooming and sand nourishment procedures for their removal (Ruiz Frau *et al.*, 2020). Improving ecological knowledge on the role of this detritus and demonstrating its role as feeding ground for fishes of commercial interest should contribute to making stakeholders more aware that this practice is a threat for the shore and for biodiversity.

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**Andrea TURSI, MASTROTOTARO F., BOTTALICO A., DE GIOSA F.,
MONTESANTO F., LISCO A., CHIMIANTI G.**

Department of Biology, University of Bari Aldo Moro, Italy.

E-mail: andrea.tursi@uniba.it

THE INFLUENCE OF HUMAN ACTIVITIES ON *POSIDONIA OCEANICA* AT TREMITI ISLANDS MARINE PROTECTED AREA (ADRIATIC SEA, ITALY)

Abstract

Posidonia oceanica meadows represent one of the richest and most diverse infralittoral habitats in the Mediterranean Sea, being highly widespread all over the coastal zones of the basin. Although these meadows provide many ecosystems goods and services, they are suffering an alarming decline mostly due to human activities such as coastal development, anchoring, bottom fishing, and dumping. We monitored a *P. oceanica* meadow at Tremiti Islands Marine Protected Area (Adriatic Sea, Italy) in 2003, 2015, and 2020 to assess the conservation status of the meadow in relation to human disturbances. Meadow density, coverage, extent, lower limit dynamics (balisage), as well as phenological and lepidochronological parameters were analyzed. A general decrease in rhizome density, total leaf area, and meadow extent from 2003 to 2020 was observed, most likely due to anchoring practices, as well as the mudding conditions related to the recent harbor implementation. Nevertheless, some encouraging signs of improvement were observed in presence of appropriate management strategies recently put in place, such as the establishment of a mooring buoy field to prevent anchoring practices, underlying the crucial role of protection measures in the conservation of this fundamental coastal habitat.

Key-words: seagrass meadow, coastal management, infralittoral habitat, meadow monitoring, anthropogenic disturbances

Introduction

Coastal habitats are considered among the most diverse and productive areas in the world, showing levels of biodiversity and primary production comparable to rainforests (Duarte and Cebrián, 1996). Despite their key role in the conservation of marine life (Hughes *et al.*, 2009), these habitats are experiencing a global massive decline due to human activities and climate changes. While in certain circumstances global changes represent the main cause of degradation, such as in the case of tropical coral reefs (Hoegh-Guldberg *et al.*, 2007) and of Mediterranean gorgonian forests (Chimienti *et al.*, 2021), other coastal ecosystems are deteriorating due to local disturbances, as in the case of semi-submerged marine caves (Nepote *et al.*, 2017) and seagrass meadows (González-Correa *et al.*, 2007). At the Mediterranean scale, the meadows of the endemic seagrass *Posidonia oceanica* represent one of the most extensive and widespread habitats in coastal areas, covering an approximative surface of 37,000 km² of the infralittoral seabed (Telesca *et al.*, 2015). *Posidonia oceanica* is a key species of the infralittoral zone, being the base of a rich trophic network involving about the 25% of the Mediterranean marine species, which rely on these meadows as spawning, nursery, feeding, and refuge areas (Francour *et al.*, 1997; Buia *et al.*, 2000). Hence, healthy meadows represent valuable hotspots of biodiversity and are considered habitats of great conservation value. Moreover, *P. oceanica* provides several ecosystem goods and services, contrasting ocean acidification and the greenhouse effect (Pergent-Martini *et al.*, 2021), stabilizing the seabed and antagonizing coastal

erosion (Guala *et al.*, 2006). Unfortunately, meadows extent has diminished by at least 34% throughout the Mediterranean Sea in the last 50 years (56% in the western basin) (Telesca *et al.*, 2015). Such meadows depletion is mostly related to human activities, locally and synergically causing the disappearance of these habitats (De los Santos *et al.*, 2019). The main anthropogenic stressors include anchoring (Montefalcone *et al.*, 2008), bottom-contact fishing (Vlachopoulou *et al.*, 2013), and coastal development (Ruíz *et al.*, 2003), causing direct and indirect damage to the meadows. Other disturbances are intensive aquaculture activities and dumping (Panayotidis *et al.*, 1990; Pazzaglia *et al.*, 2020), as well as other human-related stressors such as the spread of competitive alien species (Piazzi *et al.*, 2001; Ballesteros *et al.*, 2007), mucilaginous blooms and global warming (Marín-Guirao *et al.*, 2019). The awareness of such decline led to the establishment of EU directives, such as the Habitat Directive (92/43/CEE), Water Framework Directive (WFD, 2000/60/EC), and Marine Strategy Framework Directive (MSFD, 2008/56/EC), which listed *P. oceanica* meadows as priority habitats for conservation. Moreover, Marine Protected Areas (MPAs) and marine Special Areas of Conservation (SACs) have been designed in areas with seagrass meadows. In fact, despite the overall decline observed in the last decades, effective management measures have locally led to the partial recovery of the meadows, especially in MPAs, proving the crucial role of human actions on meadow conservation status (Guillén *et al.*, 2013; De los Santos *et al.*, 2019). In this context, our study is focused on the monitoring of *P. oceanica* at Tremiti Islands MPA, which hosts the northernmost meadow of the Italian coasts in the Adriatic Sea. The monitoring was carried out in 2003, 2015, and 2020 to assess the ecological status of the meadow over time. It allowed to understand the influence of human disturbances on *P. oceanica* in the MPA, as well as the effectiveness of management actions (e.g., establishment of mooring buoy fields).

Materials and methods

The monitoring of *P. oceanica* was carried out at Tremiti Islands MPA (42°7.38' N, 15°30.02' E), in the southern Adriatic Sea, 12 nautical miles north of the Gargano Promontory (Apulia, Italy), where a fragmented meadow is present. This meadow is divided into three main submeadows, here called submeadow A, B, and C. Submeadows A and C run along the coasts of San Domino Island, while submeadow B is located between San Nicola Island and a smaller island called Cretaccio (Matarrese *et al.*, 2000) (Fig. 1). The three submeadows are affected by several anthropogenic disturbances, including anchoring, recreational boating, artisanal fishing, and coastal development (Tursi *et al.*, 2021).

One sampling station per submeadow was considered in 2003, 2015, and 2020 (Fig. 1). In detail, M1 and M3 sampling stations were placed at the lower limit of the submeadow A and C, respectively. In fact, the lower limit often represents the meadow part that is more sensitive to human disturbances (Lopez Y Royo *et al.*, 2010). Because of the absence of a uniform lower limit, the submeadow B was studied in its intermediate portion (M2 sampling station). At each sampling station, absolute density (shoots m⁻²) was estimated by SCUBA diving by counting the number of rhizomes in a sampler frame of 40 cm × 40 cm (nine replicates).

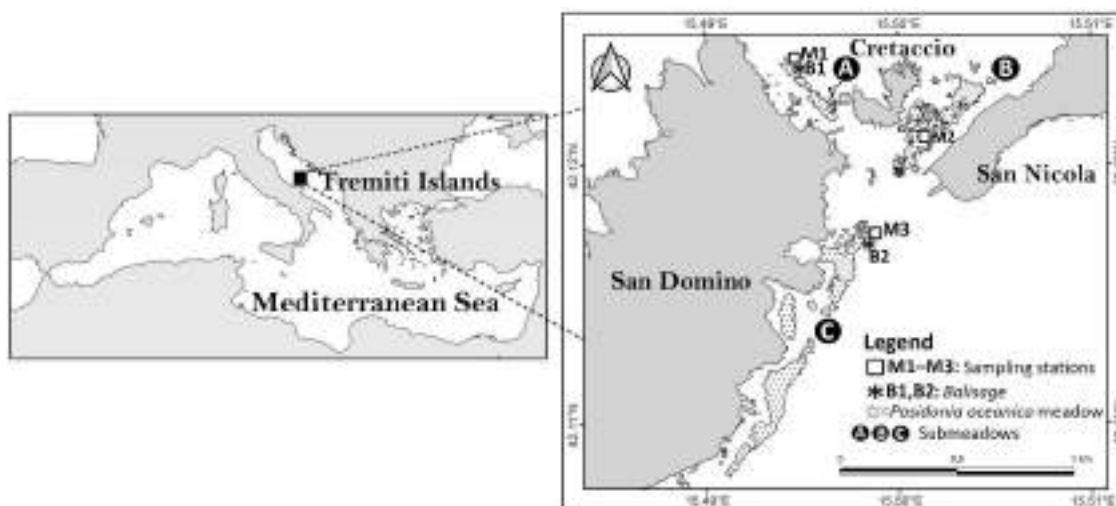


Fig. 1: Map of the study area showing Tremiti Islands' location in the Adriatic Sea, the distribution of the *Posidonia oceanica* meadow, the sampling stations (M1–M3), and the *balisage* sites (B1, B2).

Then, the mean relative density (shoots m^{-2}) was assessed by multiplying the absolute density for the coverage (%) of *P. oceanica* estimated on a surface of five-meter radius. Moreover, six living orthotropic rhizomes were collected at each sampling station. The phenological parameter Leaf Area Index (LAI, $m^2 m^{-2}$) was calculated by multiplying the mean leaf surface of each bundle ($cm^2 shoot^{-1}$) by the relative density of the meadow (shoots m^{-2}) (Buia *et al.*, 2000). The lepidochronological analysis involved the measurement of the scales' thickness (Pergent *et al.*, 1989). Then, rhizome age was estimated by counting the number of lepidochronological cycles, representing the period between two minimums of the scales' thickness, and corresponding to a year. All values were expressed as mean \pm standard deviation. The extent of the meadow was evaluated through habitat mapping carried out using the Klein 3000 Side Scan Sonar, then data were imported into a GIS environment for interpretation and cartography. The lower limit of the meadow was monitored with two *balisage* systems (B1, B2), each one consisting of five metal pickets implanted in 2015 at the lower limits: B1 at submeadow A and B2 at submeadow C (Fig. 1) (Bertrand *et al.*, 1986; Tursi *et al.*, 2021). Then, distances between pickets and meadow limits were evaluated in 2020 to assess any progression and/or regression of the lower limit.

Statistical analyses were carried out using Past 4.03 software. The Shapiro–Wilk test was used to determine the normality of the data distribution (p values > 0.05). Then, the non-parametric Kruskal-Wallis test was run to verify the presence of statistically significant differences in density, LAI, and rhizome age between the three monitoring periods.

Results

A considerable worsening of the meadow health status occurred from 2003. In fact, the meadow showed to be much more rarefied in 2015, as attested by the relative density and the LAI values, which both diminished by more than 70% in 12 years (Tab. 1, Fig. 2). Meadow extent also dropped by about 6.4% between 2003 and 2015. Rhizome age did not vary significantly, although the prevalence of old rhizomes suggested the scarce occurrence of stolonization processes. However, the decrease in density and LAI slowed down from 2015 to 2020, and the meadow even showed some improvements in some areas, although not statistically significant (Tab. 1). On the other hand, meadow extent

continued to reduce by another 14.1% with respect to 2015. The *balisage* analysis showed a steady situation of the meadow lower limit at B1 with differences of a few centimeters between 2015 and 2020, while a regression of more than 11 linear meters was observed at B2. However, most of the pickets of the two systems were lost, confirming the occurrence of mechanical impacts, such as anchoring practices, affecting the meadow.

Tab. 1: *p*-values from the comparison between the three monitoring years.

* $p < 0.05$; *** $p < 0.001$

	Relative density (shoots m ⁻²)	Leaf Area Index (m ² m ⁻²)	Rhizome age (year)
2003 vs 2015	1.87×10^{-4} ***	2.043×10^{-6} ***	0.7811
2015 vs 2020	0.102	0.3189	0.1629
2003 vs 2020	2.45×10^{-6} ***	7.77×10^{-7} ***	0.4299

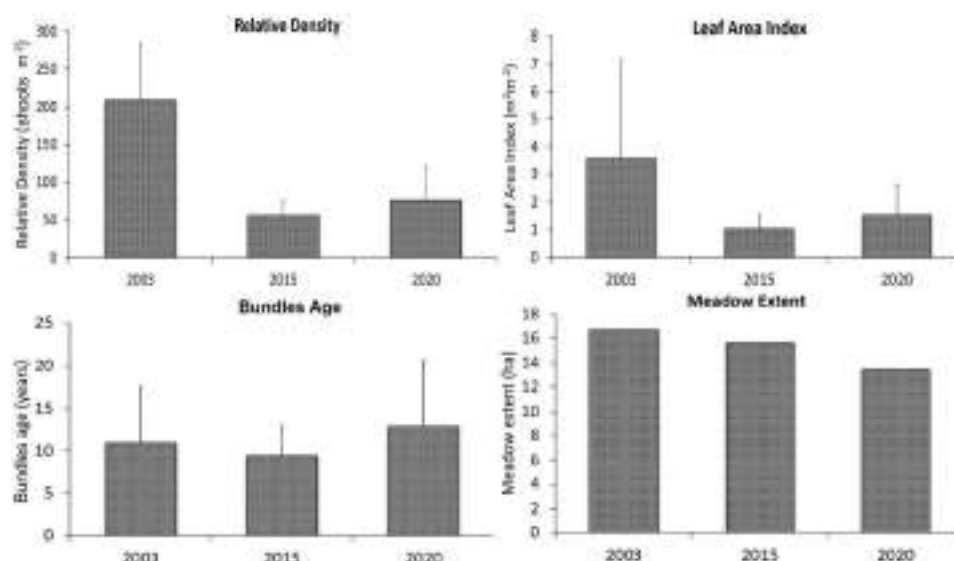


Fig. 2: Trend of the mean values (+sd) of the relative density (shoots m⁻²), Leaf Area Index (m²m⁻²), bundles age (lepidochronological years), and meadow extent (ha) of *Posidonia oceanica* at Tremiti Islands Marine Protected Area in 2003, 2015, and 2020.

Discussions and conclusions

The overall decline of the meadow observed in less than 20 years could be attributed to local anthropogenic stressors affecting this habitat, most of all anchoring practices and coastal development. However, these impacts were unevenly present along the meadow, jeopardizing only some of its portions. For instance, submeadow B underwent a progressive fragmentation over time mostly due to anchoring activities, which led to the mechanical destruction of the bundles, resulting in evident scars and a patchy coverage. The submeadow A was stressed by anchoring too, as attested by the absence of most of the *balisage* pickets during the 2020 monitoring. However, signs of recovery were observed in the last five years of monitoring, as attested by the presence of a new meadow patch in its northernmost region testifying to a certain expansion. On the other hand, the regression of the submeadow C could be mainly related to the enhancement of sedimentation rates that occurred in the last 20 years, which led to massive shading conditions affecting photosynthesis and metabolic exchanges. The increase in sedimentation is most likely a consequence of coastal development activities and the implementation of the small harbor nearby the meadow. Despite the negative trend shown by *P. oceanica* at Tremiti Islands MPA, some encouraging signs of recovery have been observed in the last five years of

monitoring, particularly where effective management measures have been put in place. This is the case of the submeadow B, where a mooring buoy field with a subsurface float system was established in 2017 to avoid both anchoring practices and the abrasion of the chains on the seabed. Although a mooring buoy field was also established in the proximity of the submeadow C, this meadow area did not show any sign of recovery, even further deteriorating from 2015. This is likely due to the high sedimentation rates, still affecting this submeadow and hindering any improvement of its conservation status.

Our study provided evidence of the harmful effects of local human activities, such as anchoring and coastal development, on this fragile habitat, confirming that local actions can strongly affect *P. oceanica* meadows. This means that local anthropogenic impacts can severely damage *P. oceanica* meadows but, at the same time, local conservation actions can effectively foster the recovery of the meadows, highlighting the importance of implementing appropriate management measures targeting the conservation of this fundamental habitat.

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Daniele VENTURA, CASOLI E., MANCINI G., BELLUSCIO A., PACE D. S., ARDIZZONE G.D.

Department of Environmental Biology, University of Rome “La Sapienza”, Italy.

E-mail: daniele.ventura@uniroma1.it

3-DIMENSIONAL MAPPING FOR FINE-SCALE CARTOGRAPHY OF *POSIDONIA OCEANICA* SEAGRASS MEADOW LIMITS USING UNDERWATER STRUCTURE FROM MOTION PHOTOGRAMMETRY

Abstract

*Seagrasses are considered one of the most important shallow-marine ecosystems and ensure, at the global scale, a large plethora of goods and services for their ecological, physical, and economic values. Although large-scale monitoring plans and transplanting operations have been carried out, a significant effort has been required to define new methods to effectively map such large underwater habitats. Here we propose an SfM-based approach for mapping seagrass limits over large areas to produce ultra-high spatial resolution orthophoto mosaics and Digital Elevation Models (DEMs) based on the processing of underwater imagery to create 3D surfaces of such key habits digitally. We applied our approach on a *Posidonia oceanica* meadow located in front of the Cala di Mezzo cove (Giglio Island, Tuscany), near the northern site impacted by Costa Concordia shipwrecking. More than 1500 HD images were acquired with a GoPro Hero 10 action cam mounted on a Diver Propulsion Vehicle (DPV) to map an area of approximately 1.6 ha. The imagery was processed by photogrammetry software to produce cartographic raster products with an ultra-high spatial resolution (< 1 cm/pixel). After image segmentation and classification in GIS environments, we achieve a fine identification of the natural meadow and the transplanted fragments used to restore such area. In addition, elevation data were also used to compute the canopy height of the natural meadow. This method provides a valid alternative to the traditional methods for creating centimetre-level accuracy cartographic products of living *P. oceanica* and could be applied to map other complex benthic habitats.*

Key-words: seabed mapping, habitat characterization, 3D point clouds, 3D modelling, high-spatial-resolution imagery

Introduction

Seagrasses are considered one of the most important shallow-marine ecosystems and ensure, at the global scale, a large plethora of goods and services for their ecological, physical, and economic values (Larkum *et al.*, 2006). Unfortunately, since the last century, we have witnessed a continual loss of seagrass habitats coupled with the decline of coastal environmental quality due to natural processes and human-mediated impacts (Francour *et al.*, 1999; Boudouresque *et al.*, 2009; Unsworth *et al.*, 2019). Nevertheless, a robust legal framework, most seagrass meadows remain under significant pressure resulting in a decline condition (Unsworth *et al.*, 2019).

Posidonia oceanica (L.) Delile is a protected Mediterranean endemic species of seagrass that forms extensive meadows from the surface to depths of 40 m, depending on water transparency and temperature (Boudouresque *et al.*, 2009). Over the last 100 years, a global decline with losses exceeding 25% worldwide has been observed for most species

of seagrass (Waycott *et al.*, 2009), including *P. oceanica*, whose loss of area has been evaluated to be 10% due to coastal human population and steadily increasing of man-made coastline, which is responsible for the fragmentation and destruction of this sensitive habitat (Marre *et al.*, 2020). Although large-scale monitoring plans and transplanting operations have been carried out, a significant effort has been required to effectively define new methods to monitor such significant underwater habitats since current methods mainly rely on traditional SCUBA divers using direct in-situ measures (Ventura *et al.*, 2022). Consequently, to support large-scale monitoring programmes, accurate mapping methods are required to integrate local evaluations into extensive assessments concerning the status of meadows.

Structure from Motion (SfM) photogrammetry provides new opportunities to extract a plethora of fine-scale variables from 3D digital models of the underwater environment. Underwater SfM photogrammetry involves the virtual reconstruction of the seabed by using numerous overlapping digital images that can be acquired from different platforms such as divers, Remotely Operated Vehicle (ROVs), Autonomous Underwater Vehicle (AUVs), and towed cameras. SfM-derived products such as 3D point clouds, triangular meshes, Digital Elevation Models (DEMs), and orthomosaics can be analyzed in several ways, providing an excellent tool for accurate measurement of lengths, areas, and volumes that are difficult or even impossible to get in situ with traditional methods (Ventura *et al.*, 2022).

In this study, we tested an operational and low-cost method for producing centimetric mapping of complex underwater environments over a large spatial extent in which both natural and transplanted meadows characterize the seabed. In addition, to speed up the identification of meadow and transplanted fragments, we tested a segmentation method followed by a supervised image classification approach to obtain informative thematic raster maps of the surveyed area.

Material and methods

We applied our approach on a *P. oceanica* meadow located in front of the Cala di Mezzo cove (Fig. 1a, Giglio Island, Tuscany), near the site impacted by Costa Concordia shipwrecking. The shallow bay was characterized by a patchy meadow extending from 5 to 25 m depth with heterogeneous seabed showing sandy, rocky, and dead “matte” areas. In this area, besides the natural meadow, a transplanted meadow is also present, which was part of the restoration programme carried out after the Costa Concordia site remediation project (Mancini *et al.*, 2022). This area was also mapped in 2018 by an acoustic survey using Multi Beam Echo Sounder (MBES) data (Fig. 1b). Bathymetric data were derived from a hull-mounted RESON Seabat 7125 SV2 operating at 400 kHz and emitting up to 512 beams across an angular coverage of 140°–165° wide swath. Vessel positioning during MBES surveys was provided by a dual frequency GNSS system (Leica 1200) using RTK corrections leading to a centimetric positioning in the x, y, and z axes. The MBES bathymetry data was interpolated at 20×20 cm grid size and visualized as shaded-relief topographic imagery. To speed up and improve the extent of the surveyed area by the diver, a diver propulsion vehicle (DPV, Suex X-joy 7) was used to optimize underwater image acquisition. DPV speed was set to ‘low’ (~25 m/min), and underwater HD images (5568 x 4176 pixels) were acquired with a GoPro Hero 10 (23 Mpix sensor) in time-lapse mode, allowing interval shooting every 2 sec.

Images processing was carried out using SfM algorithms through Agisoft Metashape v 1.6.2 (Agisoft LLC, Russia), a low-cost commercial 3D reconstruction software widely

used in the scientific community. We used the self-calibration procedure implemented in Metashape to absorb refraction at the camera port-water interfaces (Marre *et al.*, 2020). After images alignment, a sparse point cloud was generated and georeferenced by adding ten Ground Control Points (GCPs) in Metashape by using well identifiable features from MBES data such as rock edges. Subsequently, the density of sparse point clouds was enhanced by Multi-View Stereo (MVS) algorithms. This process resulted in a dense XYZ point cloud of 40 million points. The dense point supported the generation of a fine scale Digital Surface Model (DSM) used to produce both 10 cm contours and the high spatial resolution GeoTIFF orthophotomosaic of the bay. Subsequently, the three-band (RGB) raster mosaic was imported into the ESRI GIS software ArcMap v.10.6.1 to run a supervised Support Vector Machine (SVM) image classification routine following a mean shift segmentation of the native high spatial resolution imagery. The classified output was generalized by removing small, isolated regions with the majority filter, boundary clean and nibble tools. Using SfM-based DSM we estimated the mean canopy height of *P. oceanica* patches with mesh to cloud distance tool in Cloud Compare v. 2.12 (Girardeau-Montaut, 2016) after separating the *P. oceanica* class to ground points with the Cloth Simulation Filter (CSF, Zhang *et al.*, 2016).

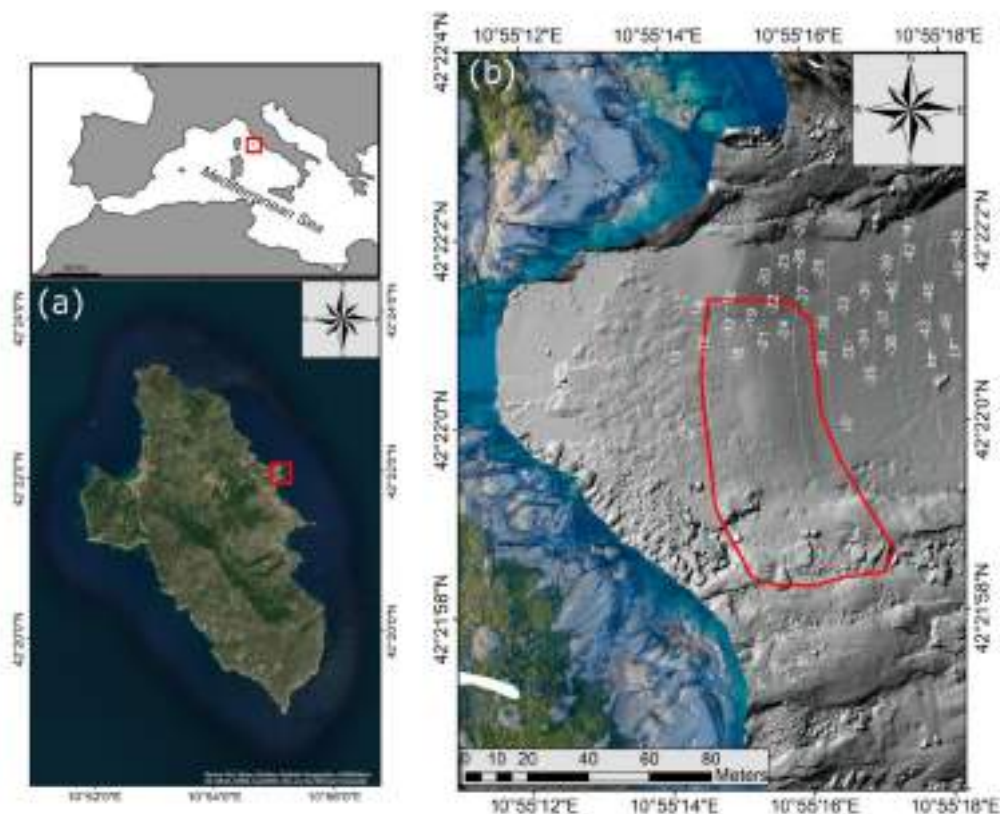


Fig. 1: a) Location of the study area on the NW coast of Giglio Island (Tuscany, Italy). b) MBES data showing the mapped area (red polygon) through our SfM-based method.

Results

After an acquisition time of 1 h 10 min dive, 1755 HD images to map the area from 11 up to 33 m depth. The generated DSM (Fig. 2a) and orthophoto mosaic (Fig. 2b) covered an area of 3800 m² with a spatial resolution of 1.3 and 1.7 cm/pix, respectively. The SVM classification allowed the identification of five main cover classes: granite rocks,

algae/matte on rocks, soft bottoms, dead ‘matte’, and *P. oceanica* (Fig. 2c). High-resolution orthophoto mosaics allowed an excellent overall classification accuracy of 85.6 % with an acceptable separation of *P. oceanica* from the other seabed cover classes. The major land-cover misclassification errors involved spectral confusion among *P. oceanica* and dead ‘matte’, illustrating the difficulty in separating land-cover classes based on filtered spectral signatures due to water column. In the transplantation area, the weak contrast and dimensions of transplanted cuttings led to poor classification accuracies. The area covered by *P. oceanica* was 954 m² corresponding to a total percent cover of 25 %. The high percentage (24 %) of dead ‘matte’ class extending both on sand and rocks coupled with the significant presence of soft bottoms (39 %) which might hide other dead ‘matte’ layers (especially in the shallow area comprise between 11 to 20 m) depict the health status of this meadow after several disturbance events on a long-term basis. By separating the meadow class from other elevation data, we estimated the mean canopy height of 0.68 ± 0.24 m, with lower values (0.30 ± 0.15 m) recorded on the lower limits of the meadow (Fig. 3).

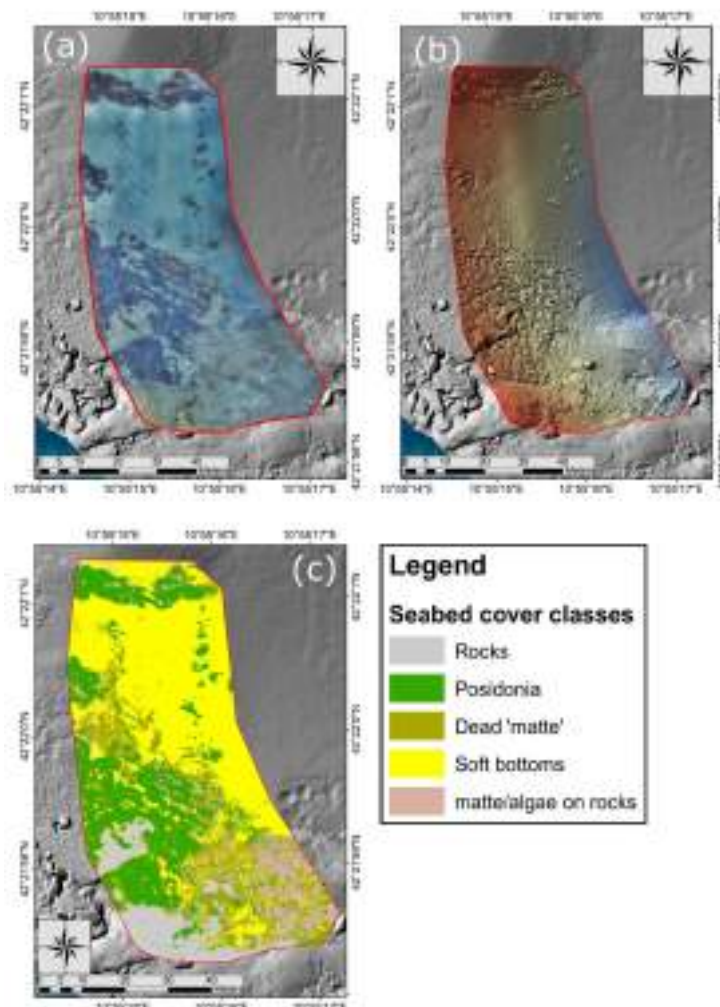


Fig. 2: (a) High spatial resolution (1.3 cm/pix) RGB orthophotomosaic. (b) Digital Surface Model (DSM) of the mapped area through SfM photogrammetry-based protocol. (c) Thematic map after image segmentation and SVM classification.

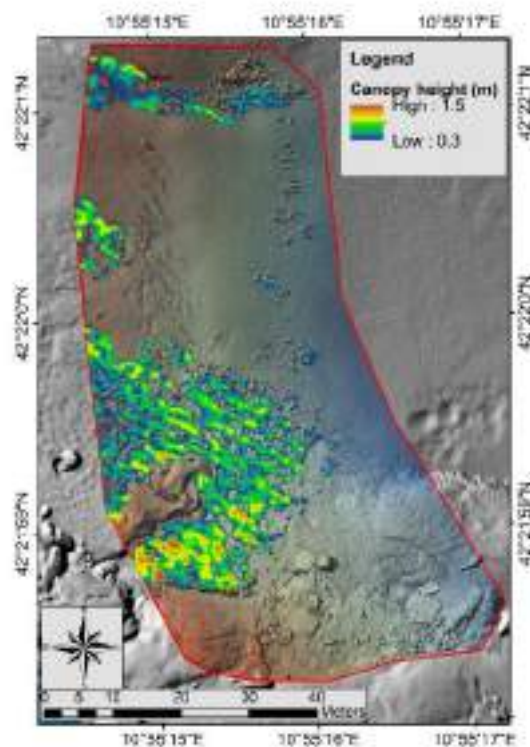


Fig. 3: Canopy height model of the meadow after computing cloud-to-mesh distances in Cloud Compare.

Discussion and conclusions

Our method aimed to develop an efficient operational methodology that is automatic and reproducible for fine-scale mapping applications of seagrass habitats. This protocol is suitable in almost all areas where patchy meadows are interrupted by other features such as rock and sandy areas or where the seagrass shows low densities, such as along the lower limits or inside transplanted areas characterized by a reduced number of fragments/m². In such circumstances, the low densities of leaves ensure good results during image alignment and tie points detection, which are critical steps in SfM processes. As in other studies that mapped seagrass beds using underwater photogrammetry and built a classifier based either on luminance and colourimetry and textures (Rende *et al.*, 2015; Mizuno *et al.*, 2017) also, the pixel-based approach we used in SVM classification can be primarily affected by water depth and turbidity, relying on red, green and blue (RGB) bands values. Therefore, the variability between meadows in terms of depth, substrate, and lighting conditions, makes it challenging to produce a generalizable classifier based only on colours or luminance. Another method, which uses seagrass movement and texture homogeneity (Marre *et al.*, 2020), reported higher accuracies of classification even in the presence of dead matte, which may affect classification algorithms based on colour or luminance. Moreover, essential metrics such as canopy height and elevation of submerged features could be computed using accurate SfM-based elevation data. However, it should be noted that in this case, we can easily define underwater GCPs by using the MBES high-resolution data; otherwise, more laborious work is needed to deploy accurate control points on the seabed to scale and georeferenced the model (Ventura *et al.*, 2022). Despite such limitations, underwater mapping using SfM algorithms can be regarded as a reliable method for capturing and storing a large amount of information on the substrate and at the community

level, providing an excellent tool for accurately monitoring seagrass areas on a large scale. When fast and frequent monitoring is required (e.g., after an impact event or during transplanting operations), this workflow allows the optimization of resources and might serve as a proof of concept to implement this cost-effective and non-intrusive monitoring method as a new tool to support the management and conservation of seagrass beds.

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Imen ZRIBI, ELLOUZI H., MNASRI I., ABDELKADER N., BEN HMIDA A., DORAI S., DEBEZ A. CHARFI-CHEIKHROUHA F., ZAKHAMA-SRAIEB R.
University of Tunis El Manar, Faculty of Sciences of Tunis, Laboratory of Diversity, Management and Conservation of Biological Systems, LR18ES06, Tunis, Tunisia.
E-mail: imenzribi4@gmail.com

EFFECT OF FILAMENTOUS *MACROALGAE CHAETOMORPHA LINUM* ON *CYMODOCEA NODOSA*: DOES CLONAL INTEGRATION ALLEVIATE THE *MAGROALGAE* IMPACTS?

Abstract

Seagrass meadows are known as one of the most valuable ecosystems on the marine environment. In shallow Tunisian coastal waters, seagrass and seaweed occur together. Ecological theory suggests that the presence of co-existing species in the same habitat would create competitive interaction. Although the performance and survivorship of seagrasses are affected by environmental stressors, clonal plants have been shown to successfully survive in stressful environments. In the present study, *in situ* experiment were conducted for 90 days in Bekalta (eastern coast of Tunisia) in order to assess the response of the seagrass *Cymodocea nodosa* (Ucria) Ascherson to shading by filamentous algae *Chaetomorpha linum*. Two rhizome connection treatments connected (allowing integration) and severed (preventing integration) were monitored every 15 days. Clonal integration was estimated by comparing different parameters such as shoot growth rate, and physiological and biomass features of rhizome-connected and rhizome-severed treatments. The hypothesis was that clonal integration positively affects the biomass allocation, growth, and physiology of the seagrass *Cymodocea nodosa* and thus improves its performance in response to shading. After 90 days of shading by *C. linum*, we found that for the two rhizome connection treatments the presence of the macroalgae induced an increase in photosynthetic pigments content and a decrease in soluble proteins and sugars concentrations. Our work suggests that these parameters could be used to monitor *Cymodocea nodosa* meadow in the early stage of eutrophication.

Key-words: Clonal integration, proteins, seagrass-seaweed interaction, shading, sugar content

Introduction

Seagrass ecosystems rank among the most productive habitat in coastal zones that play a primary role in supporting ecosystem services such as carbon sequestration, hosting biodiversity and sediment stabilization (Duarte *et al.*, 2013). Despite their importance, seagrass meadows are facing a global decline (Unsworth *et al.*, 2018). This loss is mainly due to increasing pressure from industrial, domestic and agricultural activities around these areas (Waycott *et al.*, 2009). Nutrient input from land to coastal waters causes a global regression of seagrass meadows. Excess nutrients mainly nitrogen and phosphorus trigger drift macroalgae growth (Pazzaglia *et al.*, 2020), which results in light limitations for the seagrass (Burkholder *et al.*, 2007). In eutrophic zones, the proliferation of macroalgae can induce a regime shift by converting stable seagrass meadows into less stable macroalgae beds (Petersen *et al.*, 2008). Therefore, it is crucial to understand the impact of the initial macroalgae accumulation on seagrasses to preserve seagrass habitats and the ecosystem services they provide. The present study aims to (i) assess *Cymodocea nodosa* response to shading by *Chaetomorpha linum* using different types of indicators (growth, structural and

physiological) (ii) investigate whether responses could be modulated by the clonal integration of the plant. We hypothesized that preservation of the seagrass clonal integration would ameliorate the stress induced by shading on seagrass vitality.

Materials and methods

The study was carried out in Bekalta (N 35°38'12.6"; E 11° 00' 33.5") in the eastern part of Monastir bay from September to December 2019. Monastir bay is a semi-enclosed lagoon that has been under anthropogenic pressure for many years, it is characterized by significant eutrophication (Sghaier *et al.*, 2017). The bay is an area of recurrent episodes of algae proliferation Chaouch & Harzallah (2020). *Chaetomorpha linum* is considered as one of the most abundant seaweeds in the bay Zaoulai & Ben Charrada (2010). We conducted a macroalgae addition experiment within *Cymodocea nodosa* meadow located in shallow water within a depth of 1.5-2 m. In total, we had 12 plots where six had served as control (no macroalgae addition) whereas the remaining six plots were covered by *Chaetomorpha linum* for 90 days. In order to assess the role of clonal integration in *C. nodosa* response to shading by *C. linum*, shoots from 3 plots per each treatment (control and shading) were isolated from adjacent shoots by cutting horizontal rhizomes around each plot down to 30 cm inside the seabed (Tuya *et al.*, 2013). A minimum distance of 2 m between plots from the same treatment has been implemented.

Growth and biomass measures: For shoot growth, ten shoots of *Cymodocea nodosa* were marked at each sampling occasion using the leaf punch method outlined by Short & Duarte (2001). In the laboratory, the newly formed tissues were estimated using the shift of the marking hole along each leaf. The shoot growth rate ($\text{mm shoot}^{-1} \text{day}^{-1}$) was calculated by dividing the length of the new growth by the number of days in the experimental period.

Plant Biomass was measured at the end of the experiment using a 15 cm diameter PVC corer placed randomly within each experimental plot. Biomass samples were sorted into above (shoots) and below-ground sections (horizontal rhizome and roots) and then dried at 60°C to a constant weight.

Physiological measures: Total soluble sugars were quantified at the end of the experiment according to the method described in Shields & Burnett (1960). In a nutshell, dried samples from above and below-ground parts (25 mg) were grounded and twice extracted in hot 80% ethanol. Samples were centrifuged (3000 rpm for 20 min). Soluble sugars content was analysed spectrophotometrically at 640 nm using an anthrone assay standardized to glucose and expressed on a dry weight basis ($\text{mg g}^{-1} \text{DW}$).

Photosynthetic pigments were extracted overnight in 5 ml acetone (80%) in the dark and at 4°C. The total chlorophyll and carotenoid content were quantified using a spectrophotometer at wavelengths of 470 nm, 646 nm and 663 nm.

Soluble Protein concentration was determined on triplicate leaf samples using the Bradford method (Bradford 1976) with bovine serum albumin (BSA) as a standard.

Statistical analyses: Means were compared using a two-way analysis of variance (ANOVA). In order to comply with the assumption of parametric procedures, data were tested for normality by the Shapiro-Wilk test and for homogeneity by the Levene test. All statistical analysis was performed using R v4.1.2.

Results

Growth and above-below biomass are given in Fig. 1 and 2. The results showed that at the end of the experiment shoot growth rate was significantly affected by both macroalgae

load and maintenance (or not) of the clonal integration. Still no interaction was found between the two factors ($p > 0.05$). All plants showed a steep decline in shoot growth rate. In our study, shading and preservation of the clonal integration didn't cause a significant change in below-ground biomass whereas the above biomass of *C. nodosa* was significantly impacted by the presence of *C. linum*. Furthermore, shading induced a decrease in the above-ground biomass of *C. nodosa* regardless of the presence or absence of connection with the neighbouring plants.

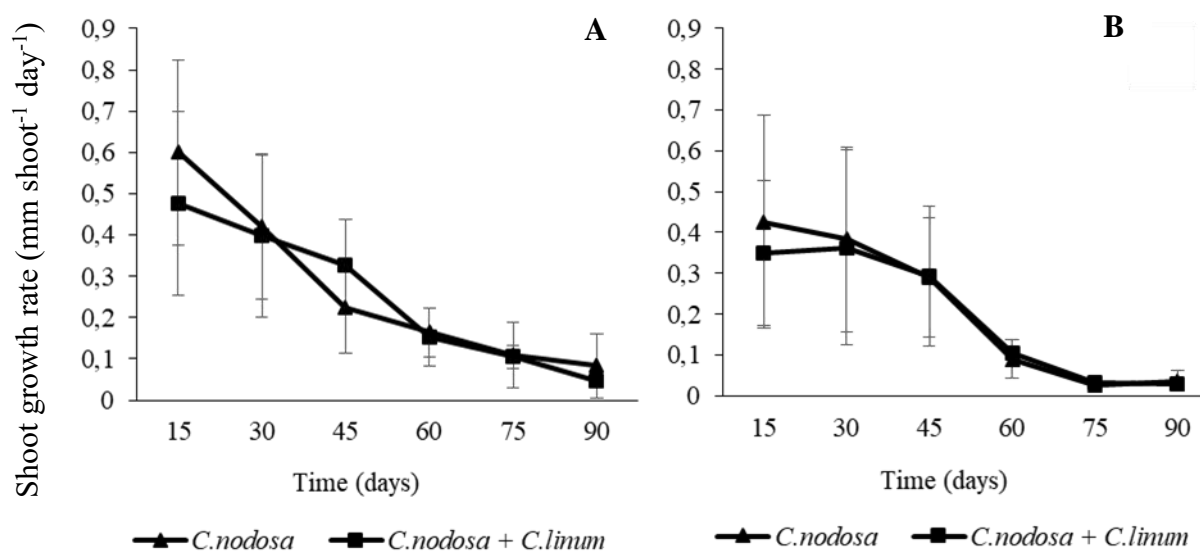


Fig. 1: Effect of *Chateomorpha linum* on the shoot growth of *Cymodocea nodosa* over time. Values are means \pm SE. (A) the clonal integration was maintained and (B) severed.

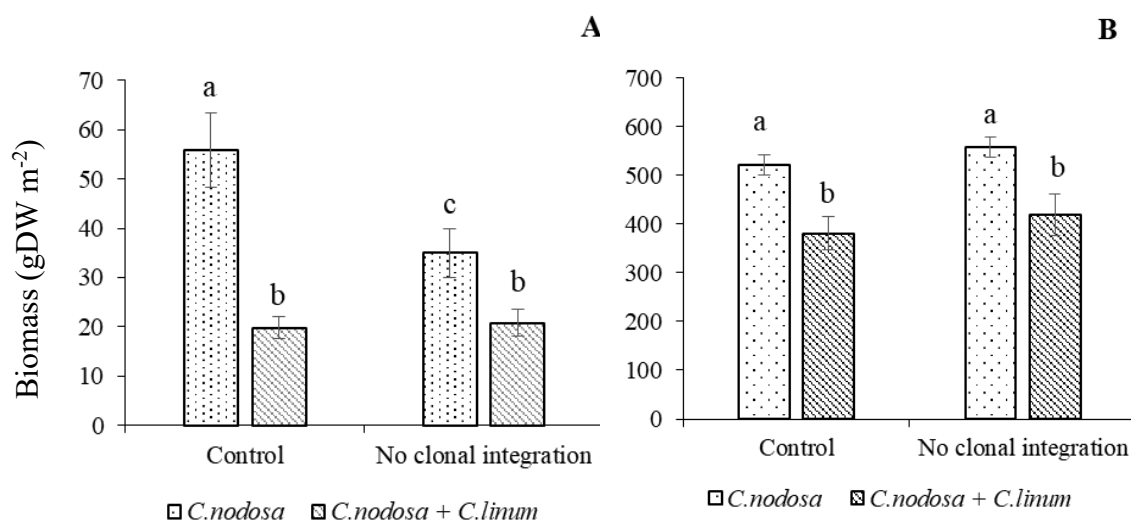


Fig. 2: Dry biomass measured in rhizomes severed treatment and control plants at the end of the 90-day manipulative experiment. Measures are mean \pm SE. (A) Above biomass; (B) Below biomass.

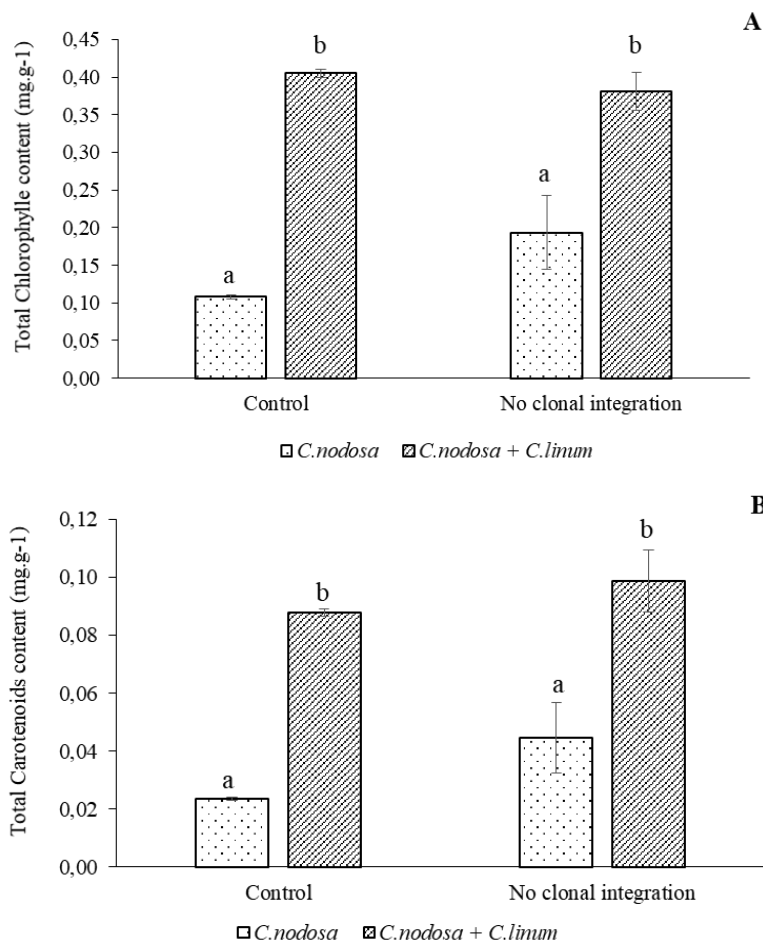


Fig. 3: Photosynthetic pigments content in leaves of *Cymodocea nodosa* after 90 days cover by loads of *Chaetomorpha linum*. (A) total chlorophyll content; (B) total carotenoids content. Different letters on bars indicate significant differences. Values are means \pm SE (n=3).

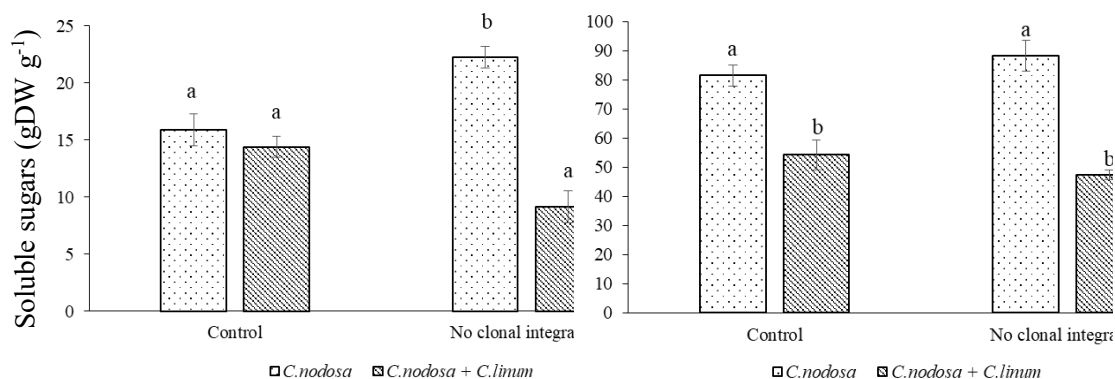


Fig. 4: Soluble sugars concentrations leaves (right) and rhizome-roots (left) of *Cymodocea nodosa* after 90 days cover by loads of *Chaetomorpha linum*). Values are means \pm SE (n=3). Shared letters indicate no significant difference.

Shading by *C. linum* loads had a significant effect on total chlorophyll and carotenoid concentrations of the leaves (Fig. 3). Chlorophyll concentration was 3 folders higher in the plots subjected to macroalgae loads whereas the carotenoid content was twice higher.

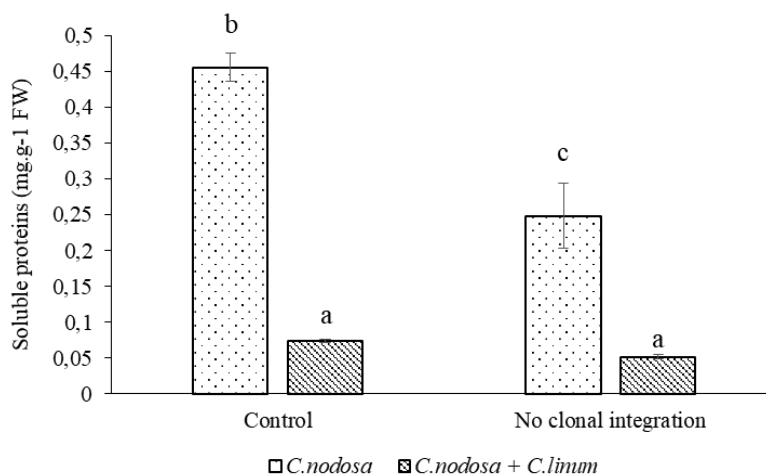


Fig. 5: Soluble protein content in leaves of *Cymodocea nodosa* after 13 weeks cover by loads of *Chaetomorpha linum*. Values are means \pm SE (n=3).

In the no clonal integration treatment, both soluble sugars concentration in leaves and rhizomes were significantly affected by the presence of *C. linum*. A significant decrease was observed in these tissues (Fig.4). The foliar soluble protein concentrations decreased by almost 50% when *Cymodocea nodosa* was covered by *C. linum* load.

Discussion and conclusions

Coastal eutrophication is often followed by the accumulation of drifting macroalgae that may cover the entire seagrass meadow. In the framework of this study, we tried to understand the short-term interaction between the seagrass *Cymodocea nodosa* and the Chlorophyta *Chaetomorpha linum* and to investigate the hypothesis that seagrass may outlive the macroalgae stress via translocation of resources.

Alcovero *et al.* (1999) highlighted that during periods of light-limited photosynthesis, the mobilisation of internal carbon reserves (carbohydrates) represents a survival strategy adopted by seagrass. The reduction of sucrose concentration in the below-ground tissues suggests a possible sucrose mobilisation throughout the plant to meet carbon demand (Burn *et al.*, 2003). The results showed that in shaded conditions, *C. nodosa* tended to reduce tissue soluble proteins. Similar patterns have been observed in submerged macrophytes exposed to low-light conditions (Zhang *et al.*, 2010).

This study clearly showed that physiological variables (photosynthetic pigments; soluble proteins and sugars) responded to reduced light availability induced by *C. linum* loads within a 90 days period. These variables may be useful as sub-lethal indicators of light reduction stress in *C. nodosa* meadows.

Some previous studies have highlighted the importance of clonal integration in buffering stress caused by nutrient enrichment or sediment burial (Tuya *et al.*, 2013). However, our experiments didn't show a clear nexus between physiological integration and *C. nodosa* responses to shading, at least in short time.

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POSTERS

ALDO ANNUNZIATELLIS, AGNESI S., LILLIS H., O'KEEFFE E., ALLEN H., CASTLE L., PINDER J., TUNESI L.

Italian institute for environmental protection and research (ISPRA), Via V. Brancati 60, Roma (Italy).

E-mail: aldo.annunziatellis@isprambiente.it

EMODNET SEABED HABITATS: A COMPLETE REPOSITORY OF STANDARDISED HABITAT AND COMPOSITE MAPS

Abstract

Since 2008, EMODnet Seabed Habitats (ESBH) has been working to fill the gap in the availability of environmental spatial data and now hosts the largest European collection of habitat maps from individual surveys and survey-based sample points (over 1,000 maps and 500,000 sample points). The portal hosts a unique web catalogue of benthic habitat spatial data providing a great opportunity to create new products which aim to answer specific questions and supporting a variety of stakeholder's needs. In the Mediterranean, the needs of the RAC/SPA, the UNEP-MAP centre for the implementation of the Regional Action Plans for the conservation of marine key benthic habitats, were discussed and addressed by the creation of specific composite maps. All the collected/created products, together with all the input layers needed to produce the models are freely disseminated online through the ESBH portal (www.emodnet-seabedhabitats.eu).

Key-words: Seagrass, Coralligenous and other biogenic concretion, Essential Ocean Variables, Habitat maps, Data collection

Introduction

The European Marine Observation and Data Network (EMODnet) began in 2008 with the main goal being the improvement of quality and typology of collected spatial data. In 2009, ESBH produced the first broad scale modelled map of some European basins, including the western Mediterranean. Subsequently this map was extended to cover all European seas and complemented with the collation of habitat maps from surveys. Since 2019, it has improved the products freely available through the portal by collating and disseminating ground-truth habitat point data from surveys and individual habitat models. Currently, the collection is being enriched also with composite products, coastal wetland data (CW) and Essential Fish Habitat (EFH). With respect to the Mediterranean, the ESBH project is collaborating with RAC/SPA, the UNEP-MAP centre in charge for the Regional Action Plans for the conservation of Key Marine Habitats (MKH), namely: Marine Vegetation; Coralligenous and other calcareous bioconcretions and Dark habitats by producing specific composite maps.

Materials and methods

All the spatial data products hosted in the ESBH repository are standardised and ranked based on their confidence, using specific rules depending the typology of the spatial product. The modelled map (AKA EuSeaMap) was created by applying a specific set of rules for the combination of environmental input layers (light, energy, seabed substratum, depth, oxygen and salinity). Where possible, the marine section of the European Nature Information System (EUNIS) habitat classification scheme was used, but it is also available in the Barcelona Convention habitat typology in the Mediterranean. The survey sample points and the individual habitat maps host habitat data in the broadest sense. In fact, habitat data can encompass all the

levels of detail from biocoenosis to broad physical descriptions. The composite maps are based on the above mentioned sample points and individual habitat maps. These maps are constructed as follow: 1. Identification of the relevant data sources. 2. Selection of the most appropriate classification systems for the target habitat(s). 3. Data query and extraction from the identified sources. 4. Application of spatial rules to remove overlapping information and clean the composite layer (if needed). 5. Compilation of a single data product, by clearly maintaining the provenance information assigned to every polygon and point in the attribute table.

Results

The portal now offers a modelled habitat map covering over 11,000,000 km², around 1,000 habitat maps, 500,000 habitat point data, 100 individual habitat models, as well as composite products that contribute towards understanding of MKHs, among other habitats. Examples include Essential Ocean Variables (EOVs) like live Hard coral cover, Seagrass cover, Macroalgal canopy cover, and Coralligenous and other bioconcretions.

Discussion and conclusions

ESBH portal is unique with respect to other web GIS portals: it provides a standardised, centralised and free access point for numerous types of habitat spatial information at a European scale. The possibility to access all this different seabed-habitat information through the map viewer section of the portal, is a very interesting novelty for both technical and non-technical stakeholders. The construction of the composite maps, even if it does not improve the original quality of the datasets, provides the representation of the distribution of MKH and allows to demonstrate the added value of compiling and standardising seabed habitat maps and sample point data (Fig. 1). In this context, the scientific community has the opportunity to support this important initiative by sharing its seabed habitats maps and ground-truth points so that also the geographical distribution of the composite layers can be improved and updated, so as to facilitate the implementation of the mentioned action plans and support the conservation of endangered habitats.

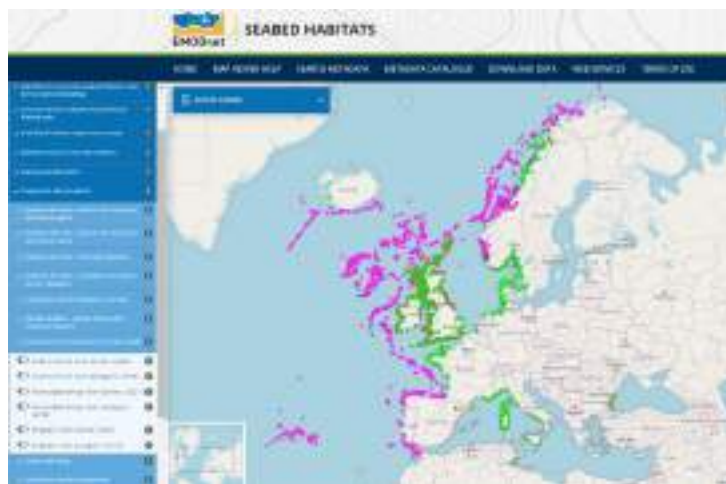


Fig. 1: EOV layers provided by the portal. Purple refers to live hard coral, green to seagrasses and brown to macroalgal

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Ali BADREDDINE

Tyre Coast Nature Reserve-Tyre-south Lebanon

E-mail: ali.badreddine@hotmail.com

THE CARLIT INDEX: AN EFFECTIVE AND USEFUL TOOL FOR ASSESSMENT OF THE ECOLOGICAL STATUS OF THE LEBANESE COAST, EASTERN MEDITERRANEAN SEA

Abstract

*The CARLIT index, based on the cartography of rocky-shore littoral communities and conceived in the European Water Framework Directive (WFD) for assessing the ecological status of coastal water bodies, was applied along the Lebanese coast during the two years 2020 and 2021. The results showed that the calculated ecological quality ratio values (EQR) varied from a max of 0.75 to a min of 0.12. Accordingly, the Ecological Status (ES) classes of 200 km of the Lebanese coastline ranged from good (with 26.6%) to moderate (46.6%), poor (0.2), and bad (6.6%) ES. In addition, the CARLIT index allows for estimating the status of macroalgae communities, especially Fucales (e.g., *Cystoseira* and *Sargassum* forests), representing more than 41% of the monitored Lebanese coast. Thus, the present study confirms the importance of the CARLIT index as a useful tool for monitoring the ecological status of the Lebanese coast.*

Key-Words: CARLIT, Ecological status, Macroalgae, *Cystoseira*, Lebanese coast

Introduction

Several methods or indexes based on macroalgae assemblages have been developed to assess rocky shores' Ecological Status (ES). Among them, the CARLIT index is conceived in the European Water Framework Directive (WFD) for evaluating the ecological status of coastal water bodies. The fact that rocky shores mainly characterize the Lebanese coastline makes the indexes based on macroalgal communities adequate for assessing the ES. The efficacy and the robustness of the CARLIT index for evaluating the Lebanese rocky coastline were confirmed in 2018 after its application along 164 km of the Lebanese rocky coastline (see Badreddine *et al.*, 2018). This study reports the results of the most recent application of the CARLIT index along the Lebanese rocky coastline.

Material and Methods

The study was performed from April-May 2020 and 2021 along 200 km of the Lebanese coast, representing about 91 % of the entire coastline. The Lebanese coast has been divided into 15 stretches of coast, considering their catchment basin and typology. The sampling consisted of a run of all the rocky coast by walking/ or using a small boat to assess littoral/and upper littoral communities and geomorphological factors of each sector. The data were visually recorded and noted directly into maps (scale 1:5000) previously prepared using aerial photographs taken from Google Earth®. Then, data were transferred into geo-referenced maps using (QGIS) software. The Ecological Quality Ratios (EQRs) were calculated based on the Reference Conditions of the Lebanese coast proposed in Badreddine *et al.*, 2018.

Results and Discussion

Based on the values of the EQR calculated in 2020 and 2021 (Fig. 1), four stretches of the Lebanese coast are in a good ES (Tyre/TYR, Adloun/ADL, Saksakiyeh/SAK, Enfeh/ENF); seven in a moderate (Saida/SAI, Tabarja/TAB, Jbeil/JBE, Barbara/BAR, Batroun/BAT, Chekaa/CHEK, and Qualamon/QUA, Sanini Island/SAN), three in a poor (Saadiyet/SAA, Tripoli/TRI, and Akkar/AKK) and one in a bad ES. The stretches of coast with a good ES are dominated by Fucales, mainly *Cystoseira* forests and *Sargassum*. In contrast, the moderate ones are dominated by ubiquitous photophilic algae, mainly Dictyotales, *Padina* spp., *Laurencia* complex. On the other hand, the poor ES rocky stretches are dominated by erect corallines and tolerant photophilic algae, mainly *Colpomenia*, *Hypnea*. Finally, the bad ES stretches are mainly dominated by green algae, mainly Ulvales, and Cyanobacteria. From a comparison point of view, the ES class has changed positively from 2018 to 2020 and 2021 (Tab. 1) due to the increase in EQR values in the monitored stretches of coast. This fact is likely due to a lower anthropogenic pressures along the Lebanese coastline during the two-year period 2020-2021 following the Covid pandemic. As a consequence, the diversity associated with the Lebanese rocky coast has been rehabilitated, and Fucales (mainly *Cystoseira* species) were present in more than 41% of the Lebanese rocky coast. From a monitoring point of view, the CARLIT index can be considered an important tool for evaluating the ecological status and estimating the condition of macroalgal communities, especially Fucales, along the Lebanese coast.

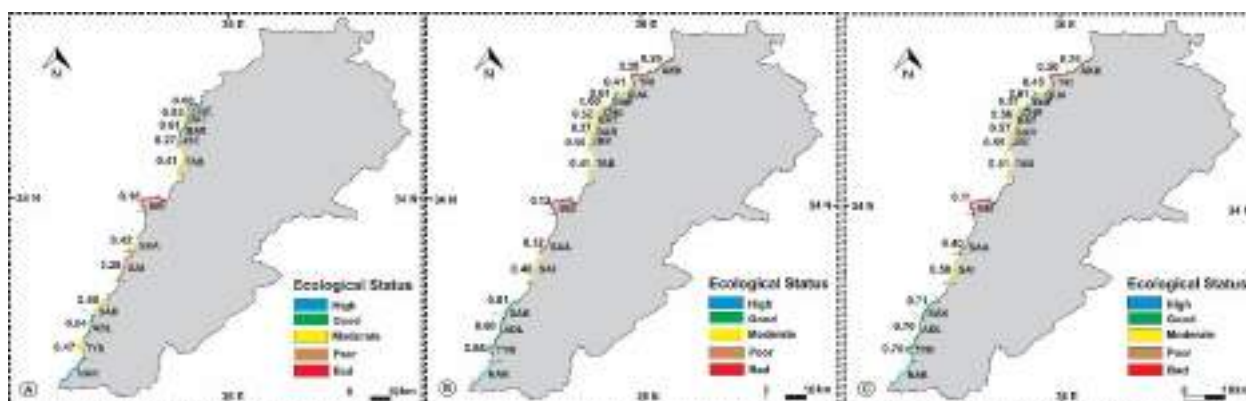


Fig. 1. Ecological quality Ratio (EQR) and Ecological status (ES) of the 15 studied stretches of coasts. EQR values calculated in: A) 2018 (Badreddine *et al.*, 2018), B) 2020 and C) 2021.

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Biodiversity, Ecology and Genome Laboratory, Faculty of Sciences, Mohammed V University, Rabat, Morocco.

E-mail: benhoussa@hotmail.com

ECOLOGICAL CHARACTERIZATION OF THE MARINE AREA OF THE FUTURE PROTECTED AREA OF JBEL MOUSSA (STRAIT OF GIBRALTAR, MOROCCO): CONSERVATION AND MANAGEMENT CONSIDERATIONS

Abstract

Marine field surveys (MedKeyHabitats I Project) revealed that the site of Jbel Moussa (Strait of Gibraltar, Morocco) has all the appropriate bioecological features to be designated as a marine and coastal protected area. Indeed, the site presents an excellent environmental state and a great ecological value linked to the presence of 25 species and seven habitats of conservation interest in the Mediterranean. All collected data were compiled and GIS-based maps of marine habitats, anthropic pressures, and tourist facilities were elaborated. Later on, within the framework of the MedMPA Network project, the GIS-based maps led to the development of an integrated management plan for the Jbel Moussa protected area, including a zoning plan with a clear definition of the related regulations in the different marine zones, in particular those with strict protection encompassing ecosystems of high conservation interest.

Key-words: Biodiversity, MPA, GIS-based maps, Conservation, Management.

Introduction

In accordance with the CBD Aichi target, Morocco plans to establish a comprehensive and coherent network of ecologically representative and well-managed protected areas. The site of Jbel Moussa (Strait of Gibraltar, Morocco) is designated as a potential Marine and Coastal Protected Area (MCPA). Here, we present an overview of a recent ecological characterization of the site and its integrated management plan.

Materials and methods

Multi-approach marine surveys (Diving, ROV, etc.) were conducted in 2014 and 2015 to appreciate the marine biodiversity of Jbel Moussa. All collected data were compiled and integrated in a Geographic Information System, allowing the elaboration of GIS-based maps of the marine habitats, anthropic pressures, and tourist facilities. An integrated management plan was elaborated then based on a participatory and inclusive approach.

Results

Jbel Moussa presents an excellent environmental state and a great ecological value linked to the presence of 25 species and seven habitats of conservation interest in the Mediterranean such as *Zostera marina* meadows and coralligenous communities (PNUE/PAM-CAR/ASP, 2016). The GIS-based maps led to the development of an integrated management plan for Jbel Moussa, including a zoning plan with a clear definition of the related regulations in the different marine zones (Fig. 1) (SPA/RAC -

ONU Environnement/PAM & HCEFLCD, 2019).

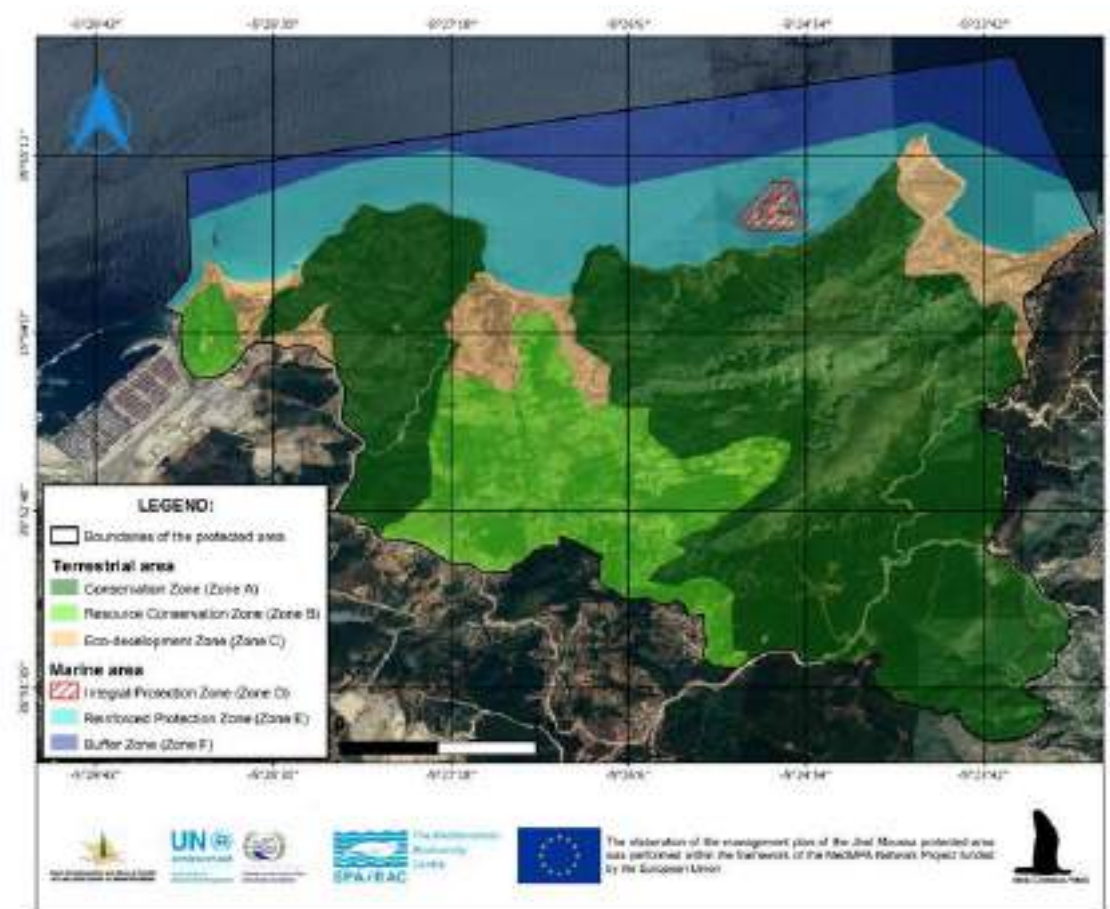


Fig. 1. Zoning plan of the potential MCPA of Jbel Moussa (SPA/RAC - ONU Environnement/PAM & HCEFLCD, 2019).

Discussion and conclusions

Jbel Moussa has all the appropriate bioecological features to be designated as an MCPA. It is one of the priority sites to be declared as an MCPA by Morocco which will enable reducing the shortage of MPAs along the southern Mediterranean coast.

Acknowledgements

This work was conducted respectively within the framework of the MedKeyHabitats I Project funded by MAVA Foundation and the EU-funded MedMPA Network Project, both implemented by UNEP/MAP-SPA/RAC in close collaboration with the Département des Eaux et Forêts (DEF).

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Ramla BOUHLEL, BENISMAIL M., CHEBAANE S., BENZARTI O., GRAMI B.
Association Notre Grand Bleu (NGB, NGO) Monastir-Tunisia;
Email: ramlabouhlel98@gmail.com

THE IMPACT OF CAULERPA CYLINDRACEA ON CYMODOCEA NODOSA IN THE MARINE AND COASTAL PROTECTED AREA OF KURIAT ISLANDS, TUNISIA

Abstract

Non-Indigenous Species (NIS) can be detrimental to a local ecosystem by overgrowing and competing with native species for space, food, and other resources. It can also affect the balance of the ecosystem by replacing these native species. However, the difficulty in making general predictions of which NIS will have negative effects suggests that each species must be subjected to rigorous ecological characterization and risk assessment of its introduction.

*The effect of the presence of the NIS algae *Caulerpa cylindracea* in the seagrass meadow *Cymodocea nodosa* was studied by comparing the biological and environmental parameters of two sites around small Kuriat Island. Environmental parameters, Chlorophyll-a content, organic content, granulometric sediment composition, macrophytes biometry and the taxonomic groups associated with *C. nodosa* were assessed for both sites. The results show the detrimental impact of *C. cylindracea* on the native biota of *C. nodosa* beds, probably through the reduction of light availability and limiting the photosynthetic performance of *Cymodocea*.*

Key-words: *Caulerpa cylindracea*, *Cymodocea nodosa*, Impact, NIS, Seagrass

Introduction

This study was done in the future Marine and Coastal Protected Area 'AMCP Kuriat' a Mediterranean area of high biological, ecological and biogeographic interest. AMCP Kuriat is characterized by the presence of a multitude of vulnerable species and marine habitats of conservation interest (e.g. the bivalve shell *Pinna nobilis*, the meadows of *Posidonia oceanica* and *Cymodocea nodosa*) (Aguir, 2009).

The present study is to: i) compare densities of the native *C. nodosa* meadows in the presence and absence of the NIS *Caulerpa cylindracea* and ii) analyze the morphological and biological parameters of *C. nodosa* and inference of potential effects of *C. cylindracea*.

Material and methods

The study was done in the small Kuriat Island, where the sampling was carried out in two different sites (the first site (T1) consisted of homogeneous beds of *C. nodosa*, and the second (T2) consisted of heterogeneous beds (*C. cylindracea* with *C. nodosa*) for each site, biotic and abiotic three samples were carried out in two different depths 0.62 m and 1.10 m (in total 12 samples).

Temperature, salinity, pH, dissolved oxygen and the conductivity were measured *in situ* with a multi-parameter. In addition, Chlorophyll a concentration was determined using the fluorimetric method proposed by Holm-Hansen *et al.*, 1965. Also samples of sediment were taken by polyvinyl chloride PVC corer of 10 cm diameter. On the top of this, density of *C. nodosa* was estimated *in situ* using a 30 cm square quadrat, randomly positioned in the herbarium (10 sampling point for each site) (Sghaier *et al.*, 2012).

Afterwards, at each site, twelve specimens of *C. nodosa* were taken haphazardly to study the associated macroinvertebrates and morphological analysis. For analyzing the data, Analysis of Variance (ANOVA) two-factors with replication was used to study the significant difference between the sites (T1, T2) for each set of samples. All the statistical analyses are carried out with the XLSTAT © software for Windows

Results and Discussion

The two studied sites were presented with characteristics of fine sand granulometry, which is a favourable habitat for the *Cymodocea nodosa* beds (Pergent *et al.*, 2012) and also for *Caulerpa cylindracea* (Verlaque *et al.*, 2000). The physicochemical parameters shows homogeneity between sites and depths ($P > 0,05$). The chlorophyll content significantly differed between the two sites at both sampling depths ($P < 0.0005$). The non-indigenous algae *C. cylindracea* seems to affect the concentration of Chlorophyll *a* as the site with this NIS has a low concentration in Chlorophyll *a* compared with the site with only the *C. nodosa* beds. The average leaf length in the *C. nodosa* was significantly different in the two depths of the same site and the two sites between them ($p < 0.05$). At shallow depth, the leaves are longer. At the site with the presence of *C. Cylindracea*, the leaves of *Cymodocea* are significantly shorter ($p < 0.05$).

Concerning the variation of density, and comparing the results found for the same depth between the two sites, a significant difference ($p < 0.01$) between the density of the *C. nodosa* in the two sites with a higher density in deeper waters. This negative effect of this NIS on the native species *C. nodosa* could be explained by establishing a competitive relationship in favour of the NIS with respect to resources (e.g. nutritive salts) (IFREMER, 2000).

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Université Côte d'Azur, CNRS, UMR 7035 ECOSEAS, Nice, France

E-mail: Simona.BUSSOTTI@univ-cotedazur.fr

KNOWLEDGE AND LEARNING PROCESSES IN MARINE ENVIRONMENTAL EDUCATION: A CASE STUDY ON *POSIDONIA OCEANICA* ACCUMULATIONS IN MEDITERRANEAN BEACHES

Abstract

*Environmental conservation objectives cannot be achieved without literacy on biodiversity and ecosystem functioning. This study thus aimed at assessing basic knowledge about the seagrass *Posidonia oceanica* and its ecological importance among pupils aged 11-16 years. We administered an ad hoc questionnaire in schools in 6 different Mediterranean countries to investigate pupils' perception about the litter accumulations of *P. oceanica* on the beaches (banquettes) that are usually perceived as a nuisance by users. In Nice (France), we re-tested with the same questionnaire pupils' knowledge acquisition after classroom lessons and outdoor activities. Results show that this method significantly supports learning processes about environmental issues.*

Key-words: Ocean literacy, teaching, seagrass, *banquettes*, seashore

Introduction

In spite of an increasing attention at societal level and the strong recommendation provided by the European Education Area and the EU Biodiversity Strategy, the issues related to biodiversity and natural ecosystems are just sporadically mentioned in science or civic education courses. To fully achieve the Sustainable Development Goals for 2030 (<https://education.ec.europa.eu>) and ensure citizen empowerment, these concepts should be learned from an early age (UNESCO, 2017). *Posidonia oceanica* is a seagrass endemic to the Mediterranean Sea included in Annex II of the Bern Convention as a species strictly protected. With the project ECOMED we aimed at setting up baseline information about knowledge and perception of the ecological importance of beach-cast *P. oceanica* seagrass litter (*banquettes*) by administering an *ad hoc* questionnaire among middle and high school pupils living in coastal areas of 6 Mediterranean countries. *Banquettes* are often removed from the beaches being perceived as a nuisance by users.

Materials and methods

We developed a questionnaire and translated it into different languages. The questionnaire included 29 questions about: (i) general knowledge on marine and beach ecology, (ii) perception and acceptance of *Posidonia* beach-cast. We distributed the questionnaire across schools of 6 Mediterranean countries (Albania, France, Italy, Montenegro, Spain, Tunisia) to students aged 11-16 years. A total of 3749 questionnaires were completed. Additionally, in some schools in Nice (France), the same questionnaire (n=245) has been re-administered a few months after to the same pupils in order to test for changes in knowledge and perception after classroom lessons and outdoor activities. Some field measures were directly collected by pupils through a scientific experimental protocol that specifically has been developed for this project.

Results

Our survey showed that: i) 62% of pupils already knew *Posidonia oceanica*; ii) >50% thought its accumulation (*banquettes*) are important for the environment; iii) only 49 % answered to accept to leave them in place. Interestingly, the experiment on learning in Nice showed a significant increase in the proportion of pupils who now agreed not to remove *banquettes* from the beaches (Fig. 1).

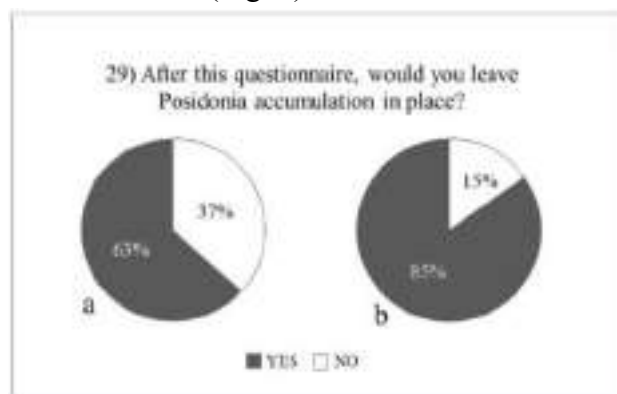


Fig.1: Pupil's responses in Nice at the question "After this questionnaire, would you leave *Posidonia* accumulation in place?", a) before and b) after classroom and outdoor activities.

Discussion and conclusions

These preliminary results show that middle and high school pupils inhabiting different Mediterranean countries and cities have a satisfactory level of basic knowledge (many had already heard the word *Posidonia oceanica*) even though further actions aimed at raising environmental awareness are recommended. Results also highlight that regular interventions involving pupils in lively science outdoor initiatives increase awareness and help to fill the wide lack in Ocean Literacy-related issues (see Cava et al., 2005). We also observed during the study period an important increase in teacher motivation. This successful interaction between teachers and researchers is of paramount importance considering the essential role that teachers have in preparing future citizen generations and the willingness of young people to know more about nature.

Acknowledgments

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Andreu DALMAU, SÁNCHEZ J.

SUBMON (Ortigosa Street, 14, 1st 3rd, 08003, Barcelona, Spain).

E-mail: andreudalmau@submon.org

RESTORING SEAGRASS MEADOWS FROM THE IMPACT OF LARGE-SIZED LITTER

Abstract

Litter is one of the main problems that the marine environment is facing and will have to face in the future. Normally, when people think about marine litter they focus on general waste and plastic pollution, forgetting about other types of debris that are lost or abandoned on the seafloor, potentially impacting sensitive habitats such as seagrass meadows. Abandoned mooring elements (concrete blocks, anchors, or chains), lost fishing gear or even sunken ships are some examples of large-sized heavy objects that accumulate underwater in coastal areas.

Here we present an initiative that we started in 2017. It consists of restoring seagrass meadows of the Spanish Mediterranean coast by locating and extracting heavy and large-sized objects that are impacting them. To do so, we elaborated a methodology that consists of: 1) selecting the area; 2) searching for this type of litter abandoned on seagrass meadows of the area and georeferencing it; 3) evaluating the suitability of the extraction by analyzing its integration in the meadows; 4) extracting the selected objects following the national regulations on commercial diving; 5) organizing an informative session addressed to public administrations, to the economic sectors implied and to the general population to raise awareness on the importance of seagrasses and to avoid further litter accumulations in the future. These actions have been carried out in different areas of the Spanish coastline, including marine Natura 2000 sites, Natural Parks, and Marine Reserves. Since 2017, a total of 469 objects have been removed from seagrass meadows, representing 56.890 kg of litter.

This initiative proves to be a way to effectively reduce some physical impacts on seagrass meadows, and an opportunity to raise awareness through society on the importance of preserving seagrasses as a key marine habitat.

Key-words: seagrass, restoration, litter, *Posidonia*, *Cymodocea*.

Introduction

It has been estimated that around 10 million tons of waste enter the ocean every year worldwide, being the 80% of them plastics and coming mostly from land-based sources (Jambeck *et al.*, 2015). This shows that litter is one of the main conservation problems that the marine environment is facing and will have to face in the future.

Marine litter can be defined as any type of human-made solid waste that enters into the marine environment. Thus, this concept is wider than what society normally thinks. There are many types of litter besides general waste and plastic pollution that can be found underwater in coastal areas, such as abandoned mooring elements (concrete blocks, anchors, or chains), lost fishing gear or even sunken ships. Those elements are normally large-sized and heavy objects that are lost or abandoned on the seafloor, where they can accumulate. Those accumulations can potentially impact sensitive underwater habitats such as seagrass meadows.

Here we present an initiative that our organization (SUBMON) started in 2017, that focuses on this type of litter. It consists of restoring seagrass meadows of the Spanish Mediterranean coast by removing heavy and large-sized objects that are impacting them.

Materials and methods

This initiative has been carried out following a methodology that we developed, which is divided in five phases:

- 1) Selecting the area. Actuations have been carried out in different protected areas of the Spanish Mediterranean coast with presence of seagrass meadows (Natural Parks, Natura 2000 sites and Marine Protected Areas). To decide where to act we gather information of potential areas and contact local administrations and organizations to know if there is a particular site where it would be needed to act. With this information we select the actuation area.
- 2) Searching for large-sized litter that is affecting seagrass meadows in the selected area. Once the area is selected, it is time to search for the litter. Members of our organization scuba dive through the area to find large-sized and heavy objects that are impacting seagrasses. All the objects found are photographed and georeferenced using a GPS to set their exact position.
- 3) Evaluating the suitability of the extraction by analyzing their integration into the meadows. Once the large-sized objects are found, it is time to evaluate if it is a better option to keep them underwater or to remove them. To do so, we analyze if they are integrated into the meadows, only removing the ones that can be extracted without producing an extra impact.
- 4) Extracting the selected objects. The removal of the selected objects is carried out following the national regulations on commercial diving, using specific gear and equipment such as diving helmets, lift bags and floating cranes. All removed objects are later brought into a certified waste management facility.
- 5) Organizing an informative session after the actuation. This session is addressed to public administrations, to the economic sectors implied and to the general population, in order to raise awareness on the importance of seagrasses and to avoid further litter accumulations in the future.

Results

Since 2017, we carried out this initiative at 16 different locations of the Spanish Mediterranean coast. A total of 469 objects have been extracted from *Posidonia oceanica* and *Cymodocea nodosa* meadows, representing 56.890 kg of litter removed. Also, 16 informative sessions have been organized, one at each location, to explain the actuations made and raise awareness throughout the local population and administrations.

Discussion and conclusions

The results obtained prove that this initiative is a way to: 1) effectively reduce some physical impacts that seagrass meadows are suffering in coastal areas; 2) raise awareness throughout society on the importance of preserving seagrasses and their role as a key marine habitat.

Acknowledgments

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Yasser ALI GENEID, EL-SAYED H., EL-HADDAD K., FOAD M.

National Institute of Oceanography and Fisheries, Egyptian Environmental Affairs Agency, EGYPT

E-mail: yasogin@yahoo.co.uk

MAPPING AND ASSESSING SEAGRASSES USING MULTI SATELLITE IMAGERY ALONG ALMAZA BAY, MEDITERRANEAN SEA, EGYPT

Abstract

*Seagrasses provide important ecosystem services, including provision of primary production supporting food webs, nutrient cycling, and habitat supporting marine biodiversity. They are also a major atmospheric carbon dioxide sink, helping mitigate global climate change. Preservation of seagrass habitats is intimately related to the sustainability of overall coastal ecosystems. The use of remote sensing in mapping underwater vegetation has increased recently with improvements in multispectral and hyperspectral sensors, enabling researchers to map and monitor the temporal changes in benthic habitats. The study was carried out along Almaza Bay, Egypt North coast (31°11'33.76"N, 27°34'21.61"E to 31°12'24.43"N, 27°32'0.97"E) on the Mediterranean seagrass *Posidonia oceanica* meadows. This area along with most of the Egypt Mediterranean seafront has witnessed huge development during the last 2 decades. Mega projects including touristic resorts, power plants and seawater desalination plants were established. Monitoring the extent and health of seagrass is important not only to track the impact of human activities on this valuable resource, but because seagrass acts as a sentinel, reflecting the condition of the surrounding environment. In this study, multispectral satellite images retrieved from both Landsat OLI and Sentinel 2 sensors were used to delineate the seagrass distribution. The obtained images were corrected for water depth, which helped in mapping process of the benthic habitats. The obtained results were validated using field observations to get the most accurate classification method. The images revealed dense *Posidonia oceanica* meadows to the west of the Bay in the subtidal zone, with water depth about 7-12 m and on a sandy substrate, while the east side of the Bay is characterized by a rocky substrate with almost no seagrass vegetation. The study showed the necessity of conducting more intensive research using multi-temporal satellite images in monitoring seagrass ecosystem change over time and space providing the basis for successful management strategies.*

Key-words: *Posidonia oceanica*, satellite imagery, benthic cover, Mediterranean Sea, Egypt

Introduction

Seagrass meadows provide important benefits to marine ecosystems. They stabilize the seafloor, protect the shorelines from erosion, and contribute to climate change mitigation thanks to their capacity to trap carbon dioxide. Additionally, seagrass meadows are widely considered as important feeding, nursery, and refuge habitats for fishes, invertebrates, and other animals (Nordlund *et al.*, 2018).

Mapping and monitoring of seagrass habitats in shallow coastal waters is performed by several methods. Remote sensing is one of the most effective means (Meehan *et al.*, 2005). Recent developments and improvements in multispectral remote sensing have increased the use of this method to map and monitor benthic habitats (Hedley *et al.*, 2018).

Materials and Methods

In the present study, Sentinel-2 Level 2A image acquired on 21 July, 2021 has been used. The images have been atmospherically corrected then the Bottom-Of-Atmosphere was created. A subset showing the study area was created and then Sen2Coral toolbox was used to apply a glint removal algorithm (Hedley *et al.*, 2005), in order to be able to observe the seafloor for the purpose of habitat mapping or bathymetry derivation. The obtained results were validated by field surveys *in situ* to get truthful classification scheme, then they were classified using iso-cluster unsupervised classification in order to get the different classes, which were compared to the field observations to generate the final benthic habitats map.

Results

The gained outcomes were repeated to get the most confirmed classification results. The images, as well as the field surveys, revealed dense *Posidonia oceanica* meadows along the western zone of the Bay in the subtidal zone, characterized by a sandy substrate with water depths of about 7-12 m. On the contrary, the eastern zone of the Bay was found to have rare seagrass vegetation, maybe due to its rocky substrate nature.

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Leila KTARI, CHEBIL AJJABI L.

Laboratoire B3Aqua, Institut National des Sciences et Technologies de la Mer (INSTM), Université de Carthage, Tunisie

E-mail: leila.ktari@instm.rnrt.tn

CASCADE APPROACH FOR VALORIZATION OF THE BROWN ALGA *DICTYOTA SPIRALIS* BY THE EXTRACTION OF BIOFERTILIZER AND BIOACTIVES COMPOUNDS

Abstract

The brown alga Dictyota spiralis, collected from Tunisian coasts has been analyzed for its biological properties in a cascading way of processing for biofertilizer and bioactive compounds extraction. Two extraction scenarios were followed in order to determine efficient way to maximize the use of biomass. Properties of the obtained fertilizers have been tested on the germination of Eucalyptus gomphocephala seeds. The results showed that extraction scenario could influence the recovery rate of bioactive compounds. Biofertilizers obtained from Dictyota spiralis showed significant stimulant activity on seed germination rate and precocity.

Key-words: Dictyotales, antioxidant, biofertilizer, fucoxanthin, phenolic compounds.

Introduction

Seaweeds have an important ecological role in maintaining environmental balance and the sustainability potential in term of economic for the “blue growth” strategy (Ktari *et al.*, 2021a). Dictyotales (Ochrophyta, Phaeophyceae) are common in warm and temperate waters. Species of this taxonomic group are known to produce a significant number of bioactive secondary metabolites (Ktari *et al.*, 2021b). Recently, Spiralyde A, an antikinoplastid dolabellane with strong activity against *Leishmania amazonensis* was isolated from *Dictyota spiralis* (Chiboub *et al.*, 2019). Furthermore, biostimulation proficiency on plant growth has been reported for different aqueous algal extracts resulting in positive effects on health, growth, and crop yield of many plants (HAMED *et al.*, 2018). The aim of this study was to evaluate the biological potential of *D. spiralis*, aqueous and organic extracts, while testing cascade extraction approach for efficient use of biomass.

Material and methods

Two scenarios were carried out: the first one being the production of biofertilizer directly from the alga (LAE) and extracting bioactives compounds from the extraction residue. While the second scenario involved extracting bioactive compounds directly from the algal biomass and preparing the biofertilizer from the organic extraction residues (LRE). Properties of the obtained fertilizers have been tested on the germination of *Eucalyptus gomphocephala* seeds *in-vitro* and *ex-vitro*. Bioactive properties of organic extracts were determined by quantifying high added value compounds such as pigments (Wang *et al.* 2005) and phenolic compounds with analysis of antioxidant properties through DPPH free radical scavenging assay (Hmani *et al.*, 2021). One-way ANOVA was performed, and significant differences ($p < 0.05$) were determined by Tukey's test.

Results

The results showed that no significant differences were obtained in pigment content of algal

biomass versus fertilizer extraction residue (Table 1). In contrast, while flavonoids and tannins contents were significantly higher when directly extracted from alga biomass, total phenolic content was found higher in the fertilizer extraction residue.

Tab. 1: Phytochemical content of *D. spiralis* organic extracts.

Extract	Pigments (mg/g dry biomass)			Phenolic compounds			DPPH
	Chl a	Chl c	Fx	TP*	F**	T*	IC ₅₀
Alga	0.34±0.02	0.22±0.03	0.13±0.05	13.3±1.55	62.71±4.87	47.06±2.45	64.02
Residue	0.36±0.0	0.30±0.04	0.11±0.0	19.7±0.72	39.1±1.37	19.73±1.15	76.04

Values are means ± standard deviation, *Total Polyphenol: TP (mg GAE/g extract); **Flavonoids: F (mg CatE/g extract); ***Tannins: T (mg CatE/g extract); IC₅₀ (mg/ml)

Considering the properties of obtained fertilizers from both extraction scenarios, no significant differences were observed on seed germination precocity or rate (Figure 1 a, b and c) except for high concentration (100%). The obtained biofertilizers showed significant stimulant activity on seed germination rate *in-vitro* and *ex-vitro*.

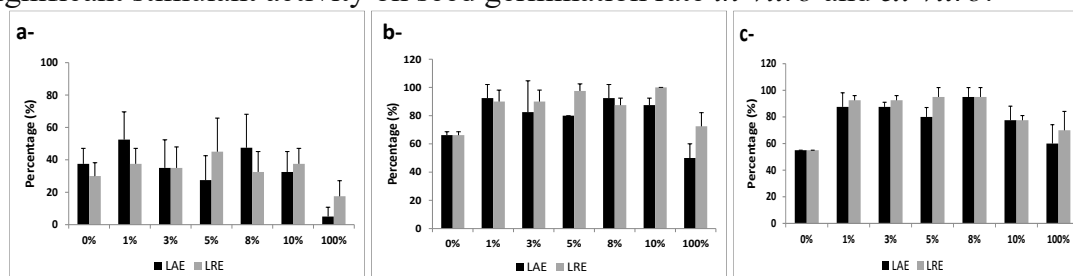


Fig. 1: Percentages of seed germination precocity (a) and rate (b *in-vitro*; c *ex-vitro*).

Discussion and conclusion

Results obtained for total polyphenol content were comparable to data found by Chiboub *et al.* (2017) for the same species (11.4±0.1 mg GAE/g extract) collected from northern coast of Tunisia (Tabarka). The analysis of phenolic compounds highlighted the richness of *D. spiralis* in flavonoids and tannins. However, the prior extraction of the liquid fertilizer leads to a reduction in the content of more than 30% for flavonoids and 50% for tannins.

This study showed that *Dictyota spiralis* represent an interesting source for biofertiliser and bioactive metabolites extraction. Therefore, cascade processing approach can be applied as an efficient way for biomass exploitation with less waste generated.

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Simone MODUGNO, PUSTINA I., KROMIDHA G.

INCA, Institute for Nature Conservation in Albania.

E-mail: info@ilbludelmare.it

RESULTS OF THE FIRST ASSESSMENT OF *POSIDONIA OCEANICA* MEADOWS IN PALASE AREA, ALBANIA

Abstract

Posidonia oceanica is a Mediterranean endemic phanerogam widely exploited as bio-indicator to assess both water quality and health status of coastal ecosystem, in accordance with the WFD and the IMAP Barcelona Convention. Main objectives of this work were to provide a baseline of the ecological conditions of *P. oceanica* meadow and the surrounding environment and mapping meadow distribution as a preliminary tool for studying Palasë's coast. Underwater scientific divers evaluated the meadow on the basis of UNEP/MAP-RAC/SPA guidelines. During survey physical and physiographical descriptors, meadow distribution in relation to the nature of the bottom and main habitat structural descriptors have been investigated. Results indicated good conditions of both the meadow and the marine environment, but a upper limit retreating phenomenon has been noticed.

Key-words: Posidonia, meadow, Water Framework Directive, IMAP, Underwater Scientific Diver.

Introduction

Studying *Posidonia oceanica* is fundamental for the evaluation of the ecological status of Mediterranean marine coastal ecosystems. In order to increase the protection of coastal areas, new updated data about marine phanerogams are really important. Mapping and census studies are required to update data year after year, in order to study any meadow's changes occurring.

The main objective of this work was to provide a first evaluation of the geomorphological, chemical-physical and ecological conditions of the Palasë 's meadow (Vlorë County, Albania). A first assessment of *P. oceanica* meadow along the coast of Palasë has been realized during summer 2021, in the frame of the project “Assessment of *P. oceanica* meadows in Palasë area” funded by the Global Environment Facility - Small Grants Program and RAC-SPA.

Materials and methods

Palasë's meadow has been studied and mapped on the basis of UNEP/MAP-RAC/SPA guidelines (UNEP/MAP-RAC/SPA, 2015). During survey the following descriptors have been investigated in five stations (St, Tab.1): depth of upper and lower limits (m), substrate type, bottom nature, % coverage of living *P. oceanica* and shoot density at the upper limit.

Results

The main results are reported and summarized below in Table 1.

Tab. 1: Synthesis of the main descriptors recorded in the investigated stations.

St	Upper limit (m)	Lower limit (m)	Substrate type	Bottom nature	% coverage of living <i>P. oceanica</i>	Density of leaf shoots (upper limit)	Assessment based on shoot density and depth (see UNEP/MAP-RAC/SPA, 2015)
1	-12,4	-27,4	sandy	a first line of shoreline consisting of coarse pebbles and smooth rocky boulders; a brief accumulation of gravel at the first bathymetric jump; from -4/4.5 m up to the upper limit (-12,4 m), a very long stretch of white sand with suspended mud and particles; initial presence of belts, stripes, island and many old channels and patches of dead <i>Posidonia</i> till the real meadow (-15/16 m)	50	450±12,75	moderate
2	-15,1	-29,3	sandy	a first line of shoreline consisting of coarse pebbles and smooth rocky boulders; a brief accumulation of gravel at the first bathymetric jump; from -4/4.5 m up to the upper limit (-15,1 m), a very long stretch of white sand with suspended mud and particles; initial presence of belts, stripes, island and many old channels and patches of dead <i>Posidonia</i> till the real meadow (-15/16 m)	46	522±25,88	high
3	-14,4	-32,2	sandy	a first line of shoreline consisting of coarse pebbles and smooth rocky boulders; a brief accumulation of gravel at the first bathymetric jump; from -4/4.5 m up to the upper limit (-14,4 m), a long stretch of coarse sand; initial presence of belts, stripes and patches of <i>Posidonia</i> till the real meadow (-15/16 m)	40	477±17,89	good
4	-12,6	-27,6	sandy	a first line of shoreline consisting of coarse pebbles and smooth rocky boulders; a brief accumulation of gravel at the first bathymetric jump; from -4/4.5 m up to the upper limit (-12,6 m), a long stretch of coarse sand; initial presence of belts, stripes, island, channels and patches of <i>Posidonia</i> till the real meadow (-13,2/14,5 m)	37	470±15,81	good
5	-12,5	-24,3	Rocky, mud-sandy	a first stretch of rocky coast consisting of limestone and granite cliffs and slabs with coarse pebbles and boulders accumulated up to -7/9 m; a brief accumulation of gravel between the rocks; from about -7,5/8,5 m up to the upper limit (-12,5 m), an expanse of sand and light mud; initial presence of bands, stripes and patches with dug corridors of <i>Posidonia</i> up to the real lawn -12.4/13.5 m)	25	470±38,08	good

Discussion

Results indicated good conditions of the ecological status of marine coastal environment and good condition of the meadow at the upper limit on the basis of the shoot density classes (UNEP/MAP-RAC/SPA, 2015), as summarized in Table 1. However, a phenomenon of retreat of the upper limit was detected, probably due to both hydrodynamic and anthropogenic impacts.

Although preliminary, these data can represent a reference baseline. The development of a new long-term and broader monitoring plan is recommended, in order to assess the temporal evolution of the ecological status of the meadow under increasingly intense anthropogenic pressures along the south coast of Albania.

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Briac MONNIER, PERGENT G., MATEO M.Á., CLABAUT P., PERGENT-MARTINI C.

Equipe Ecosystèmes Littoraux, UMR CNRS SPE 6134, Université de Corse, Faculté des Sciences et Techniques, Campus Grimaldi BP 52, 20250 Corte, France.

E-mail: briac.monnier@gmail.com

BLUE CARBON STOCKS ASSOCIATED WITH *POSIDONIA OCEANICA* MEADOWS: A LARGE-SCALE INVENTORY IN CORSICA (NW MEDITERRANEAN)

Abstract

*In the last decades, the increasing necessity to reduce atmospheric carbon dioxide (CO₂) concentrations has intensified interest in quantifying the capacity of coastal ecosystems to sequester carbon, referred to commonly as 'Blue Carbon' (BC). In the Mediterranean Sea, *Posidonia oceanica* seagrass meadows are considered as long-term carbon sinks due to the formation of a belowground organic deposit (the matte). To specify the role of *P. oceanica* meadows in climate change mitigation, an inventory of organic and inorganic carbon stocks has been conducted along the east coast of Corsica based on the study of 39 sediment cores. The biogeochemical analyses revealed a high variability of stocks with environmental drivers (water depth, depositional environment and sediment matrix). The carbon stocks in the first 250 cm of matte (average thickness) were estimated at 14.0 million t C (studied site) and 36.9 million t C (Corsica), corresponding to 29.2 and 76.8 years of CO₂ emissions from the population of Corsica.*

Key-words: *Posidonia oceanica*, seagrass, carbon stock, Corsica, climate change.

Introduction

The strategies to reduce greenhouse gas (GHG) emissions, and notably carbon dioxide (CO₂) concentration, are crucial for climate change mitigation. Recently, 'Blue Carbon' (BC) initiatives have been implemented to conserve and restore coastal vegetated ecosystems contributing to the capture of CO₂ and its storage as organic carbon in their sediments for long periods of time. Among these BC ecosystems, *Posidonia oceanica* seagrass meadows have been recognized as major carbon sinks due the formation of significant deposits (mattes) reaching several meters in height and preserving carbon over millennia. In the last decades, the increasing necessity of reducing CO₂ concentrations has intensified interest in quantifying the carbon storage and its variability related with biological and environmental drivers (Mazarrasa *et al.*, 2018). This study aimed (i) to understand the variability of carbon stocks in the *P. oceanica* matte of Corsica, and (ii) to provide a global estimate carbon stored at regional scale.

Material and methods

Posidonia oceanica matte cores (n = 39) were collected along the east coast of Corsica (France) in a wide range of environmental conditions (depth: -10 to -40 m; depositional environment: coastal vs estuary; substrate: sandy vs rocky). The sediment sampling was performed in August 2018 using Kullenberg gravity corer during the oceanographic survey Carbonsink aboard the R/V 'L'Europe' (Ifremer). Cores were subsampled in 1 cm-thick slices, dried and weighted to measure the dry-bulk density (g cm⁻³). Biogeochemical

analyses (loss on ignition, elemental and isotopic measures) were achieved on grounded samples to determine the organic (%C_{org}) and inorganic carbon (%C_{inorg}) contents. The C_{org} and C_{inorg} stocks were normalized to stratigraphic depths of 100 cm (standard) and 250 cm thick deposits (mean thickness of matte; Monnier *et al.*, 2021).

Results and discussion

The mean (\pm SE) C_{org} and C_{inorg} stocks show a significant variability with depth, sediment matrix and depositional environment (Fig. 1). Seagrass meadows located in shallow stations and settled on sandy substrate exhibited significantly higher C_{org} stocks and lower C_{inorg} stocks than those located in deeper areas or growing on rocky matrix. In the top 100 cm of matte, the stocks have been estimated at 32.7 ± 1.5 kg C_{org} m⁻² and 24.5 ± 4.5 kg C_{inorg} m⁻². Considering the mean thickness of matte on the east of Corsica, the C_{org} and C_{inorg} stocks reached 72.3 ± 4.6 kg m⁻² and 60.5 ± 9.9 kg m⁻², respectively (Fig. 1).

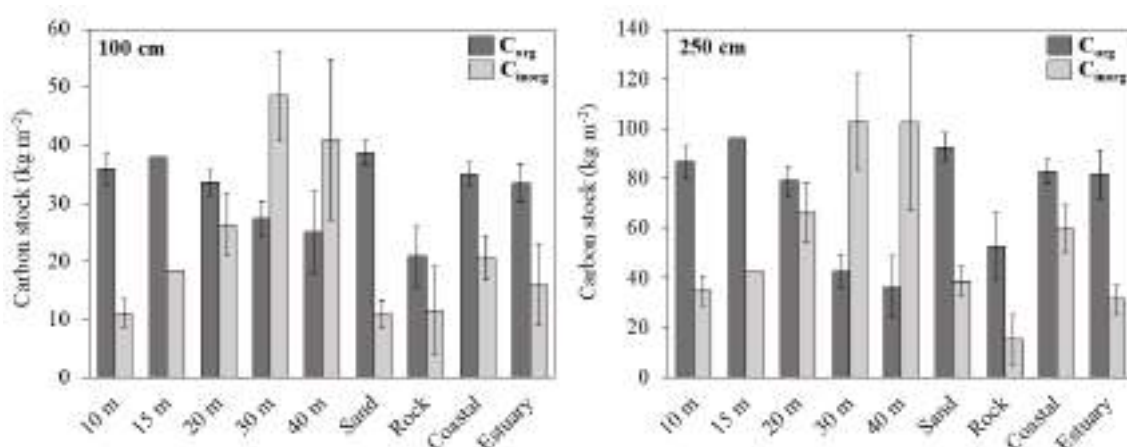


Fig. 1: Mean (\pm S.E.) C_{org} and C_{inorg} stocks in the top 100 cm (left) and 250 cm (right) of *P. oceanica* matte determined at different depths, sediment matrix (sandy or rocky) and environmental influence (coastal or estuary).

The estimation of C_{org} stocks in the top 250 cm of matte at studied site and along Corsica Island have been assessed at 14.0 million t C_{org} (51.5 million t CO_{2e}) and 36.9 million t C_{org} (135.4 million t CO_{2e}) that is to say the equivalent to 29.2 years and 76.8 years of CO₂ emissions by the entire Corsican population, constituting one of the most significant estimates of C_{org} stocks associated with *P. oceanica* meadows in the Mediterranean basin.

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Pedro Clemente NAVARRO-MARTÍNEZ, BERNARDEAU-ESTELLER J., BELANDO M.D., MARÍN-GUIRAO L., MARTÍNEZ-SALVADOR A., CONESA-GARCÍA C., MARIN-GARCÍA D., AGULLES-GÁMEZ M., RUIZ-FERNÁNDEZ J.M.
Department of Ecology, University of Murcia, Spain.
E-mail: pedroclemente.navarro@um.es

THE RECENT INFLUENCE OF CLIMATE CHANGE RELATED TO EXTREME EVENTS ON PRODUCTION DYNAMICS OF *POSIDONIA OCEANICA* (L.) DELILE MEADOWS

Abstract

*In semi-arid areas of the Mediterranean region, climate change is increasing the incidence of marine heatwaves, torrential rains and big storms. In this study, the influence of these extreme events on the vertical rhizome production (annual growth and biomass) of the iconic seagrass *Posidonia oceanica* was explored by lepidochronological modeling using generalised additive models (GAMs). Extreme temperature events had a negative effect on rhizome biomass production and vertical annual growth. This effect was intensified by high rates of terrigenous sedimentation linked to wadi discharges under torrential rains. Therefore, meadows close to wadis show a higher vulnerability to extreme events related to climate change in semi-arid areas. In this context, lepidochronological techniques and environmental approaches used can be useful tools for long-term studies of the production dynamics of this species and the effects associated with extreme climatic and weather events.*

Key-words: Seagrasses, heatwaves, terrestrial sediment inputs; lepidochronology, GAMs

Introduction

Climate change is progressively increasing the incidence of climate events such as heatwaves, torrential rains and big storms. In the Mediterranean region, these extreme events may negatively affect the endemic seagrass *Posidonia oceanica* (Giorgi & Lionello, 2008). In these areas, during torrential rains, wadis transport terrestrial sediments to the sea, where they can interact with marine heatwaves (MHW) and storms (Marbà & Duarte, 1997). Noteworthy, the combined effect of extreme events on *P. oceanica* seagrass meadows has been little studied. The aim of this study was to explore the influence of these extreme events on the production of *P. oceanica* rhizomes in several meadows of a semiarid Mediterranean coastal area (Region of Murcia, Southern Spain)

Materials and methods

Time series of vertical annual growth and biomass production of *P. oceanica* rhizomes were obtained by lepidochronology (tool for the analysis of seagrass dynamics based on the estimation of the age of leaf sheaths). Terrestrial sediment inputs were estimated by Watershed Erosion Prediction Project - WEPP models, sea storms were characterised by mean energy flux of waves, and marine heatwaves were disaggregated into standardised metrics. The potential influence of different descriptors of extreme events on the interannual variation of *P. oceanica* rhizome production variables was analysed using generalised additive models (GAMs).

Results

The models developed showed a negative relationship of annual maximum temperatures and the intensity of extreme events (maximum temperatures reached during an extreme event) with the rhizome production capacity. Marine heatwaves above 28°C had a clear negative effect on both biomass production and vertical annual growth. Also, important evidence of a potential interaction between heatwave intensity and terrestrial inputs associated with torrential rainfall was detected. Negative impact was further intensified by high rates of terrigenous sedimentation (Fig. 1).

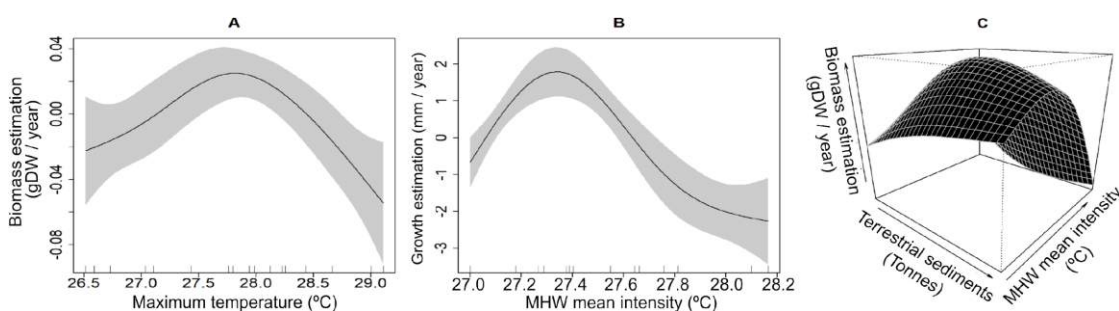


Fig. 1: Alternative model estimates for vertical rhizome production of *P. oceanica* at different sampling stations. We show the effect of maximum temperatures (A), MHW mean intensity (B), and the interaction (C) between terrestrial sediment discharges by La Azohía wadi (Region of Murcia, Southern Spain) and MHW mean intensity.

Discussion and conclusions

Extreme temperature events had a negative effect on rhizome biomass production and vertical annual growth. This effect was intensified by high rates of terrigenous sedimentation linked to wadi discharges under torrential rains. Therefore, meadows close to wadis show a higher vulnerability to extreme events related to climate change in semi-arid areas. In this context, lepidochronological techniques and environmental approaches used can be useful tools for long-term studies of the production dynamics of this species and the effects associated with extreme climatic and weather events.

Acknowledgements

This research has been funded by the projects "*Climate change and morphological adjustments in Mediterranean ephemeral watercourses: dynamics and geomorphic resilience, and proposals for action (CCAMICEM)*" funded by the Ministry of Science, Innovation and Universities of the Spanish Government and the European Union (ERDF) and the *Monitoring network of the meadows of *P. oceanica* of the Region of Murcia (Spain)* funded by the Autonomous Government. PCNM was supported by a PhD grant from the University of Murcia (Spain).

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Atik RAJA, KHAYA K., BELATTMANIA Z., SABOUR B.

Phycology, Blue Biodiversity and Biotechnology RU, Laboratory of Plant Biotechnology, Ecology and Ecosystem Valorization- CNRST, Research Unit N°10, Faculty of Sciences, University Chouaib Doukkali, El Jadida, Morocco.

E-mail: atic.raja@gmail.com

SEASONAL VARIABILITY OF PHENOLOGY OF *ERICARIA MEDITERRANEA* (FUCALES, OCHROPHYTA) FROM THE MEDITERRANEAN COAST OF MOROCCO

Abstract

In the Mediterranean Sea, macroalgal forests are dominated by fuclean brown algae (i.e., Cystoseira sensu latu, including the genera Cystoseira, Ericaria and Gongolaria) spreading out from the intertidal to the subtidal zone with different species along the bathymetric gradient. The present study investigated the morphological and reproductive phenology of Ericaria mediterranea from Mediterranean coast of Morocco. Density, size, and fertility of this species were measured monthly from January to December 2021. The E. mediterranea density reached values of cover ranging from 50.4 ind/m² in autumn to 60 ind/m² in spring. The thalli showed a seasonal pattern characterized by a marked phenological variation of the fronds. The monthly average values of thallus size vary from a minimum of 10.9 cm in January to a maximum of 20 cm in June. The maximum vegetative development was observed in spring/summer, in accordance with high temperature and photoperiod length. The E. mediterranea fertility progressively increases from summer to early autumn, reaching the highest value (56% fertile branches) in July. Matured receptacles can be observed during the August- November period.

Key-words: Phenology, *Cystoseira*, *Ericaria mediterranea*, Mediterranean, Morocco.

Introduction

Most of Mediterranean coasts are dominated by macroalgal forests (Assis *et al.*, 2020). The majority of these forests are fuclean brown algae (i.e., *Cystoseira sensu latu*, including the genera, *Cystoseira*, *Ericaria*, and *Gongolaria* (Molinari-Novoa and Guiry, 2020). Our study monitors the morphological and reproductive phenology of *Ericaria mediterranea* from Mediterranean coast of Morocco, for one year.

Materials and methods

The study was carried out at the Cape of Three Forks (C3F), a headland on the Mediterranean coast of northeastern Morocco (35°25'14.6"N; 2°57'21.7"W), classified as SIBE of priority 2 since 1991. The biodiversity of the C3F, both terrestrial and marine, is so rich (UICN, CAR/ASP, HCEFLCD, 2014).

Density of *E. mediterranea* is determined by estimation of the average number of fronds per m² after counting the fronds present in 10 plots of 625 cm², each delimited by a quadrat of 25 cm side. The plots are randomly selected along a 10 m long transect. The counting is made at low tide, monthly, during the year 2021.

Maturity and Fertility are assessed monthly during a year according the AFRIMED project protocol for *Cystoseira* phenology.

In the field, Fertility, expressed as the percentage of plants bearing receptacles (fertile), is determined from observations of 50 individuals randomly sampled. The thallus is considered fertile when the percentage of fertile branch estimated visually is greater than 25 % (by direct visual estimation).

In the laboratory, 5 individuals randomly sampled monthly during the year 2021 were used to study maturity. For each individual, the receptacles of 5 branches are incised transversely under a microscope and the gametes inside the conceptacles are counted and measured. Individuals are considered mature when the gametes are fully developed and close to release. Maturity is expressed as the percentage of mature individuals.

Results

The *E. mediterranea* density reached values of cover ranging from 50.4 ± 6.7 ind/m² in autumn to 60.0 ± 6.7 ind/m² in spring. The thalli showed a seasonal pattern characterized by a marked phenological variation of the fronds. The monthly average values of thallus size vary from a minimum of 10.9 ± 3.3 cm in January to a maximum of 20 ± 3.3 cm in June. The maximum vegetative development was observed in spring and summer. The *E. mediterranea* fertility is relatively high in summer and autumn ($54\% \pm 0.3$ and $51\% \pm 0.3$, respectively) with a peak percentage fertility of 56% reached in July. Matured receptacles can be observed from August and may persist until early winter.

Conclusion

This first study in the Cape of Three Forks showed that *E. mediterranea* is in good health unlike what has been observed on the French Mediterranean coasts where Thibaut *et al.* (2005) reported that *E. mediterranea* (as *Cystoseira mediterranea*) populations were regressing and has almost disappeared from some area with, however, persistence as a continuous belt along the Albères coast.

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ROUNDTABLE

**Christine PERGENT-MARTINI, BOUAFIF C., BURZIO P., RAÏS C., TORCHIA G.,
MAGANZA A., BEZZO T., SOHLOBJI E., VILLERS F.**

EqEL, UMR CNRS SPE 6134, Université de Corse, Campus Grimaldi, 20250 Corte,
France.

E-mail: pmartini@univ-corse.fr

ANCHORING ON *POSIDONIA OCEANICA* MEADOWS: SYNTHESIS ON THE EXISTING REGULATIONS AND DISTRIBUTION MAPS AS A BENCHMARK FOR ACTION IN THE MEDITERRANEAN

Abstract:

Posidonia meadows are widely distributed throughout almost the whole Mediterranean Sea, from the shoreline to 20-45 m of depth and play a key-role in terms of functioning of the marine environment. They provide important ecosystem services and also act as a carbon sink with a highly efficient carbon storage capacity. However, due to their location and ecological needs, Posidonia meadows are subjected to many anthropic pressures. Among these, anchoring is reported as a major and increasing threat, particularly in recent years, due to the significant increase in leisure boating. Following the conference "Anchors Away", organized by the European Union in 2019, the Mediterranean Posidonia Network (MPN) was created to address this issue at the whole Mediterranean scale by gathering together all the Mediterranean Countries concerned by the presence of Posidonia oceanica in their waters. In this context, a study was launched to collect the available information regarding (i) the known distribution of Posidonia oceanica meadows in GIS, (ii) its overlapping with protected areas (MAPAMED), and (iii) an exhaustive review of the regulatory framework protecting the species, in particular from anchoring. Relevant experts, scientists, academics and/or NGOs of each one of the Countries facing the Mediterranean Sea and having Posidonia meadows were involved in such study.

The results show that, even if some progress has been made (mapping coverage or legal protection of Posidonia), only few countries have promulgated regulations allowing for the specific control of anchoring on Posidonia meadows. Further efforts are needed to have a more comprehensive inventory of the meadows reflecting their distribution in all the basin, and allows for identifying priority areas where measures are most needed for the control and management of boat anchoring activities. In addition, the findings of such studies created a comprehensive state of the art potentially acting as a benchmark for common measures.

Key-words: Posidonia; legal protection; anchoring regulation.

Introduction

The meadows formed by *Posidonia oceanica* are considered a key habitat of the Mediterranean Sea where they are widely distributed from the shoreline to 20-45 m and host approximately 20 % of the species known for the Mediterranean Sea (Boudouresque *et al.*, 2012). They provide important ecosystem services such as the protection of the shores against erosion and their role of a carbon sink with a highly efficient carbon storage capacity.

The *Posidonia oceanica* meadows are subjected to multiple threats due to the strong anthropic pressure that characterize the Mediterranean. Above all, anchoring has been historically reported as a major threat to Posidonia meadows with increasing impacts particularly in recent years, due to the significant increase in leisure boating.

Following the conference "Anchors Away", organized by the European Union in 2019, the Mediterranean Posidonia Network (MPN) was created to address this issue at the whole Mediterranean scale by gathering all the Mediterranean Countries concerned by the presence of *Posidonia oceanica* in their waters. One of the priorities of the network being to support the strengthening of regulations related to anchoring, including by (i) ensuring consistency among relevant national regulations and (ii) promoting compliance with these regulations through monitoring, and (iii) evaluating the effects of their measures on the preservation of Posidonia meadows.

Within the framework of its activities, the MPN launched a study aimed at collecting the available information regarding the known distribution of *Posidonia oceanica* meadows available in GIS format and its overlapping with protected areas. The study also included a review of the regulatory framework protecting the species and/or its meadows from anchoring.

Materials and methods

The existing data on the status of Posidonia meadows in the Mediterranean Sea and the regulation tools adopted in each country were gathered through an in-depth desktop study and consultations with relevant experts, scientists, academics and/or NGOs of each one of the countries facing the Mediterranean Sea and having Posidonia meadows. To this end, standard data forms were prepared to be filled in with the information gathered by both a bibliographic research and consultations in order to have comparable data concerning the protection of *Posidonia oceanica*, its monitoring and the management measures implemented to safeguard the meadows from anchoring. Particular attention was paid to the existing information in terms of digital maps of Posidonia meadows. All countries facing the Mediterranean were considered, except the following ones where *P. oceanica* was found to be absent (Telesca *et al.*, 2015): Bosnia and Herzegovina; Israel; Lebanon, Morocco and Syria. An additional form targeting Marine Protected Area (MPA) managers was circulated to gather additional data (if any) and, in particular to address the implementation of measures to prevent the impacts of anchoring on Posidonia (e.g. eco-mooring).

Using ESRI ArcGIS©, a geodatabase was prepared for Posidonia distribution building on the data of Telesca *et al.* (2015) that was completed and/or updated with available and downloadable data on geoportals retrieved during filling in the forms (e.g. Ligurian Region in Italy and Corsica in France).

When projection errors occurred in the georeferenced data consulted, a correction was implemented after a careful comparison with other geodatabases of seagrass distribution (e.g. UNEP-WCMC & Short, 2021) and analysis of the shoreline and bathymetry.

Some Countries (i.e. Egypt and Montenegro) were not considered in the estimation of the Posidonia coverage, since only punctual data are available in the consulted databases.

The Posidonia meadow distribution in the basin was then overlapped with the most updated MPA distribution data provided by MAPAMED (the database of Marine Protected Areas in the Mediterranean) to assess the extent of Posidonia meadows protected by nationally regulated institutions, such as the nationally designated MPAs, SPAMIs. Natura 2000 sites and Ramsar sites were also included in the calculations but separated from the other protected areas.

For each Country facing the Mediterranean (and having Posidonia meadows in its territories) a standard factsheet was prepared in order to provide all the regulatory data gathered for the Country itself and focused on Posidonia safeguard, in particular from anchoring (when applicable), and the extent of Posidonia in its territories.

Results

According to the available knowledge, in September 2021, 16 Countries facing the Mediterranean Sea out of 21, have *Posidonia* in their territorial waters. Only six Countries (Croatia, France, Greece, Italy, Malta, Spain) have a cartography at national (or at local/department) level, of the distribution of their *Posidonia* meadows, available online. The percentage of *Posidonia* meadows mapped can be estimated to 100 % only for seven Countries (Tab. 1). For each country a part of the *Posidonia* meadows are included in national Marine Protected Areas, Natura 2000 site or Ramsar site (between 2 to 99 %), that constitutes a mean to safeguard these meadows against some human activities like anchoring. This value can be taking into account only for the Countries where all the coastlines have been mapped (Tab. 1).

Tab. 1: Surface of the *Posidonia* meadows included in a MPA, or Natura 2000 or Ramsar sites and percentage of the whole *Posidonia* meadows protected by this way, for the different countries. Al: Albania; Fr: France; It: Italy; Mo: Monaco; Sl: Slovenia; Sp: Spain.

	Albania	France	Italy	Malta	Monaco	Slovenia	Spain
Surf. (ha)	210	68 716	131 801	5 280	13	0.5	101 618
% of <i>Posidonia</i> included in a protected site	5	78	41	92	99	72	75

All the Countries, concerned by *Posidonia* meadows have at least ratified the Barcelona convention and its Protocol concerning the specially Protected Areas and the biological Diversity, which mentions *Posidonia oceanica* as an endangered species and most of them have signed other international commitments (Tab. 2). 69% of these countries have national laws that allow protecting directly the species or its meadows or indirectly by: (i) forbidding some activities above the *Posidonia* meadows, (ii) taking into account the presence of *Posidonia* meadows in the implementation of the procedures of environmental impact assessment and (iii) banning trawling and other fishing gears that could damage the meadows...; Tab. 2). But on the contrary few Countries already have specific laws to protect the meadows against anchoring (Tab.2).

Tab. 2: Synthesis of the rules and regulatory framework protecting *Posidonia* meadows, with a focus on anchoring. Y: existing law or regulation.

	International Commitments specifically listing <i>Posidonia</i>			National laws for <i>Posidonia</i> protection		Specific national laws for <i>Posidonia</i> protection from anchoring
	Barcelona Conv.	Bern Conv.	Habitat Direct.	Protected Spec.	Indirect Protect.	
Albania	Y	Y	Y		Y	None
Algeria	Y			Y		None
Croatia	Y	Y	Y	Y		Y/ Anchoring and mooring outside planned zones prohibited in protected areas
Cyprus	Y	Y	Y			Y/ Through national Fisheries Legislation, MPAs are designated with prohibitions on fisheries and vessel passage and anchorage
Egypt	Y					None on the Mediterranean part

	International Commitments specifically listing Posidonia			National laws for Posidonia protection		Specific national laws for Posidonia protection from anchoring
France	Y	Y	Y	Y		Y/ Anchoring prohibited on protected vegetal species. In several areas, anchoring prohibited for boat > 24m length between the surface and - 40 m
Greece	Y	Y	Y	Y		None
Italy	Y	Y	Y			None
Libya	Y					None
Malta	Y	Y	Y	Y		None
Monaco	Y	Y		Y		Y/Identification of areas of anchoring restriction; most of them overlap with Posidonia meadows
Montenegro	Y	Y	Y	Y		None
Slovenia	Y	Y	Y			None, anchoring not possible on Posidonia meadows
Spain	Y	Y	Y	Y		Y/ Anchoring prohibited in the Balearic Islands
Tunisia	Y	Y			Y	None
Turkey	Y	Y	Y	Y		Y/ Anchoring and mooring prohibited outside the port administrative site except for purpose of travel and sport

This lack of regulation does not imply that these Countries are not concerned by this issue, and practical solutions have been implemented like eco-mooring, public information, uses of web application to identify area free of Posidonia to anchor (Tab. 3). Concerning eco-mooring several countries mentioned the complex administrative steps as one of the main difficulties faced to implement these initiatives and some of them were not even finalised because of these administrative constraints. Webapps and smartphone apps (e.g. Donia) were also developed for some Countries (e.g. Spain, France, Greece and some places in Italy) to assist sea users in finding safe anchoring places outside fragile ecosystems, such as Posidonia meadows.

Tab. 3: Synthesis of the solutions and monitoring plans implemented in the different Countries concerning the anchoring on Posidonia meadows

	Adopted solutions, case studies or guidelines for managing anchoring	Monitoring plans for anchoring on Posidonia
Albania	34 anchoring buoys in Vlora bay and Karaburun peninsula	None
Algeria	None	None
Croatia	“Special purpose ports – anchorages” in Split-Dalmatia County – “Sea and Karst”. Mooring buoys system and electronic ticketing for Lastovo Island Nature Park.	Medpan South Croatia Pilot Project (2012). National Park Kornati, monitoring in two anchoring sites (Project - SASPAS - Italia-Croatia)
	Adopted solutions, case studies or guidelines for managing anchoring	Monitoring plans for anchoring on Posidonia
Cyprus	Mooring buoys equipped with light at Kavos Gkreko (Konnos).	None
Egypt	Mooring buoys installed to safeguard coral reefs in the Red Sea	None

	Adopted solutions, case studies or guidelines for managing anchoring	Monitoring plans for anchoring on Posidonia
France	Mooring strategy (2019) established and implemented with eco-moorings, limitedly affecting the substrate in several departments and mooring boxes for large yachting. 32 areas equipped with eco-mooring system and 11 planned. RAMOGE Area- Measures for raising awareness (i.e. flyers) on the safeguard of Posidonia from anchoring within the framework of the RAMOGE agreement (France/ Monaco/ Italy) Availability of the Donia application free to allow boaters to know where are Posidonia meadows before anchoring. Sensibilisation of boaters with specific information campaigns (eco-geste)	Environmental monitoring required in mooring areas and light equipment
Greece	Installation of sea grass friendly mooring buoys in the area of Andros Island. Installation of ecological anchoring buoys and prohibition of free anchoring in the National Marine Park of Zakynthos	Monitoring plan in the Aegean and Ionian Seas and within the frame of the CYCLADES LIFE Project
Italy	“Spiorsi” seagrass-friendly mooring system (Punta Chiappa - Portofino MPA). Mooring buoys (Punta Manara -Riva Trigoso; Campulungu, Cavoli Island and Molentis cape - Sardinia) RAMOGE Area- see France	Monitoring project study on mooring systems in Sardinia (Acunto <i>et al.</i> 2017). Monitoring project study on anchoring in Liguria (Montefalcone <i>et al.</i> , 2013)
Libya	None	None
Malta	None	None
Monaco	Mooring on Posidonia meadows banned in the MPA “Larvotto, Reserve”. RAMOGE Area- see France	None
Montenegro	Seasonal mooring buoys in Katič potential MPA screwed into sand and using ballasts. Anchoring prohibited in the Platamuni MPA.	Monitoring of the effects of the mooring buoys in in Katič potential MPA is ongoing.
Slovenia	Anchoring regulated in MPAs.	None
Spain	Anchoring prohibited and eco-moorings installed in the Balearic Islands. Ecological anchors (buoys) in the natural space Castell-Cap Roig in Palamós (Catalogna). In Serra Gelada Natural Park, the drafting of a decree (similar to that of the Balearic Government) is ongoing. 29 ecological anchoring points and a boat-based surveillance service that sanctions anchoring in areas prohibited. Implementation of new tool as “Spatial Monitoring”. Webapps (GARMIN and NAVIONICS) and smartphone apps (Donia) reporting meadows distribution and buffers.	None
Tunisia	Ecological mooring system in the future Kuriat Archipelago MPA.	None
Turkey	Ecological mooring system in Göcek and Dalaman bays.	None

Conclusion & perspectives

The protection of Posidonia meadows against boat anchoring in the Mediterranean is far from being satisfactory since it is effective in only a limited number of Countries. Indeed, although all the Countries with *Posidonia oceanica*, have regulations concerning the species protection in general, only few have promulgated regulations allowing the

specific control of anchoring on *Posidonia* meadows. In addition to the actual enforcement of specific regulations, some technical tools have been taken in consideration and proved to be effective. In this context, eco-mooring systems have been set up in some areas, especially in marine protected areas. Webapps and smartphone apps constitute an easy mitigation approach, but require information on the distribution of the habitats, at least in the areas submitted to anchoring pressures. However, the cartography of the distribution of *Posidonia* meadows, when present and available, is mostly at local level or based on distribution modelling (e.g. Croatia). For most of the European Union Countries, where significant progress has been done during the last decade.

Further investigation is required to ensure an adequate mapping coverage with the view of providing the relevant decision makers, and other stakeholders, with accurate data (maps) allowing them (i) to have an inventory of meadows that reflects their distribution in all the Mediterranean basin and (ii) to identify areas where measures are most needed for the control and management of boat anchoring activity and other pressures. This may be considered as an urgent priority for most Mediterranean Countries in relation to *Posidonia oceanica* conservation in general and to mitigate the impacts of anchoring on the meadow.

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CONCLUSIVE REMARKS AND RECOMMENDATIONS OF THE 7TH MEDITERRANEAN SYMPOSIUM ON MARINE VEGETATION

1. Posidonia meadows are at the top of the ecosystems in terms of Carbon fixation and sequestration in their matte. Considering the importance of the ecosystem services provided by seagrass meadows (including carbon sequestration), a particular attention should be paid on their conservation and restoration.
2. The scientific community has underlined the general regression of *Posidonia oceanica* meadows, mainly due to trawling, anchoring, and coastal development; although some local signs of natural recovery have been observed where effective protection measures have been undertaken. The same situation of decline can be observed also for macroalgae. This evidence urgently asks for the need to strengthen efforts to further reinforce the protection of key habitats by reducing pressures and by boosting the creation of MPAs and No Take zones, and to develop monitoring activities following the IMAP guidance of the Barcelona Convention Ecosystem Approach. Thus, stricter and more binding measures are necessary for the protection of *Posidonia oceanica* meadows, especially against bottom trawling and anchoring (e.g., through the improvement of ecological moorings), and these measures should be considered in the update of the regional action plan on marine vegetation.
3. We need to reinforce the exchange of the best practices and to improve the mapping effort of Posidonia meadows at the whole basin scale, also taking advantage from the availability of new investigation tools (3D mapping, photogrammetry, Structure from Motion, high resolution remote sensing data). A particular attention should be paid to the southern part of the Mediterranean to reduce the gap of knowledge and of monitoring activities between the northwestern and the southern Mediterranean countries.
4. Local knowledge and historical data should be considered as a useful research tool for the monitoring of the dynamic of Posidonia meadows. Historical data must be used as a baseline for assessing the status of this habitat at national, sub-regional and regional levels, following the Ecosystem Approach roadmap decisions and in synergy with the regional directives and the Regional Sea Conventions.
5. Local variability and specific ecological/environmental conditions (such as hydrological conditions) should be taken into account during the implementation of the periodic assessment activities to understand the resilience of habitats in a changing environment, with a particular attention to the key habitats listed in the reference list of benthic habitats.
6. Deposit and debris of dead leaves of *Posidonia oceanica* (in banks, wracks, or banquettes) are ecologically important for a healthy ecosystem. Banquettes, in particular, could represent an asset for a sustainable tourism if we will be able to raise people's awareness that they are a strong sign of the Mediterranean identity.
7. Although our priority should always be to conserve the existing natural habitats and ecosystems, an improvement in the protocols used in restoration activities of both seagrass and macroalgae interventions is foreseen to facilitate and speed the recovery of the habitats.

SCIENTIFIC COMMITTEE MEMBERS
(In alphabetical order)

Ali BADREDDINE

PhD in Marine Biology
Tyre Coast nature reserve-TCNR
Benthic habitat (Benthos) Specialist
Marine Turtles Expert
Conservation of coastal ecosystems
E-mail: ali.badreddine@hotmail.com

Ivan GUALA

IMC - International Marine Centre - Loc.
Sa Mardini, Torre Grande, 09170
Oristano, Italy.
E-mail: i.guala@fondazioneimc.it

Habib LANGAR

Professor
UR17ES10, Département de Biologie,
Faculté des Sciences Mathématiques,
Physiques et Naturelles de Tunis,
Université
de Tunis El Manar - Campus Universitaire
Farhat Hached, 2092 Tunis, Tunisia
E-mail: habib.langar@fst.utm.tn

Monica MONTEFALCONE

PhD in Marine Science
Seascape Ecology Lab
DiSTAV, University of Genoa
Corso Europa 26, 16132 Genoa, Italy
E-mail: monica.montefalcone@unige.it

Christine PERGENT-MARTINI

Maitre de Conférences (MCF)
Habilitée à Diriger des Recherches (HDR)
University of Corsica – FRES 3041 /
UMR 6134 - Faculty of Sciences, BP 52,
20250 Corte, France.
E-mail: pmartini@univ-corse.fr

This symposium was prepared and organized under the Overall supervision of Mr **Khalil ATTIA**, director of UNEP/MAP-SPA/RAC.

ORGANISING COMMITTEE MEMBERS
(In alphabetical order)

Naziha BEN MOUSSA

Administrative Assistant
UNEP-MAP-SPA/RAC
Boulevard du Leader Yasser Arafet, B.P. 337,
1080, Tunis Cedex, Tunisia
E-mail: naziha.benmoussa@spa-rac.org

Cyrine BOUAFIF

Consultant, PhD in Biological Sciences
23, Avenue La Gazelle, Cité La Gazelle, 2083,
Ariana, Tunisia
E-mail: bouafif.cyrine@gmail.com

Imtinen KEFI

Financial Officer
UNEP-MAP-SPA/RAC
Boulevard du Leader Yasser Arafet, B.P. 337,
1080, Tunis Cedex, Tunisia
E-mail: imtinen.kefi@spa-rac.org

Dorra MAAOUI

Communication Assistant
UNEP-MAP-SPA/RAC
Boulevard du Leader Yasser Arafet, B.P. 337,
1080, Tunis Cedex, Tunisia
E-mail: dorra.maaoui@spa-rac.org

Monica MONTEFALCONE

PhD in Marine Science
Seascape Ecology Lab
DiSTAV, University of Genoa
Corso Europa 26, 16132 Genoa, Italy
E-mail: monica.montefalcone@unige.it

Atef OUERGHI

Ecosystems Conservation Programme Officer
UNEP-MAP-SPA/RAC
Boulevard du Leader Yasser Arafet, B.P. 337,
1080, Tunis Cedex, Tunisia
E-mail: atef.ouerghi@spa-rac.org

Yassine Ramzi SGHAIER

Project Officer - NTZ/MPA
UNEP-MAP-SPA/RAC
Boulevard du Leader Yasser Arafet, B.P.
337,1080, Tunis Cedex, Tunisia
E-mail: yassineramzi.sghaier@spa-rac.org

Leonardo TUNESI

*Research Director - Head of the Area
"Marine biodiversity, habitat and species
Protection"*
Italian National Institute for Environmental
Protection and Research, ISPRA – Via
Vitaliano Brancati, 60 I-00144 Roma, Italy.
E-mail: leonardo.tunesi@isprambiente.it

This symposium was prepared and organized under the Overall supervision of Mr **Khalil ATTIA**, director of UNEP/MAP-SPA/RAC.



Mediterranean
Action Plan
Barcelona
Convention



Specially Protected Areas Regional Activity Centre (SPA/RAC)
Boulevard du Leader Yasser Arafat
B.P. 337 - 1080 - Tunis Cedex - Tunisia +216 71 206 649 / +216 71 206 485
car-asp@spa-rac.org
www.spa-rac.org