



# PROCEEDINGS OF THE 5<sup>th</sup> MEDITERRANEAN SYMPOSIUM ON MARINE VEGETATION

Portorož, Slovenia, 27-28 October 2014

# ACTES DU 5<sup>ème</sup> SYMPOSIUM MEDITERRANEEN SUR LA VEGETATION MARINE

Portorož, Slovenie, 27-28 octobre 2014



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INSTITUTE OF THE REPUBLIC OF SLOVENIA  
FOR NATURE CONSERVATION



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With the support of MedKeyhabitats project Financed by the MAVA Foundation

*Avec le support du projet MedKeyhabitats Finance par la fondation MAVA*



May 2015

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Citation :

UNEP/MAP – RAC/SPA, 2015. Proceedings of the 5th Mediterranean Symposium on Marine Vegetation (Portorož, Slovenia, 27-28 October 2014). LANGAR H., BOUAFIF C., OUERGHI A., edits., RAC/SPA publ., Tunis: 264 p.

*PNUE/PAM – CAR/ASP, 2015. Actes du 5ème Symposium Méditerranéen sur la Végétation Marine (Portorož, Slovénie, 27-28 octobre 2014). LANGAR H., BOUAFIF C., OUERGHI A., édits., CAR/ASP publ., Tunis :264 p.*

ISBN: 978-9938-9574-0-2

## AVANT-PROPOS

Suite aux recommandations du **Plan d'action pour la conservation de la végétation marine en mer Méditerranée** (adopté par les Parties contractantes à la Convention de Barcelone, en 1999), du **Plan d'action pour la conservation du coralligène et des autres bio-constructions de Méditerranée** (adopté par les Parties contractantes à la Convention de Barcelone, en 2008), du **Plan d'Action pour la conservation des habitats et espèces associés aux monts sous-marins, aux grottes et canyons sous-marins, aux fonds durs aphotiques et aux phénomènes chimio-synthétiques en mer Méditerranée (Plan d'action pour les habitats obscurs)** (adopté par les Parties contractantes à la Convention de Barcelone, en 2013) et dans le but du développement des connaissances, une série de symposiums scientifiques, dédiée à ces habitats, a été initiée en 2000 par l'organisation du 1<sup>er</sup> symposium Méditerranéen sur la végétation marine. Ces initiatives visent essentiellement à faire le point sur les données scientifiques disponibles et à promouvoir la coopération entre les spécialistes qui travaillent en Méditerranée.

Cette année, avec la mise en œuvre du projet de Cartographie des habitats marins clés de la Méditerranée et la promotion de leur conservation par l'établissement d'Aires Spécialement Protégées d'Importance Méditerranéenne (ASPIM) « Projet Medkeyhabitats » financé par la fondation MAVA, l'opportunité s'est présentée pour organiser ensemble les symposiums suivants:

- 5<sup>ème</sup> Symposium Méditerranéen sur la Végétation Marine
- 2<sup>ème</sup> Symposium Méditerranéen sur la conservation du Coralligène et autres Bio-concrétions
- 1<sup>er</sup> Symposium Méditerranéen sur la conservation des Habitats Obscurs

Suite à l'offre de « the institute of the republic of Slovenia for nature conservation » lors du quatrième symposium organisé à Yasmine-Hammamet (Tunisie) du 2 au 4 décembre 2010 d'abriter la 5<sup>ème</sup> édition du même symposium, il a été convenu de les organiser ensemble back to back à Portorož, Sloveie, du 27 au 31 octobre 2014 comme suit :

- 5<sup>ème</sup> Symposium Méditerranéen sur la Végétation Marine du 27 au 28 octobre 2014
- 2<sup>ème</sup> Symposium Méditerranéen sur la conservation du Coralligène et autres Bio-concrétions du 29 au 30 octobre 2014)
- 1<sup>er</sup> Symposium Méditerranéen sur la conservation des Habitats Obscurs le 31 octobre 2014

Cette édition a vu l'inscription de plus de 140 participants en provenance de 17 pays Méditerranées, ce ne sont pas moins de 126 communications orales et posters qui devraient y être présentés.

Cette édition sera aussi l'occasion d'aborder des sujets d'actualités tels que les invasions biologiques, le réchauffement global, et leurs impacts sur les habitats clés de Méditerranée et de renforcer les liens entre les scientifiques et entre les institutions scientifiques.

Khalil ATTIA  
Directeur du CAR/ASP





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## PROGRAMME

### Monday 27 October 2014

- 8:00-9:00** Participants welcome and registration
- 9:00-10:00** **Opening of the Symposium**
- **Welcome speech** by **Khalil ATTIA**, *RAC/SPA Director*
  - **Welcome Speech** by **Martina Kačičnik Jančar** *from Institute of the Republic of Slovenia for Nature Conservation, on behalf of the director of the Institute*
  - Opening presentation: **Slovenian Coastal Sea: The Ark of Noah** by **Pr. Lovrenc Lipej** *from National Institute of Biology, Marine Biology Station Piran*
- 10:00-10:30** *Coffee break*
- 10:30-11:30** Keynote conference: **Terrestrial versus marine diversity of ecosystems. And the winner is: the marine realm** by **Pr. Charles F. BOUDOURESQUE**, RUITTON S., BIANCHI C.N., CHEVALDONNÉ P., FERNANDEZ C., HARMELIN-VIVIEN M., OURGAUD M., PASQUALINI V., PEREZ T., PERGENT G., THIBAUT T., VERLAQUE M.
- Session 1: Mediterranean Marine Vegetation: population, biology, ecology and dynamics – Marine "macroalgae"**  
Chair: **Charles F. BOUDOURESQUE**, Rapporteur: **Amelia Gomez GARETTA**
- 11:30-11:45** "**Marine forests at risk: solutions to halt the loss and promote the recovery of Mediterranean canopy-forming seaweeds**" by **Laura AIROLDI**, BALLESTEROS E., BUONUOMO R., VAN BELZEN J., BOUMA T.J., CEBRIAN E., DE CLERK O., ENGELEN A.H., FERRARIO F., FRASCHETTI S., GIANNI F., GUIDETTI P., IVESA L., MANCUSO F.P., MICHELI F., PERKOL-FINKEL S., SERRAO E.A., STRAIN E.M., MANGIALAJO L.
- 11:45-12:00** "***Cystoseira Sedoides* (desfontaines) C. Agardh des côtes tunisiennes : état actuel des connaissances**" by **Cyrine BOUAFIF**, OUERGHI A., LANGAR H.
- 12:00-12:15** "**Ecology and perturbations of Mediterranean deep-water algal communities: linking population biology and community ecology for conservation**" by **Bernat HEREU**, CAPDEVILA P., CEBRIAN E., DÍAZ D., GARRABOU J., KERTING D., LINARES C., NAVARRO L., PAUNER O., TEIXIDO N.
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- 12:45-13:00** Discussion
- 13:00-14:00** *Lunch*

**Session 2: Mediterranean Marine Vegetation: population, biology, ecology and dynamics – Marine Magnoliophytes**

Chair: **Rachid SEMROUD**, Rapporteur: **Robert TURK / Habib LANGAR**

- 14:00-14:15** "Insight into the typology of reef formations of the Mediterranean seagrass *Posidonia oceanica*" by **Charles F. BOUDOURESQUE**, BONHOMME D., ASTRUCH P., BONHOMME P., GOUJARD A., THIBAUT T.
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- 14:30-14:45** "The importance of genetic make-up for restoration success - a case study of the seagrass *Zostera noltii* hornem in a mediterranean lagoon" by **Marlene JAHNKE**, SERRA I.A., BERNARD G., PROCACCINI G.
- 14:45-15:00** "Modelling the reference conditions of the upper limit of *Posidonia oceanica* meadow" by **Gloria MISSON**, VACCHI M., MONTEFALCONE M., ARCHETTI R., BIANCHI C.N., FERRARI M.
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- 15:30-15:45** "Distribution and habitat requirements of *Zostera noltei* along the northern coast of Jerba Island (Southern Tunisia, Mediterranean sea)" by **Abdessalem SHILI**, BEN HMIDA A., BEN MAÏZ N., BOUDOURESQUE C.F.
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- 16:00-16:15** Discussion
- 16:15-16:45** *Coffee break*
- 16:45-18:00** **Poster Session**
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- 18:00-19:00** *Side event*
- One day in Albania coastline** by **Violeta ZUNA**, **Eno DODBIBA**
-

Tuesday 28 October 2014

**Session 3: Mapping, monitoring and management of Mediterranean marine vegetation**  
Chair: Gérard PERGENT, Rapporteur: Marina BONACORSI

**8:30-8:45** "Assessment of the conservation status of *Posidonia oceanica* meadows in the Samaria National Park, an MPA in Crete, Greece" by Dimitris POURSANIDIS, Antonios BARNIAS, LYMBERAKIS P.

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**9:30-9:45** Discussion

**9:45-10:00** "Combining modelling and historical data to define the status of *Posidonia oceanica* meadows" by Alice OPRANDI, MONTEFALCONE M., VACCHI M., COPPO S., DIVIACCO G., MORRI C., FERRARI M., BIANCHI C.N.

**10:00-10:15** "Monitoring and expansion of *Posidonia* monitoring networks along Corsican coastline" by Gérard PERGENT, BEIN A., BLANFUNE A., DEDEKEN M., OBERTI P., ORSINI A., PERGENT-MARTINI C., RUITTON S., SHORT F.

**10:15-10:30** "Monitoring of *Posidonia* meadows under the EC habitats directive: vehicular videography can estimate trends in coverage at low cost and high precision" by Stewart T. SCHULTZ, BAKRAN-PETRICIOLI T., KRUSCHEL C., PETRICIOLI D.

**10:30-10:45** Discussion

**10:45-11:15** *Coffee break*

**11:15-11:30** "A rapid and non-destructive assessment of your *Posidonia* meadow" by Gérard PERGENT

**11:30-12:15** Poster Session

**Session 3: Mapping, monitoring and management of Mediterranean marine vegetation (continued)**  
Chair: Gérard PERGENT, Rapporteur: Marina BONACORSI

**12:15-12:30** "Analyse critique de l'évaluation de l'état de conservation de l'herbier de posidonie dans le cadre du programme de Cartographie des Habitats Marins – CARTHAM" by Boris DANIEL, LAMOUREUX A.



- 12:30-12:45 "Are Mediterranean MPAs protecting marine forests?" by Fabrizio GIANNI, MANGIALAJO L.
- 12:45-13:00 Discussion
- 13:00-14:00 *Lunch*
- Session 4: Mediterranean vegetal assemblages: Impact and disturbance**  
Chair: Christine PERGENT-MARTINI, Rapporteur: Sandrine RUITTON
- 14:00-14:15 "Impact de la pêche par mini-chalut benthique sur les herbiers à *Posidonia oceanica* dans le secteur nord-est des îles Kerkennah (Tunisie) " by Ahmed BEN HMIDA, SHILI A., SGHAIER Y.R., RAIS C.
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- 14:30-14:45 "Assessing *Posidonia oceanica* beds regressions using anthropogenic pressures maps along a French coastal region" by Florian HOLON, BOCKEL T., BOISSERY P., DETER J.
- 14:45-15:00 "Arsenic concentrations in seagrass around the Mediterranean coast and seasonal variations" by Christine PERGENT-MARTINI, SALIVAS-DECAUX M., LANGAR H., PERGENT G., AKÇALI B., ALVAREZ-PÉREZ E., APOSTOLAKI E., BAKRAN-PETRICIOLI T., BELBACHA S., BORG J., BUIA C., CASALTA B., CELEBI B., FERNANDEZ-TORQUEMADE Y., HADJICHRISTOFOROU M., LLAGOSTERA I., LIPEJ L., LOPEZ Y ROYO C., MARCOU M., MAVRIC B., PANZALIS P., ROMERO J., SEMROUD R., SKOUFAS G., TURK R., WEITZMANN B., ZAPATA-SALGADO F.J.
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- 15:30-15:45 Discussion
- 15:45-16:15 *Coffee break*
- 16:15-17:00 **Poster Session**
- 17:00-17:30 **Awards for best poster**  
Jury: Charles F. BOUDOURESQUE, Rachid SEMROUD, Gérard PERGENT and Christine PERGENT-MARTINI, Secretaries: Cyrine BOUAFIF and Habib LANGAR
- 17:30-18:00 **Closure of the Symposium**

## PROGRAMME

Lundi 27 octobre 2014

- 8:00-9:00** Accueil et inscription des participants
- 9:00-10:00** **Ouverture du symposium**
- **Mot de bienvenue** par **Khalil ATTIA**, *Directeur du CAR/ASP*
  - **Mot de bienvenue** par **Martina Kačičnik Jančar** *de l'Institut de la République de Slovénie pour la conservation de la nature, au nom du directeur de l'institut*
  - **Présentation d'ouverture : Slovenian Coastal Sea: The Ark of Noah** par **Pr. Lovrenc Lipej** *de l'Institut National de Biologie, Station de Biologie Marine Piran*
- 10:00-10:30** *Pause café*
- 10:30-11:30** Conférence introductive : **Terrestrial versus marine diversity of ecosystems. And the winner is: the marine realm** par **Pr. Charles F. BOUDOURESQUE**, RUITTON S., BIANCHI C.N., CHEVALDONNÉ P., FERNANDEZ C., HARMELIN-VIVIEN M., OURGAUD M., PASQUALINI V., PEREZ T., PERGENT G., THIBAUT T., VERLAQUE M.
- Session 1 :** **Végétation marine de Méditerranée: population, biologie, écologie et dynamique – "Macroalgues" marines**  
Président : **Charles F. BOUDOURESQUE**, Rapporteur : **Amelia Gomez GARETTA**
- 11:30-11:45** "**Marine forests at risk: solutions to halt the loss and promote the recovery of Mediterranean canopy-forming seaweeds**" par **Laura AIROLDI**, BALLESTEROS E., BUONUOMO R., VAN BELZEN J., BOUMA T.J., CEBRIAN E., DE CLERK O., ENGELEN A.H., FERRARIO F., FRASCHETTI S., GIANNI F., GUIDETTI P., IVESA L., MANCUSO F.P., MICHELI F., PERKOL-FINKEL S., SERRAO E.A., STRAIN E.M., MANGIALAJO L.
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- 12:45-13:00** Discussion
- 13:00-14:00** *Déjeuner*

**Session 2 : Végétation marine de Méditerranée : population, biologie, écologie et dynamique – Magnoliophytes marines**

Président : Rachid SEMROUD, Rapporteur : Robert TURK / Habib LANGAR

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- 16:45-18:00 Session Posters
- 
- 18:00-19:00 *Evènement parallèle*
- One day in Albania coastline par Violeta ZUNA, Eno DODBIBA
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Mardi 28 octobre 2014

**Session 3 :**        **Cartographie, surveillance et gestion de la végétation marine de Méditerranée**  
Président : Gérard PERGENT, Rapporteur : Marina BONACORSI

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**10:30-10:45**     Discussion

**10:45-11:15**     *Pause café*

**11:15-11:30**     "**Une évaluation rapide et non destructive de votre herbier de Posidonie**" par Gérard PERGENT

**11:30-12:15**     Session Posters

**Session 3:**        **Cartographie, surveillance et gestion de la végétation marine de Méditerranée**  
(suite)            Président : Gérard PERGENT, Rapporteur : Marina BONACORSI

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- 15:30-15:45 Discussion
- 15:45-16:15 *Pause café*
- 16:15-17:00 **Session Poster**
- 17:00-17:30 **Remise de prix pour le meilleur poster**  
Jury : Charles F. BOUDOURESQUE, Rachid SEMROUD, Gérard PERGENT and Christine PERGENT-MARTINI, Secrétaires : Cyrine BOUAFIF and Habib LANGAR
- 17:30-18:00 **Clôture du Symposium**

# **KEYNOTE CONFERENCE**

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# **CONFERENCE INTRODUCTIVE**



**Charles F. BOUDOURESQUE, RUITTON S., BIANCHI C.N., CHEVALDONNÉ P., FERNANDEZ C., HARMELIN-VIVIEN M., OURGAUD M., PASQUALINI V., PEREZ T., PERGENT G., THIBAUT T., VERLAQUE M.**

Aix-Marseille University and Toulon University, Mediterranean Institute of Oceanography (MIO), CNRS/IRD UM 110, University campus of Luminy, 13288 Marseille cedex 9, France.

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## **TERRESTRIAL VERSUS MARINE DIVERSITY OF ECOSYSTEMS. AND THE WINNER IS: THE MARINE REALM**

### **Abstract**

*The concept of biodiversity encompasses a wide range of scales and metrics, from genetic and species diversity to functional and ecosystem diversity. Ecosystems can be characterized by a number of descriptors, such as species richness, type of primary production (where present), e.g. photosynthesis and chemosynthesis, production and biomass of primary producers (where present), the balance between nutrients and primary production (HNLC, HNHC, LNLC and LNHC systems), fate of primary production (e.g. herbivores vs detritivores), type of the primary production recycling (slow vs. rapid), production and biomass of secondary producers structure and length of food webs, dominant control (bottom-up, wasp-waist or top-down), import of organic matter from adjacent ecosystems, export of organic matter to other ecosystems and the carbonate cycle. Comparison of a number of marine ecosystems (Posidonia oceanica meadows, Cystoseira forests, Macrocyctis forests, coralligenous constructions, coral reefs, underwater sea caves and hydrothermal vents), with some terrestrial ecosystems (temperate and tropical forests, matorral and grasslands), evidences an obviously higher functional ecosystem diversity in the marine realm. Similarly, marine phyletic diversity is far higher than the terrestrial, whose higher species diversity is virtually due to a few phyla. This is consistent with the fact that Life originated in the oceans, ~3.8 Ga ago, while the conquest of land by Life occurred comparatively in recent times, ~0.5 Ga ago.*

**Key-words:** Ecosystem diversity, ecosystem functioning, marine ecosystems, terrestrial ecosystems

### **Introduction**

The concept of biodiversity encompasses a wide range of scales and metrics, from genetic and species diversity to functional and ecosystem diversity, from the sample and local scale to global scale, in addition to the distribution of individuals among species (heterogeneity diversity or evenness). Biodiversity is therefore a multidimensional concept that is impossible to assess with a single measure (Gray, 2000; Sala & Knowlton, 2006; Boudouresque, 2011a).

At the species level, the diversity of the marine realm is much lower than that of the terrestrial realm: only 13-15% of the ~2 000 000 currently described species (Bianchi & Morri, 2000; Sala & Knowlton, 2006; Boeuf, 2011; Mora *et al.*, 2011). In contrast, marine diversity is higher at phylum level: nearly all of the 31 phyla of metazoans are present in the marine realm and 12 of them are exclusively marine (Boeuf, 2011).

In contrast to the taxonomic diversity, from the species level to high-level taxa such as phyla, little attention has been paid to the ecosystem diversity of the marine vs. the terrestrial realms. Here, we consider the marine and terrestrial realms proper, excluding intermediate habitats and ecosystems such as coastal lagoons, mangroves, lakes, rivers, peat bogs and underground more or less flooded caves. We compare the characteristics (species richness and functioning) of a number of marine ecosystems, such as the Mediterranean *Posidonia oceanica* meadow, the Mediterranean *Cystoseira* forests and barren grounds, the Mediterranean coralligenous, the *Macrocystis* forests, coral reefs, underwater sea caves and deep-sea hydrothermal vents, with characteristics of terrestrial ecosystems, such as temperate and tropical forests, matorrals and grasslands.

### **Ecosystem descriptors**

An ecosystem can be defined as a unit of biological organization made up of all the organisms in a given area (that is 'community'), interactions between these organisms (i.e. symbioses: mutualism, parasitism, predation, etc.) and interactions between organisms and the abiotic environment (habitat), so that a flow of energy leads to a characteristic trophic structure and material cycles within the system (Odum, 1969; Likens, 1992; Frontier, 1999).

Ecosystems can be characterized by a number of descriptors, such as species richness, type of primary production (where present), production and biomass of primary producers (where present), the balance between nutrient availability and primary production, the fate of primary production, type of the primary production recycling, production and biomass of secondary producers, structure and length of food webs, dominant types of control (bottom-up, wasp-waist or top-down), import of organic matter from adjacent ecosystems, export of organic matter to other ecosystems, and the carbonate cycle (where present).

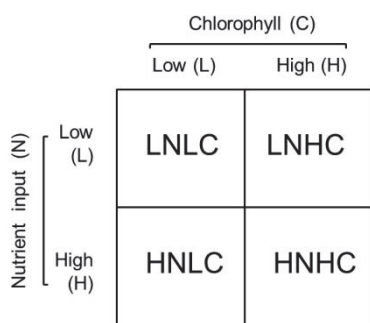
**(i)** Species richness is the mean number of species by sample (point diversity) and the cumulative number of species at the ecosystem level ( $\alpha$  diversity) and regional scale ( $\gamma$  and  $\epsilon$  diversity) (Whittaker, 1972; Gray, 2000; Boudouresque, 2011a). The former can be relatively high while the latter is relatively low and vice versa (Boudouresque, 2011a).

**(ii)** Type of primary production (where present) is photosynthesis versus chemosynthesis. In photosynthesis, the source of energy is sunlight. Classically, 3 types of photosynthesis are recognized: C3, C4 and CAM. In C3 primary producers (PPs), the RuBisCO enzyme fixes the incoming CO<sub>2</sub> into a 3-carbon molecule. In C4 PPs, which typically dwell in warm and dry terrestrial climates, the CO<sub>2</sub> is first fixed into a 4-carbon molecule. CAM PPs are roughly viewed as a mix of C3 and C4 PPs, adapted to very dry terrestrial environments, although this is an oversimplification (Dodd *et al.*, 2002). In chemosynthesis, the conversion of inorganic carbon molecules (carbon dioxide or methane) and nutrients into organic matter uses the oxidation of inorganic molecules (e.g. hydrogen sulphide), rather than sunlight, as a source of energy (Campbell *et al.*, 2008; Bertrand *et al.*, 2011).

**(iii)** Production and biomass of primary producers (where present). Primary production is the synthesis of organic compounds via photosynthesis and chemosynthesis, per surface area and time interval; it is distinguished as either net (NPP; Net Primary Production) or gross (GPP; Gross Primary Production), the former accounting for the losses due to respiration, maintenance, production of volatile compounds and offspring. The NPP therefore roughly corresponds to the biomass increase, due to growth, per time interval.

(iv) Balance between nutrients and primary production. Four categories of ecosystem can be distinguished on the basis of nutrient availability (proxy: nitrogen N) and primary production (proxy: chlorophyll), both roughly distinguished into low (L) and high (H) levels (Fig. 1): LNLC (e.g. Mediterranean pelagic ecosystems and terrestrial forests), HNLC (e.g. Antarctic pelagic ecosystems), LNHC (e.g. coral reefs and Mediterranean *P. oceanica* meadows) and HNHC (e.g. upwelling pelagic ecosystems and agricultural systems). When two types of PPs are present, as in seagrass meadows, the plant can be LNHC while the leaf epibionts can be HNHC under eutrophication conditions.

(v) Fate of primary production. The primary production can be directly consumed by herbivores (browsers or grazers). However, if PPs are protected by chemical and/or physical defences, or if herbivores are scarce (Lawrence, 1975; Rhoades & Cates, 1976; Boudouresque & Verlaque, 2013), consumption may be low and the products of PPs end up in the litter, in a carbon sink, or are exported toward adjacent ecosystems. If not sequestered in a sink, the organic matter coming from PPs ends up in the detritus-feeder pathway, within either the producing or adjacent ecosystems.



**Fig. 1: The 4 categories of ecosystems based upon the relation between availability of nutrients (e.g. N) and abundance of chlorophyll (a proxy of primary production).**

(vi) Type of primary production recycling. Magnoliophyta (seagrasses and land plants; kingdom Archaeplastida) produce material hard to degrade that requires months or years to enter the detritus food webs (slow recycling). ‘Macroalgae’, a polyphyletic group of organisms belonging to Chlorobionta,

Rhodobionta (green and red algae; Archaeplastida) and Phaeophyceae (brown algae; kingdom Stramenopiles), are easier to degrade (weeks or months) and quickly enter the food webs (rapid recycling). Most terrestrial ecosystems rely on the first type of primary producers while most benthic photophilous marine ecosystems are based upon the second. The combination of two sets of PPs is a very rare feature, met with in seagrass ecosystems and mangroves (the latter not considered here) (Boudouresque *et al.*, 2006).

(vii) Production and biomass of secondary producers. Secondary producers encompass a suite of trophic levels, from consumers of primary producers, detritus-, filter- and suspension-feeders, to top-predators. Secondary production is usually very low as compared with primary production.

(viii) Structure and length of food webs. Food webs are generally not a linear suite of species, from the prey to the predator, but a complex network of relationships. As a result, counting trophic level number is not an easy task, especially when omnivores and detritus-feeders are involved. Due to the relatively low ecological efficiency of the energy transfer from one level to the next, especially in the terrestrial environment, the number of trophic levels rarely exceeds 4 in terrestrial ecosystems and cannot exceed 7 in marine systems (Fenchel, 1988; Ricklefs & Miller, 2005)

(ix) Dominant control. The control of the ecosystem functioning can be bottom-up, top-down or wasp-waist (Cury *et al.*, 2003). Bottom-up control means that the lowest level, that of primary producers, controls the higher levels, *via* the nutrient supply. Top-down control means that the highest trophic level (top predators) controls the lowest levels, *via* a sort of cascade effect (‘the enemy of my enemy is my friend’; Holt, 2000).



Finally, in the wasp-waist control, the population dynamics of an intermediate trophic level controls the others, both downwards and upwards. In the real world, the control is rarely (or never) of only one type; rather, there is a full gradient between e.g. mainly top-down and mainly bottom-up controls.

(x) Import of organic matter from adjacent ecosystems. Some ecosystems, especially in the terrestrial realm, are self-sufficient, i.e. they totally, or almost totally, depend upon the autochthonous PPs. Other ecosystems depend, in part or totally, on imported organic matter.

(xi) Export of organic matter to other ecosystems. Some ecosystems export a greater or lesser part of the organic matter coming (directly or indirectly) from PPs, in the form of e.g. carcasses, faeces and detritus, to other ecosystems. This feature, related to gravity or hydrodynamics, is common in marine ecosystems.

(xii) The carbonate cycle. Precipitation of calcium (and/or magnesium) carbonate, mainly in the form of calcite and aragonite, by living organisms, has a high energetic cost. It often corresponds to a defence strategy. Calcified organisms, mostly represented among the marine biota, are generally less palatable than fleshy ones, although some species, such as sea urchins and fish, do graze them. Precipitation of calcium carbonate may result in a sink of carbon, whatever the source of this carbon.

### Examples of marine and terrestrial ecosystems

A small number of marine and terrestrial ecosystems were chosen for this comparative approach (Tab. 1).

*Posidonia oceanica* is a seagrass endemic to the Mediterranean Sea (Boudouresque & Verlaque, 2008). It dwells from the sea level down to 35-45 m depth and is the ecosystem engineer of an ecosystem that plays a major role in the Mediterranean coastal zone (Boudouresque *et al.*, 2006, 2009; Pergent *et al.*, 2012; Personnic *et al.*, 2014).

Sub-littoral rocky substrates of the Mediterranean harbour an ecosystem that can occur under multiple 'stable' states (MSS) (see e.g. Boudouresque *et al.*, 2005, for the MSS concept), the functioning of two of these MSS is so contrasted that we will treat them hereafter as two different ecosystems: (i) The 'forest' state, dominated by long-living species of the *Cystoseira* genus (Phaeophyceae, Stramenopiles), here mainly *C. brachycarpa*. (ii) The barren grounds, where overgrazing by sea urchins, fish and other macro-herbivores removes the layer of *Cystoseira* and other MPO (Multicellular Photosynthetic Organisms), leaving a more or less bare rocky substrate covered with encrusting corallines (calcareous red algae) and 'invertebrates' (Sala *et al.*, 2011, 2012; Boudouresque & Verlaque, 2013)

*Macrocystis pyrifera* is a pseudo-perennial giant (up to 40 m tall) brown alga (Phaeophyceae, Stramenopiles) that mainly thrives in southern hemisphere cold waters, with isolated stations in the northern hemisphere, in California. It builds impressive underwater forests, with leaves spreading at the sea surface (Dayton *et al.*, 1992; Steneck *et al.*, 2002).

The Mediterranean coralligenous is a complex of ecosystems. It is characterized by basal calcareous concretions of biogenic origin and by a canopy of large sessile filter- and suspension-feeders (gorgonians and sponges). The biogenic concretion is produced by the accumulation of encrusting red macro-algae (order Corallinales, Rhodobionta, Archaeplastida) and calcareous metazoans (bryozoans, serpulid annelids, etc.) growing in dim light conditions, between 20 and 120 m depth. Accretion of the concretions results from

a positive balance between bio-constructing organisms and bio-erosion by e.g. sponges, cyanobacteria and grazers (Ballesteros, 2006).

Sub-littoral marine caves are found in a wide variety of rocks, from sedimentary (e.g. karst caves) to metamorphic and igneous rocks. Here, only caves with a sub-littoral entrance, either horizontal or descending (the latter resulting in the trapping of cold water), are considered. With the exception of the dim-light entrance, they are characterized by more or less total darkness. Caves constitute a functional unit, although extending along a sharp gradient of light, food availability, confinement and communities, from the entrance to the most remote zones (e.g. Laborel & Vacelet, 1958; Bianchi & Morri, 1994; Vacelet *et al.*, 1994; Rastorgueff *et al.*, 2011).

Coral reefs are inter-tropical and sub-littoral biogenic formations. Accretion is due to a positive balance between bio-constucting organisms, mainly scleractinians (metazoans), but also to e.g. calcareous red algae and molluscs, and bio-erosion by e.g. cyanobacteria, Chlorobionta, sponges, molluscs, and grazing fish (Tribollet & Payri, 2001). Scleractinians have a mutualistic relationship with unicellular Dinobionta of the genus *Symbiodinium*, living within the host cells. A unique process of nitrogen recycling between scleractinians and Dinobionta enables coral reefs to develop a lush ecosystem in highly oligotrophic waters.

Deep-sea hydrothermal vents are oases of microbial and metazoan life, associated with deep thermal springs occurring at ocean ridge spreading centres (2-3 km deep) and which are based upon chemosynthesis. Metazoans utilize chemosynthesis performed by sulphur-oxidizing prokaryotes to derive most or all of their energy supply, rather than photosynthesis, at depths where light cannot penetrate (Corliss *et al.*, 1979).

Mediterranean matorral is a thermophilous bushy formation dominated by shrubs such as *Arbutus unedo*, the dwarf oak *Quercus coccifera* and the rosemary *Rosmarinus officinalis*. It is furthered by fire and its long-term fate, in the framework of the succession and without fire recurrence, is generally to evolve into an evergreen oak forest.

The evergreen *Quercus* spp. forest, the deciduous oak forest (*Quercus* spp. and *Fagus sylvatica*) and the tropical rain forest are climax forests in Mediterranean, temperate Europe and inter-tropical areas, respectively. They are characterised by long-lived trees (at least several centuries) that act as ecosystem engineers. The tropical rain forest is an ecosystem type that occurs within the inter-tropical zone. It experiences high average temperature and rainfall. Rain forests exhibit high levels of species diversity and very high primary production (Clark *et al.*, 2011).

The *Pinus nigra* ssp. *laricio* (Corsican pine, lariciu) (also known as *P. nigra* ssp. *salzmannii* var. *corsicana*: its taxonomy is a much debated question) forest is an emblematic ecosystem of the Corsican mountains (Gamisans, 1975, 2010).

Plain grasslands of Western Europe can be regarded as a stage in the succession towards climax oak or *Fagus* forests, artificially maintained by farmers and cattle rearing. However, it can also be considered that cattle just take the place of large herbivores, driven to extinction by Humans during the Holocene, such as Przewalski's horse *Equus ferus przewalskii* and the European bison *Bison bonasus*. These grasslands could therefore constitute a natural climax ecosystem.

### **Calibration of the ecosystem descriptors used**

We calibrated the selected ecosystem descriptors on a 5-step semi-quantitative scale (very low to very high; changed into 1 through 5 for statistical treatment). Where not applicable (e.g. descriptors defined by presence-absence only), this scale was simplified into a 2-

step scale (very low or 1 vs. very high or 5) (Tab. 2). Some of the above-mentioned descriptors, e.g. C3 vs. C4 and CAM photosynthesis, were not taken into consideration here. For clarity, we kept hereafter the numbering of the descriptors used previously, even when descriptors were split or non-retained.

**(i) Species richness (SR):** point diversity. Here, we only considered the number of species of MPOs per sample (SR-MPO) and the number of species of metazoans per sample (SR-M) (Tab. 2). MPOs are a functional polyphyletic group of taxa belonging to Embryophyta, Chlorobionta, Rhodobionta (kingdom Archaeplastida) and Phaeophyceae (kingdom Stramenopiles); see Boudouresque (2011b) for taxonomic treatment. SR-MPO can usually be found in phytosociological *relevés* (Coppejans & Boudouresque, 1975). The minimal area usually constitutes a convenient proxy for point diversity (see Boudouresque, 1974; Panayotidis, 1979). Sample size differs between taxa, e.g. between insects, fish and birds, so that the value for the descriptor SR-M is a rough cumulative estimate.

**(iii-a) Photosynthetic primary production (PPP):** DM (dry mass) of NPP per m<sup>2</sup> per year (SI symbol for year: a): DM m<sup>-2</sup> a<sup>-1</sup>. **(iii-b) Chemosynthetic primary production (CPP):** NPP as DM m<sup>-2</sup> a<sup>-1</sup>. Where sessile metazoans with mutualistic symbiosis with chemosynthetic prokaryotes draw their mass from this relationship, these metazoans were considered as PPs. **(iii-c) Biomass of photosynthetic primary producers (B-PPP):** DM m<sup>2</sup>. In terrestrial ecosystems, it includes root biomass, which can be higher than above-ground biomass (Crouzet, 1973), but is unfortunately often poorly known. Where living organisms incorporate calcium (and sometimes magnesium) carbonate (calcite or aragonite), we considered the non-calcified mass only. However, the non-calcified mass is often not available from literature data, especially when calcification is inconspicuous. When biomass fluctuates over the year, e.g. is high during the spring blooms and low in autumn when dead leaves are shed, we considered the highest annual value (DM m<sup>-2</sup>). **(iii-d) Ratio between primary production and biomass (PP/B).**

**(iv) Balance between nutrients and primary production (HNLC, HNHC, LNLC and LNHC systems).** Here, according to the ecosystems taken into account, i.e. in the absence of agricultural systems and pelagic ecosystems, only LNHC was considered (Tab. 2).

**(v) Fate of the primary production.** **(v-a)** The herbivore pathway (HP) corresponds to the percentage of direct consumption of the primary production (PPP or CPP). **(v-b)** The detritus-feeder pathway (DP) corresponds to the percentage of the primary production that is consumed in the form of dead products within the producer ecosystem. **(v-c)** The carbon sink (CS) corresponds to the mass of organic matter (DM m<sup>-2</sup> a<sup>-1</sup>) that is durably (i.e. at the geological timescale) sequestered within the ecosystem. That means that the temporary sequestration within biomass, which only lasts the lifespan of organisms (generally PPs), possibly centuries in trees (in the form of wood), is not a true sink; some authors and politicians erroneously consider the temporary sequestration of carbon within biomass as a sink, in the framework of the attempts to mitigate climate change.

**(vi) The primary production recycling can be slow (vi-a: PP-SR), rapid (vi-b: PP-RR) or both (vi-c: PP-SRR).** No metric was associated with these descriptors (Tab. 2).

**(vii) Production and biomass of secondary producers.** Here, only the biomass (B-SP) will be considered, as data on production are not available in the literature for most ecosystems. As for PPP, we intended to consider the non-calcified mass only.

(viii) Structure and length of food webs (FW). Only the length, i.e. the number of trophic levels, was considered here (Tab. 2). Data are generally not available in the literature and were therefore mostly derived from the interpretation by the authors of published food webs, and from the authors' expertise.

(ix) Dominant control. Wasp-waist control was not considered, since it is mainly known in some pelagic ecosystems not taken into account here. The metric was the estimated ratio between the top-down and the bottom-up control (TDC) (Tab. 2).

(x) Import of organic matter from adjacent ecosystems (IM). The metric was the percentage of the organic matter (OM) of allochthonous origin consumed by secondary consumers.

(xi) Export of organic matter to adjacent ecosystems (EX). The metric was the percentage of the primary and secondary production that is exported to other ecosystems, as e.g. dead leaves, migrant individuals leaving the ecosystem, faeces and carcasses.

(xii) The carbonate cycle (CA). Here, we consider the precipitation of carbonates ( $\text{DM m}^{-2} \text{ a}^{-1}$ ), without taking into consideration bio-erosion.

### Possible flaws and biases

Most ecosystems, in the terrestrial realm, are known from a descriptive point of view: typology, spatial structure (e.g. stratification), list of taxa. Primary production and biomass are more rarely tackled, and if so only few taxa (such as embryophytes) are considered. Secondary production and biomass are often ignored. Available studies generally deal with a single compartment (e.g. soil, embryophytes, fungi in a customary meaning, birds). Functional characteristics of the ecosystem are therefore often difficult to obtain. The same is true for the marine realm, although to a lesser extent. In both realms, a comprehensive approach going beyond a few well-studied taxa (e.g. embryophytes, birds, fish) and taking into account the real diversity (including bacteria, archaea and unicellular eukaryotes such as excavates, alveolates and Rhizaria, and assessing their crucial functional role), is often lacking. To fill the table 3, the authors were therefore often reduced to relying on their intuitions and their own experience (expert judgement).

We here intended to consider theoretical 'natural ecosystems'. But what is a natural ecosystem? First, there is probably no ecosystem on Earth, including deep Amazonia and offshore oceans, which has not been more or less strongly impacted by modern Humans. Overfishing in the marine realm, massive extinctions in Europe and Asia during the Holocene, massive extinctions in America and Australia just after the arrival of humans (the 'blitzkrieg'), profoundly changed the functioning of ecosystems (Planhol, 2004; Zalasiewicz *et al.*, 2011). What would European terrestrial ecosystems be with large wandering herds of Przewalski's horses, European bison, maybe mammoths, in addition to high density of bears, wolves, etc.? Second, the baseline of ecosystem functioning is not only generally unknown, in the absence of reliable ancient data (Tegner & Dayton, 1997; Sáenz-Arroyo *et al.*, 2005), but constantly shifting with natural climate oscillations (Le Roy-Ladurie, 2004; Martrat *et al.*, 2004; Kuhlemann *et al.*, 2008), in addition, of course, to the climate change induced by man (Stott *et al.*, 2000; Lejeune *et al.*, 2010).

Finally, the choice of the 7 marine ecosystems and the 6 terrestrial ecosystems (more exactly: types of ecosystem) considered here is subjective (Tab. 1). These ecosystems hardly account for the much broader diversity spectrum of Earth ecosystems. Sandy sublittoral bottoms and deep abyssal ecosystems in the marine realm, tundra and deserts in the terrestrial realm, prone to increasing the functional diversity, are obviously lacking.

The choice of the 19 considered ecosystem descriptors (Tab. 2) is probably less subjective. However, they should be weighed: does magnoliophytes species richness play the same role in ecosystem functioning as the import of organic carbon and the fate of the primary production? Here, we chose not to weight the descriptors. Preliminary attempts to weight descriptors did not result in obvious changes in the MDS (see below) of the ecosystem diversity (results not shown).

### **Main functional traits of the considered ecosystems and ecosystem diversity**

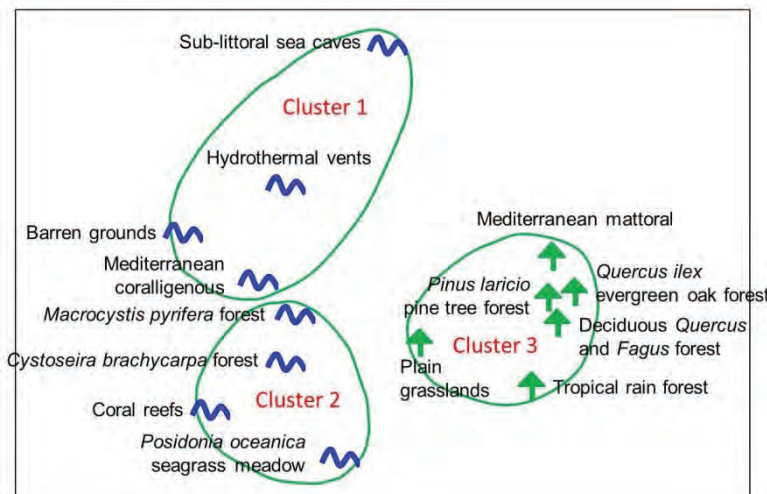
Some ecosystem functioning traits of the considered ecosystems (Tab. 3) are shared by several ecosystems, whether terrestrial or marine; others are mainly shared by terrestrial ecosystems or by marine ecosystems. Finally, some functioning traits are distinctive of a single or few ecosystems.

The huge precipitation of carbonates is specific to coral reefs, although it also occurs in the Mediterranean coralligenous. The occurrence of an important carbon sink is specific to the *Posidonia oceanica* meadow and coral reefs, *via* two different pathways, the organic carbon and the carbonates, respectively. Massive importation of organic carbon is specific to the *Macrocystis pyrifera* forest and to marine caves. Chemosynthetic primary production characterizes hydrothermal vents. Juxtaposition of slow and rapid recycling of the primary production characterizes the *Posidonia oceanica* ecosystem. Overall, it seems that more functional traits are unique to marine than to terrestrial ecosystems.

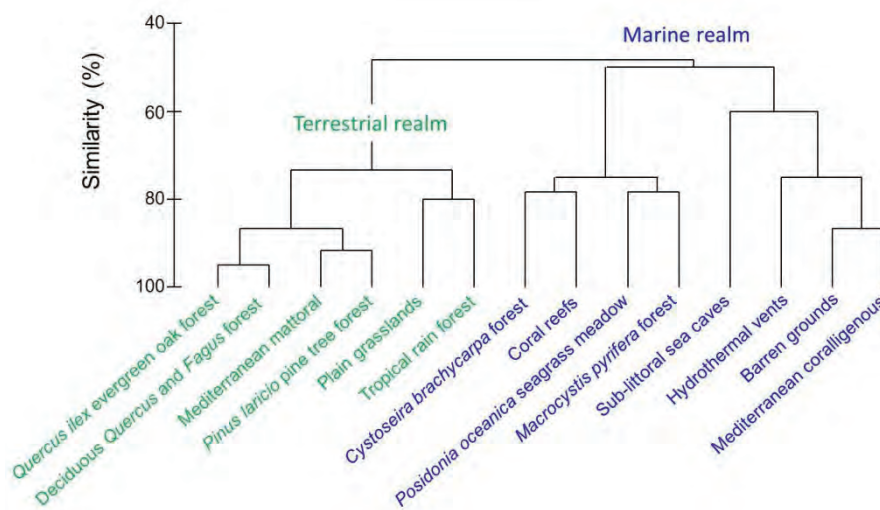
We compared the functioning diversity of marine and terrestrial ecosystems by means of an MDS (Multidimensional scaling) analysis, using S17 Bray-Curtis similarity index (Clarke & Warwick, 1994; Clarke & Gorley, 2006). Grouping, based on a complete linkage clustering, was superimposed on the ordination plot. This ordination analysis was followed by a PERMANOVA on S17 Bray-Curtis similarity index (Anderson *et al.*, 2008) (Figs. 2, 3). The results showed marine and terrestrial ecosystems as clearly separated, which was far from unexpected. Interestingly, marine ecosystems were more spread out on the MDS space than terrestrial ones (Fig. 2). Cluster analysis evidenced three main groups (Fig. 3). Marine ecosystems gathered into two clusters (clusters 1 and 2; Fig. 2), while terrestrial ecosystems constituted only one cluster (cluster 3; Fig. 2); cluster 1 seemed to correspond to cases where photosynthetic PPs are absent or inconspicuous (unexpectedly including the Mediterranean coralligenous), while cluster 2 corresponds to high photosynthetic primary production.

The *P. oceanica* ecosystem has been claimed to be ‘the most terrestrial of marine ecosystems’ (Boudouresque *et al.*, 2006), due to a set of characters supposed to be unique in the marine realm, while more common on land, such as the relatively high biomass of PPs, the low herbivore pressure and the importance of the detritus-feeder pathway. This was consistent with the continental origin of seagrasses. However, this relationship was not confirmed by the present analyses (Figs. 2, 3).





**Fig. 2: Two-dimensional MDS (Multidimensional scaling) ordination plot of the terrestrial (green) and marine (blue) ecosystems considered, according to the score of each descriptor. Grouping, based on complete linkage at 60%, using S17 Bray-Curtis similarity index, is superimposed on the ordination plot. Stress: 0.07.**



**Fig. 3: Clustering of the ecosystems considered based on complete linkage. S17 Bray-Curtis similarity index. Terrestrial (green) and marine (blue) ecosystems.**

### Discussion and conclusion

The results showed the functional diversity of ecosystems to be higher in the marine realm, which is characterized by a number of outstanding ecosystems with unique functioning patterns. It may be recalled that a similar pattern is known for phyletic diversity, which is much higher in the marine than in the terrestrial realm, whose higher species diversity is virtually due to a few phyla. This is consistent with the fact that Life began in the sea, ~3.8 to 3.5 Ga ago, while the conquest of land by Life occurred in comparatively recent times, ~0.5 Ga ago.

However, the sharp contrast between strictly marine and strictly terrestrial ecosystems should probably fade if transitional ecosystems, such as mangroves, coastal lagoons, salt marshes, lakes and rivers, were taken into consideration. In such ecosystems from intermediate habitats, marine and land features may coexist to challenge any rigid schematisation.

### Acknowledgements

The authors are indebted to Cyrine Bouafif and Habib Langar, who organized the symposium, for this invited talk and paper, and to Michael Paul for improving the English text.



Tab. 1: Ecosystems considered: main regional origin of data, altitude or depth and some of the used references (in addition to the authors' expertise).

Ecosystem	Region	Altitude or depth (m)	Some of the references used
Marine ecosystems			
<i>Posidonia oceanica</i> seagrass meadow	Port-Cros Island (NW Mediterranean)	-1 to -10 m	Panayotidis (1979), Pérès & Picard, 1964; Boudouresque <i>et al.</i> (2006), Personnic <i>et al.</i> (2014)
<i>Cystoseira brachycarpa</i> forest	NW Mediterranean	-1 to 5 m	Ballesteros (1990)
Barren grounds	NW Mediterranean	-1 to 5-10 m	Sala <i>et al.</i> (2011); Boudouresque & Verlaque (2013)
Mediterranean coralligenous	Provence, Catalonia and Adriatic Sea	-20 to -50 m	Laubier (1966), Boudouresque (1974), Sartoretto (1995, 1996); Ballesteros (2006), Caselato & Stefanon, (2008)
<i>Macrocystis pyrifera</i> forest	California (NE Pacific)	Sea surface to -30 m	Foster & Schiel (1985), Harrold & Pearse (1987), Tegner <i>et al.</i> (1997), Steneck <i>et al.</i> (2002)
Sub-littoral marine caves	NW Mediterranean	-5 to -30 m	Laborel & Vacelet (1958), Bianchi & Morri (1994), Vacelet <i>et al.</i> (1994), Southward <i>et al.</i> , 1996; Harmelin <i>et al.</i> (1998), Rastorgueff <i>et al.</i> (2011), Rastorgueff (2012)
Coral reefs	Moorea (French Polynesia)	Sea surface to -5 m	Borowitzka (1981), Salvat (1992), Tribollet (2001), Tribollet <i>et al.</i> (2006)
Hydrothermal vents	East Pacific Rise	ca.-2 500 m	Laubier (1986), Desbruyères <i>et al.</i> (1998)
Terrestrial ecosystems			
Mediterranean maotral	Corsica	50 to 200 m	Molinier (1959), Terradas (1991)
<i>Quercus ilex</i> evergreen oak forest	Southern France	50 to 300 m	Lossaint & Rapp (1978)
Deciduous <i>Quercus</i> and <i>Fagus</i> forests	Western Europe	200 to 800 m	Lemée (1978), Duvigneaud (1980)
<i>Pinus laricio</i> pine tree forest	Corsica	1 300 to 1 800 m	Gamisans (1975), Ranger (1978), Gamisans (2010)
Plain grasslands	Western Europe	100 to 500 m	Ricou (1978), Frontier (1999), Bernhardt-Römermann <i>et al.</i> (2011)
Tropical rain forest	Western Africa	100 to 500 m	Gillespie <i>et al.</i> (1992), Clark <i>et al.</i> (2001), Liddell <i>et al.</i> (2007), Clark <i>et al.</i> (2011)

Tab. 2: Calibration of ecosystem descriptors. na = non-applicable. See text for acronyms.

Ecosystem descriptor	Metric	Absent, na or very low (VL)	Low (L)	Medium (M)	High (H)	Very high (VH)
Species richness MPOs (SR-MPO)	Number of species /sample	< 10	10-40	40-100	100-200	> 200
Species richness metazoans (SR-M)	Number of species /sample	< 20	20-60	60-150	150-300	> 300
Photosynthetic primary production (PPP)	g DM m <sup>-2</sup> a <sup>-1</sup>	< 250	250-500	500-1 000	1 000-2 000	> 2 000
Chemosynthetic primary production (CPP)	g DM m <sup>-2</sup> a <sup>-1</sup>	< 100	100-200	200-400	400-800	> 800
Biomass of photosynthetic primary producers (B-PPP)	g DM m <sup>-2</sup>	< 500	500-2 000	2 000-5 000	5 000-20 000	> 20 000

*(continued)*

Ratio between primary production and biomass (PP/B)	Ratio	< 0.05	0.05-0.50	0.50-5.00	5-50	> 50
Nutrient/biomass balance: LN-HC	No metric	No	na	na	na	Yes
Fate of primary production (PP): herbivore pathway (HP)	%	< 10 %	10-20 %	20-40 %	40-75 %	> 75 %
Fate of primary production (PP): detritus-feeder pathway (DP)	%	< 10 %	10-20 %	20-40 %	40-75 %	> 75 %
Carbon sink (CS)	g DM m <sup>-2</sup> a <sup>-1</sup>	< 10	10 to 20	20 to 40	40 to 100	> 100
Type of the primary production recycling: only slow re-cycling (PP-SR)	No metric	No	na	na	na	Yes
Type of the primary production recycling: only rapid re-cycling (PP-RR)	No metric	No	na	na	na	Yes
Type of the primary production recycling: both slow and rapid recycling (PP-SRR)	No metric	No	na	na	na	Yes
Biomass of secondary producers (B-SP)	g DM m <sup>-2</sup>	< 20	20 to 40	40 to 80	80 to 150	> 150
Structure and length of food webs (FW)	Number of trophic levels	3	3 to 4	4 to 5	5 to 6	6 to 7
Top-down control TD (vs. bottom-up BU) (TDC)	Ratio TD/BU	0 to 0.2	0.2 to 0.4	0.4 to 0.6	0.6 to 0.8	> 0.8
Import of organic matter from adjacent ecosystems (IM)	% of allochthonous OM	< 5 %	5 to 20 %	20 to 40 %	40 to 60 %	> 60 %
Export of organic matter to adjacent ecosystems (EX)	% of PP	< 5 %	5 to 20 %	20 to 40 %	40 to 60 %	> 60 %
The carbonate cycle (CA)	g CaCO <sub>3</sub> m <sup>-2</sup> a <sup>-1</sup>	< 50	50 to 150	150 to 500	500 to 2 000	> 2 000

**Tab. 3: Characterisation of the ecosystems considered here: ecosystem descriptors.**

Ecosystem	SR-MPO	SR-M	SR-M	PPP	CPP	B-PPP	PP/B	LN-HC	HP	DP	CS	PP-SR	PP-RR	PP-SRR	B-SP	FW	TDC	IM	EX	CA
Marine ecosystems																				
<i>Posidonia oceanica</i> seagrass meadow	M	H	H	VH	VL	M	M	VH	L	H	VH	VL	VL	VH	M	M-H	M	L	M-H	L
<i>Cystoseira brachycarpa</i> forest	M	L	L	H	VL	L	M	VH	M	VL	VL	VL	VH	VL	M	L	M	VL	L	VL
Barren grounds - state <i>C. brachycarpa</i> forest	VL	VL	VL	L	VL	VL	M	VL	VH	VL	VL	VL	VH	VL	H	VL	VH	M	VL	H
<i>Macrocystis pyrifera</i> forest	L	M	M	VH	VL	L	M	VL	L	H	VL	VL	VH	VL	M	VH	VH	VH	H	L
Mediterranean coralligenous	M	M	M	L	VL	VL-L	M	VL	H	M	M	VL	VH	VL	H	M	H	H	VL	H
Sub littoral sea caves	VL	VL	VL	VL	L	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	VL	L
Coral reefs	M	VH	VH	VH	VL	VL	L	VH	VH	VL	VH	VL	VH	VL	VH	H	VH	L	L	VH
Hydrothermal vents	VL	L	L	VL	VH	VL	M	VL	L	M	VL	VL	VH	VL	H	L	L	L	L	L
Terrestrial ecosystems																				
Mediterranean maotral	L	L	L	VL	VL	M	L	VL	VL	H	VL	VH	VL	VL	VL	L	VL	VL	VL	VL
<i>Quercus ilex</i> evergreen oak forest	L	M	M	L	VL	VH	VL	VL	VL	VH	VL	VH	VL	VL	VL	L	VL	VL	VL	VL
Deciduous <i>Quercus</i> and <i>Fagus</i> forests	L-M	M-H	M	H	VL	VH	VL	VL	VL	VH	L	VH	VL	VL	VL	L	VL	VL	VL	VL
<i>Pinus laricio</i> pine tree forest	L	L-M	M	M	VL	H	L	VL	VL	VH	VL	VH	VL	VL	VL	L-M	VL	VL	VL	VL
Plain grasslands	M	H	H	H	VL	L	M	VL	H	L	VL	VH	VL	VL	L-M	M	M	L	L	VL
Tropical rain forest	VH	VH	VH	H-VH	VL	VH	VL-L	VL	VL	VH	L	VH	VL	VL	L-M	M	VL	VL	VL	L

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# **ORAL COMMUNICATIONS**

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# **COMMUNICATIONS ORALES**

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## **MARINE FORESTS AT RISK: SOLUTIONS TO HALT THE LOSS AND PROMOTE THE RECOVERY OF MEDITERRANEAN CANOPY-FORMING SEAWEEDS**

### **Abstract**

*Along Mediterranean coasts, canopy-forming seaweeds used to form diverse, productive and valuable “forest” habitats, but in the past decades conspicuous declines, sometimes to local extinction, have been reported in many regions. Canopies are retracting particularly close to urban areas, and are replaced by turf-forming and ephemeral algae or barrens. The persisting forests are under continued threat, and current protection measures are insufficient. We provide evidence that declines of canopy algae are dramatically extensive, and are driven by multiple local (nutrient enrichment and high sediment loads, fishing, heavy metal pollution) and global stressors (increasing temperature, high wave exposure). We also show that the combined management of local stressors (such as nutrients and sediments) would increase significantly the resilience of canopy algae to future climatic stressors, preventing their further deterioration. Finally, we discuss restoration prospects in areas where these systems have been lost. We conclude identifying the main needs to understand, guide and motivate effective conservation actions in these valuable ecosystems.*

**Key words:** Canopy algae, habitat loss, Mediterranean Sea, multiple threats, conservation

### **Introduction**

Along Mediterranean coasts, canopy-forming seaweeds (most frequently brown algae belonging to the order Fucales) form diverse, productive and valuable “forest” habitats. These habitats are becoming rare at local, regional and basin scales at an alarming rate (Airoldi & Beck, 2007). This is concerning because algal canopies play a key role in coastal primary production and nutrient cycling, and facilitate rich flora and fauna communities. In the past decades, algal canopies have suffered widespread and apparently irreversible loss, much of which may have gone unnoticed. Algal canopies are retracting particularly close to urban areas, and are replaced by turf-forming and ephemeral algae or sea urchin barrens, with major negative consequences for associated benthic and fish communities (Benedetti-Cecchi *et al.*, 2001). The persisting forests are under continued threat, and the benefits of current protection measures have been low.

We synthesize past research efforts aiming at quantifying the losses, and identifying what factors drive the loss or enhance the resilience of these systems. We also discuss the restoration prospects in areas where canopies have been lost and the main needs.

### **Materials and methods**

We reviewed published primary literature and summarized it in a table. The review is organized into three sections: 1) a compilation of data on historical loss of canopies along

Mediterranean coastlines and main drivers of loss; 2) a compilation of data on known factors enhancing resilience and restoration success; 3) a discussion of gaps in the data, ecological knowledge, and protection measures for these coastal habitats and recommendations for how to address these gaps.

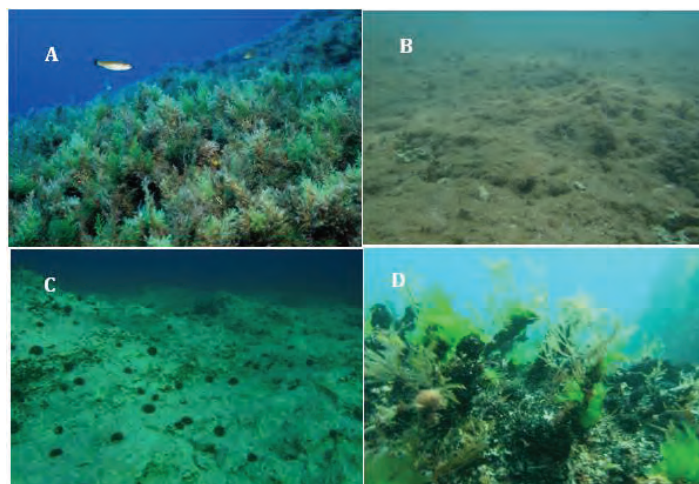
## Results

### Historical loss and main drivers

Conspicuous declines of algal canopies, sometimes to local extinction, have been reported in many regions along the coasts of Spain, France, Italy, Croatia, Albania, Greece and Turkey (Fig. 1 and Tab. 1). Along the Albères coast only 5 out of 14 species of Fucales (*Cystoseira* spp. and *Sargassum* spp.) documented as abundant in 1912 were present in 2003 (Thibaut *et al.*, 2005). Lost algal forests tend to be replaced by assemblages of lower structural complexity, such as turf-forming, filamentous or other ephemeral seaweeds, mussels or “barrens” (Mangialajo *et al.*, 2008; Connell *et al.*, 2014; Strain *et al.*, 2014). Canopy algae, turfs and barrens have been suggested to represent alternative states in shallow temperate rocky coasts under different disturbance and stress regimes (Airoldi *et al.*, 2009). There is a growing consensus and empirical evidence that these habitat shifts are driven by multiple anthropogenic stressors, including overfishing of higher trophic groups leading to outbreaks of grazers, eutrophication, excess sediment loads, coastal development, heavy metal pollution, point source pollutants such as oil spills, detergents and anti-fouling paints and invasive species (Table 1). These local anthropogenic stressors can interact negatively with environmental stressors or global climatic stressors (such as increasing temperature and CO<sub>2</sub>) resulting in accelerated declines of canopy-algae (Perkol-Finkel & Airoldi, 2010; Asnaghi *et al.*, 2013; Olabarria *et al.*, 2013; Strain *et al.*, 2014).

### Factors enhancing resilience or restoration efforts

While the proximate drivers of canopy loss are now relatively well understood, the factors that control the recovery have been more difficult to identify, and over a certain deterioration threshold, these systems may not be able to recover at all (Perkol-Finkel & Airoldi, 2010). The alternative habitat replacing lost canopies seems



**Fig. 1: Canopy algae characterise Mediterranean rocky coasts (e.g. A-*Cystoseira balearica* forest, Scandola, Corse), but many forests have been replaced by algal turfs (e.g. B- Haifa, Israel), urchin barrens (e.g. C- Porto Cesareo, Italy), or mussel beds (e.g. D - Monte Conero, Italy). Photographs by: A) E Ballesteros, B) L Airoldi, C) P Guidetti, D) L Airoldi**

to play a crucial role in controlling the return to a canopy dominated state once the stressors have been removed (Airoldi *et al.*, 2009). Thus, while there are examples of recovery of canopies from outbreak of urchins when effective protection measures have been established (*e.g.* Guidetti, 2006), when algal forests become replaced by turfs, sediments, or mussels it is not yet clear what prevents the recovery of the system, other than severe recruitment failure (Perkol-Finkel & Airoldi, 2010). Recent work has shown that management of water quality and particularly sediment loads is critical for ensuring the persistence of *Cystoseira* forests (Sales *et al.* 2011, Strain *et al.* unpub.). Reduction of nutrients would provide the greatest opportunity to prevent the shift from canopy to turf algae because of the prevalence of synergistic interactions between nutrients with other local and global stressors (Strain *et al.*, 2014). If depletion of forests has already occurred over wide areas, natural recovery could be slow or even implausible (Perkol-Finkel & Airoldi, 2010). The artificial restoration of fucoids in the Mediterranean Sea has been understudied compared to kelps and fucoids in other parts of the world, but results so far suggest it could be an effective strategy (Sales *et al.*, 2011; Perkol-Finkel *et al.*, 2012; Gianni *et al.*, 2013).

### Discussion

Mediterranean canopy forests are affected by many threats. The greatest impacts are associated with degraded water quality, coastal development, outbreaks of herbivores and invasive species, while effects of diseases and climate changes are uncertain. Current losses are alarming and protection is insufficient. Some key needs and opportunities for conservation and management are suggested below:

- 1) There is no comprehensive summary of the distribution of canopy forests, particularly deep sea ones, and their management is impeded by lack of knowledge on their status. Detailed habitat mapping should be given priority. The ecosystem services that these coastal habitats provide (such as nurseries for fisheries and recreation) also need to be better assessed to illustrate the costs of their loss and provide impetus and economic incentives for their protection and restoration.
- 2) An adequate evolutionary framework is needed to inform decisions on local and regional species diversity and to differentiate local extinction from species extinction. The loss of genetic biodiversity as populations undergo bottlenecks is also undescribed. New molecular tools need to be applied (RAD-seq) or developed (SNPs or microsatellites) to assess genetic diversity and link it to population resilience and ecosystem functioning, assess connectivity of populations, and study parentage and recruitment at local scales.
- 3) Like in other ecosystems (*e.g.* lakes, coral reefs, or forests) a gradual degradation of resilience paves the loss of these algal forests to alternative habitats, so that the mere restoration of environmental conditions preceding the loss may be insufficient to restore the system (Perkol-Finkel & Airoldi, 2010). Strategies for conservation of canopy forests should focus on “early-warning signals” of approaching shifts and on effective and rapid management of local stressors to maintain resilience in face of global stressors. This knowledge is presently limited for Mediterranean canopy forests, but results so far suggest that management of water quality and sediment loads would provide some of the greatest opportunities, particularly in enclosed bays or estuaries.

**Tab. 1: Selected studies reporting the loss (as either percentage area lost and/or number of species lost) of canopy-forming algae in the Mediterranean Sea, suggested drivers of loss, and signs of recovery if observed. Na= no quantitative data reported**

Lost species	Geographic location	Amount of loss	Driver of loss	Alternative habitat	Signs of recovery	References
<i>Cystoseira</i> spp. and <i>Sargassum acinarium</i> (as <i>S. linifolium</i> )	Monte Conero (Italy, Central Adriatic Sea)	90% (6 out of 8 sp lost)	High sediment loads/poor water quality, increased substratum instability, increased storminess	Turfs or mussels	no	(Romagnoli & Solazzi, 2003; Irving <i>et al.</i> , 2009; Perkol-Finkel & Airoldi, 2010)
<i>Cystoseira</i> spp. and <i>Sargassum</i> spp	Albères Coast (France, NW Mediterranean)	80% (9 out of 14 sp lost)	Poor water quality, overgrazing by urchins, coastal development, human trampling	Turfs, mussels or urchin barrens	no	(Thibaut <i>et al.</i> , 2005)
<i>Cystoseira</i> spp. and <i>Fucus virsoides</i>	Istrian coast (Croatia, North Adriatic Sea)	11 out of 15 sp lost	Poor water quality, overgrazing by urchins	Ephemeral algae	yes	(Munda, 1979, 2000; Zavodnik <i>et al.</i> , 2002)
<i>Cystoseira</i> spp	Genova (Italy, Ligurian Sea)	>50%	Coastal urbanisation	Algal turfs	no	(Mangialajo <i>et al.</i> , 2008)
<i>Cystoseira</i> spp	Albania	90%	Coastal development and urbanisation			(Fraschetti <i>et al.</i> , 2011)
<i>Cystoseira</i> spp. and <i>Sargassum</i> spp	Tremiti islands (Italy, South Adriatic Sea)	na	Poor water quality, overgrazing by urchins	Ephemeral algae or urchin barrens	yes	(Cormaci & Furnari, 1999; Fraschetti <i>et al.</i> , 2012)
<i>Cystoseira</i> spp	Salento Peninsula, (South Adriatic and Ionian Sea)	90%	Overgrazing by urchins	Urchin barrens	yes	(Guidetti <i>et al.</i> , 2003; Guidetti, 2006)
<i>Cystoseira</i> spp and <i>Sargassum</i> spp	Linosa Island (Italy, Sicily Channel)	60% (10 out of 15 sp lost)	Increased water temperature and changes in water circulation	Turfs and ephemeral algae	no	(Serio <i>et al.</i> , 2006)
<i>Cystoseira</i> spp	Several locations (Italy, NW Mediterranean)	90%	Coastal urbanisation, poor water quality, high sediment loads	Turfs	no	(Benedetti-Cecchi <i>et al.</i> , 2001)
<i>Cystoseira</i> spp	Several locations, (Greece and Turkey, Southern Aegean Sea)	>90%	Invasive fish	Urchin-less barrens	no	(Sala <i>et al.</i> , 2011; Giakoumi, 2014)
<i>Cystoseira crinita</i>	Cap Corse (France, NW Mediterranean)	7%	Coastal development	Turfs	no	(Sales & Ballesteros, 2010)
<i>Cystoseira crinita</i> , <i>C. barbata</i>	Maó harbour, Menorca, (Spain, Balearic Sea)	100%	Poor water quality, heavy metal pollution, and aquaculture	Ephemeral algae	Yes after transplant	(Sales <i>et al.</i> , 2011)
<i>Cystoseira zosteroides</i>	Medes Islands, (Spain, NW Mediterranean)	90%	Exceptional storm	Turfs	yes	(Navarro <i>et al.</i> , 2011)



4) Fucoids show high reproductive potential but low dispersal, which limits their natural recovery of wide lost/degraded areas. Given the extent of damage, restoration will be required in many places to meet any reasonable goals for conservation and management. Artificial restoration of *Cystoseira* forests in the Mediterranean Sea is much behind compared to other systems (i.e. seagrass beds), and much more work is needed to develop effective tools and approaches (Gianni *et al.*, 2013).

There are still opportunities for conservation of Mediterranean canopy forests. This protection should be achieved quickly because conservation is cheaper than restoration. Reducing cumulative local human impacts would represent the most effective strategy for the conservation and recovery of these systems, but, whenever this alone cannot reverse the loss, well-designed restoration projects can assist. Overall, there should be greater public, political and even scientific awareness of the extent, importance, and consequences of the loss of canopy forests, and greater commitment to motivate serious conservation and restoration actions in these highly threatened ecosystems.

### Acknowledgments

The study was supported by project TETRIS (Prin 2010-2011, MIUR).

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## **ASSESSMENT AND QUANTIFICATION OF THE ANTHROPIC IMPACT ON THE *POSIDONIA OCEANICA* SEAGRASS MEADOW**

### **Abstract**

*The regression of seagrass meadows has been extensively studied over the past few years. Although the causes of this regression may be locally natural, it is more generally related to human impact.*

*In the framework of the HalGolo (2010) and CoralCorse (2013) oceanographic campaigns, acoustic data (mosaic of sonograms and bathymetry), validated by field data (Scuba diving, ROV), were acquired at depths of -10 m and -50 m at the NATURA 2000 site “Grand Herbier de la Plaine Orientale” (Western Mediterranean, Corsica). Processing of this data provided evidence of the scale of this mechanical degradation (trawling scars, mooring, etc.), and enabled its quantification with regard to surface area and scar density. The main degradation was observed between -20 and -40 m depth (98%); the surface area of seagrass meadow destroyed is estimated at 280 ha with more than 40 scars per hectare recorded in the northern part of the site. Given the slow growth rate of the meadow, assuming the hypothesis of the ending of these practices, it would require almost 150 years to recover these scars.*

**Key-words:** Anthropogenic impact, Corsica, Side Scan Sonar, *Posidonia oceanica*, trawling.

### **Introduction**

The ecological, economic and heritage importance of the *Posidonia oceanica* seagrass meadow in the Mediterranean, and notably in Corsica, has been extensively demonstrated over several decades (see synthesis in Boudouresque *et al.*, 2012). Its importance has resulted in the introduction of conservation measures for this ecosystem at regional and / or national scale (UNEP-MAP-RAC/SPA, 1999; Boudouresque *et al.*, 2012). Benthic mapping constitutes an essential element in this conservation strategy (reference state, patterns of change over time) and Corsica may be seen as the precursor in this field since all of its *Posidonia* meadows have been mapped since the beginning of the 1990s (Pasqualini *et al.*, 1998) and monitoring over time has been carried out there, at representative sites, since 2004 (Bein *et al.*, 2013; Pergent *et al.*, 2007).

During the cartographical surveys undertaken between 2010 and 2013, numerous anthropic scars were evidenced, in particular at the “Grand Herbier de la Plaine Orientale” - Zone NATURA 2000 FR 9402014 site. Most of these scars would appear to be of mechanical origin and correspond to the action of benthic trawling and the mooring of large vessels off a petroleum facility. Although the legislation in force should limit the impact of trawling on the meadow (distance from the coastline, depth; CE, 2006), it is apparent that it is rarely applied in many Mediterranean countries, which results in the significant degradation of the *Posidonia oceanica* meadows, with in particular the uprooting of the leaf shoots and rhizomes, the suspension of particles, the alteration of the benthic communities in favour of opportunistic species and the reduction of the diversity and abundance of the fauna (Pergent *et al.*, 2013).

The automatic quantification of this impact, on the basis of the available acoustic data (e.g. side-scan sonar), comes up against methodological problems and is often limited to manual interpolation over limited sectors (Pasqualini *et al.*, 2000; Ramos-Esplas *et al.*, 1994). These measurements involve mainly the assessment of the surface areas covered by the dead mat or of the soft sediment in relation to the surface area of living seagrass, even if other factors, whether natural or anthropic, may act in synergy and be partly responsible for the absence of the seagrass meadow (Boudouresque *et al.*, 2009).

The aim of the present study was therefore to test a new method for optimising the identification, characterisation and quantification of the anthropic scars on the test zone of the “Grand Herbier de la Plaine Orientale”.

### **Materials and methods**

The study site extends from the sea outlet channel of the Biguglia lagoon to south of the mouth of the Golo, on the east coast of Corsica. It was chosen because of its interest in terms of conservation, the availability of complete coverage by acoustical data and the occurrence there of trawl fishing.

The identification and the characterisation of the anthropic mechanical scars are based on the direct interpretation of sonograms (depth range 10 – 50 m) and on the map of habitats (Bonacorsi *et al.*, *in press*). This acoustic imaging was obtained by means of a Klein 3000 side scan sonar (range 100 m and frequency 100 KHz – CoralCorse campaign), and by an interferometric sonar (frequency 250 KHz – Halgolo campaign). The depth was obtained using the same device (Halgolo campaign - frequency 250 KHz) and by a Simrad EM 2040 multibeam echosounder (CoralCorse campaign - frequency 300 KHz). The absolute decimetric position was determined using the DGPS system.

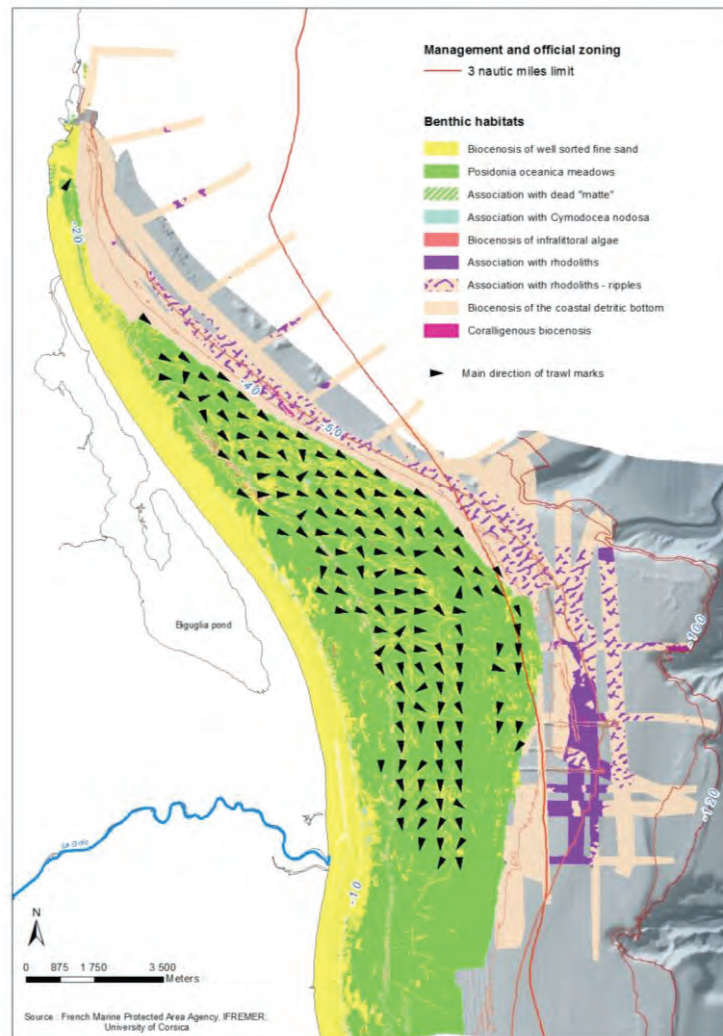
The raw data are processed with the Caraibes software (@ IFREMER). The imaging data are corrected (suppression of the blind band), correction of obliquity errors, gain homogenisation, contrast enhancement and correction of the geographical position). The mosaics are integrated in a Geographical Information System (GIS - ArcGIS 10.0) at a resolution of 0.5 m. The bathymetric data are corrected by filtering and manual sounding invalidation. The MNT is meshed at 5 m resolution and integrated under GIS. Raster processing (shading, focal statistics and algebra) is used to improve the visualisation of the relief. Analysis of the anthropic scar density is performed, under GIS, by the setting of a grid of points every 200 m (Smith *et al.*, 2007). For each point, within a 200 x 200 m window, an operator counts the number of scars observed and gives their dominant orientation. The points recorded by side scan sonar are weighted by a higher weighting than those recorded by interferometric sonar, which only takes into account the largest scars.

An interpolation by linear kriging is then performed in order to obtain a map of the scar density. The number of scars per point is discretised in seven classes according to the natural threshold method (Jenks method), which best fits with the statistical series. An analysis of the areas impacted is then carried out on the basis of a manual map (1/1000) of the scars on the sonograms, the scar object being considered as a polygon.

A theoretical estimation of the resilience capacity of the *Posidonia oceanica* meadow is calculated by the application of buffer zones corresponding to time periods of 10 to 150 years on the basis of a growth rate of 4 cm.year<sup>-1</sup> and the calculation of the corresponding surface areas.

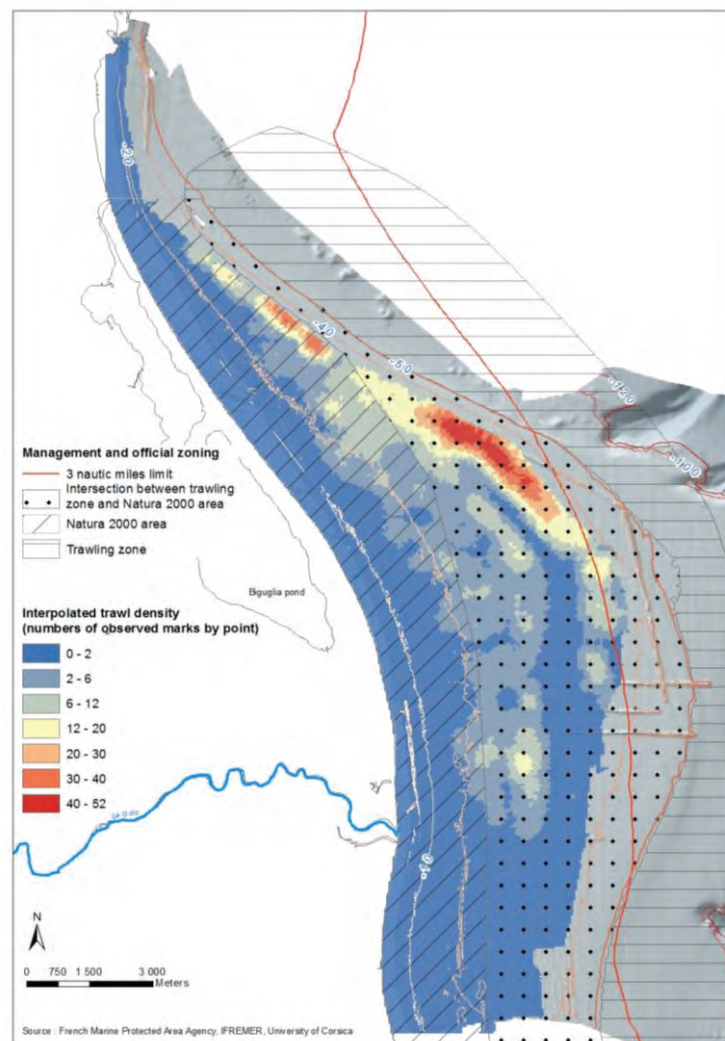
## Results

The map of the main habitats and types of bottom provides a basis for visualising the distribution of the anthropic impact within the *Posidonia oceanica* meadow (Fig. 1). Faint scars (probably old) and sharp scars, no doubt more recent, with a characteristic acoustic aspect, co-exist throughout the area.



**Fig. 1: Benthic habitats map and orientation of main trawling scars**

Several of these scars are more than two kilometers long. In the surface zones (down to 20 m depth), there are fewer anthropic scars. The trawling scars are mainly oriented parallel with the coastline (east-south/east in the northern sector, then south). The surface area of *Posidonia oceanica* meadow ‘destroyed’ is estimated at 280 hectares of the 7 258 hectares covered by the meadow, or almost 4% for the whole of the sector studied. The interpolated density of trawling scars (Fig. 2) shows that most of these scars are situated between -20 and -40 m depth (98%), with a concentration in the north-east sector and greater sparseness to the south. In the most highly impacted sectors, more than 10 scars per hectare were identified (Fig. 2).



**Fig. 2: Interpolated trawl density map (in a 200 x 200 m grid)**

On the hypothesis of the ending of these practices, the complete regeneration of the *Posidonia* meadow is estimated at 150 years, even if the first decade would enable a recovery of more than 30%, allowing the recolonisation of the narrowest scars.

### **Discussion and conclusions**

In the light of our results, the effectiveness of acoustic sensors in characterising the mechanical scars within the *Posidonia oceanica* meadow is clearly demonstrated, while side scan sonar would appear to be more effective than interferometric sonar. Nevertheless, the strategy of acquisition may play a major role in the quantification of the scars in that they are less easily detected when they are perpendicular to the direction of navigation (Coggan *et al.*, 2001). Similarly, the speed of the boat, the frequency of the acoustic impulse and the position of the towfish above the bottom are all parameters that require optimisation (Humborstad *et al.*, 2004). Carrying out a continuous video survey, in parallel with the acoustic survey, would also offer a means to better characterise the scars (Humborstad *et al.*, 2004 ; Smith *et al.*, 2007).

The method chosen to analyse the scar density, while faster and more representative than the ‘destroyed areas’ methods of assessment, requires improvement in order to attempt to overcome the bias inherent in the subjectivity of the operators. An automatic scar surface



area analysis would complete the density values and provide a better characterisation of the phenomenon. The first trials along these lines, carried out on small areas, are encouraging (Pasqualini *et al.*, 2000) and should be pursued, in particular with the use of a ‘canny filter’ contour detection algorithm. Nevertheless, the discrimination between the different types of anthropic scars (anchoring, trawling) and ‘natural destruction’ (e.g. intermattes) merits particular attention that is poorly compatible with automatic methods.

This work has made available a first assessment of mechanical anthropic impact, in particular trawling pressure, on one sector of the Zone NATURA 2000 site of the “Grand Herbier de la Plaine Orientale”, and has provided a basis for identifying the worst affected areas. Of the 7 258 hectares covered by the *Posidonia oceanica* meadow, 6 ha have been destroyed between 0 and -20 m depth and 274 ha between -20 and -40 m. Even if the data available for other sectors of the Corsican littoral would appear to show much higher values, between 12% (Pasqualini *et al.*, 2000) and 38% (Bonacorsi, 2012), they only concern very limited areas (a few tens of hectares). However, in the Gulf of Gabes, Tunisia (Zaouali 1993) and in the region of Alicante, Spain (Ramos-Esplas *et al.* 1994), trawling may be responsible for the loss of extensive areas of *Posidonia* meadows (respectively 80% and 50%).

The rate of recolonisation, and therefore of the resilience of the *Posidonia oceanica* meadow to mechanical damage, constitutes an essential element in terms of conservation management, and even of the natural restoration of this ecosystem. The mean growth rate values used here are only given as an indication, and only growth rate measurements *in situ* in the sector where the impact no longer occurs would provide a suitable basis for recalibrating the model. Because of the degradation of the substrate (matte) and of the introduction of replacement species (e.g. *Caulerpa racemosa* var *cylindracea*), the rate of recolonisation might be much slower (Delgado *et al.*, 1999; Kiparissis *et al.*, 2011).

### Acknowledgments

This research is a part of the University of Corsica and the Collectivité Territoriale de Corse CHANGE programme (FRES 3041). This work would not have been possible without the support of the N/O L'Europe (Ifremer-Genavir-Insu/UMS Flotte Océanographique Française), the efficiency of the crew of the Oceanographic vessel, and the financial support of the Direction Régionales de l'Environnement, de l'Aménagement et du Logement de Corse.

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## **IMPACT DE LA PÊCHE PAR MINI-CHALUT BENTHIQUE SUR LES HERBIERS À *POSIDONIA OCEANICA* DANS LE SECTEUR NORD-EST DES ÎLES KERKENNAH (TUNISIE)**

### **Résumé**

*Le présent travail est une contribution à l'évaluation de l'impact d'une pratique de pêche illicite, la pêche au « Kiss », sur les herbiers à *Posidonia oceanica* dans le secteur Nord-Est des îles Kerkennah. Dans cette zone l'herbier constitué par la Magnoliophyte marine *P. oceanica* est très bien développé et présente différentes structures assez particulières notamment l'herbier tigré, une formation typique de la région. Ces herbiers, très importants pour l'équilibre écologique de la zone côtière sont menacés par le chalutage benthique. Dans ce travail les caractéristiques de l'herbier (recouvrement, densité des faisceaux, biométrie et biomasse foliaire) sont décrites dans un secteur impacté par le chalutage et dans un site de référence afin d'évaluer l'incidence de cette pratique de pêche. Cette première étude montre que c'est surtout le recouvrement de l'herbier qui est affecté par le chalutage benthique.*

**Key-words:** *Posidonia oceanica*, Seagrass, Kerkennah islands, Fishing, Trawling (kiss).

### **Introduction**

Les îles Kerkennah appartiennent à la côte Nord du Golfe de Gabès et ne sont que la partie émergée d'un vaste plateau sous-marin dont les hauts fonds s'étendent de 9 à 50 km autour de cet archipel. L'herbier à *Posidonia oceanica* (L.) Delile représente une composante majeure de l'écosystème littoral des îles Kerkennah. Il forme localement une structure unique appelée « Herbier tigré » ; cette structure, décrite pour la première fois autour des îles Kerkennah, est considéré comme la plus importante et la mieux développée en Méditerranée (Burolet, 1983 ; Pergent *et al.*, 2010).

L'herbier à *Posidonia oceanica* des îles Kerkennah couvre des surfaces très importantes du fait de la faible déclivité des fonds, il est présent à plus de 15 km du littoral. Malgré son importance les travaux réalisés sur *P. oceanica* dans cette zone ont été fragmentaires portant sur la répartition (Burolet, 1983); la floraison (Pergent *et al.*, 1989); ou bien, sur l'état de l'herbier (Ben Mustapha et Hattour, 1992 ; PNUE-PAM-CAR/ASP, 2009). Cet écosystème subit des grandes pressions anthropiques (Boudouresque *et al.*, 2009) dues surtout au chalutage par à un engin de pêche illégal, un mini chalut benthique, connu localement sous le nom du "kiss".

Dans le but de mieux connaître l'état des herbiers à *P. oceanica* dans la région des îles Kerkennah et pour évaluer les dégâts dus au chalutage, une première évaluation de l'impact du "kiss" sur les herbiers à *Posidonia oceanica* est réalisée.

### **Matériel et méthodes**

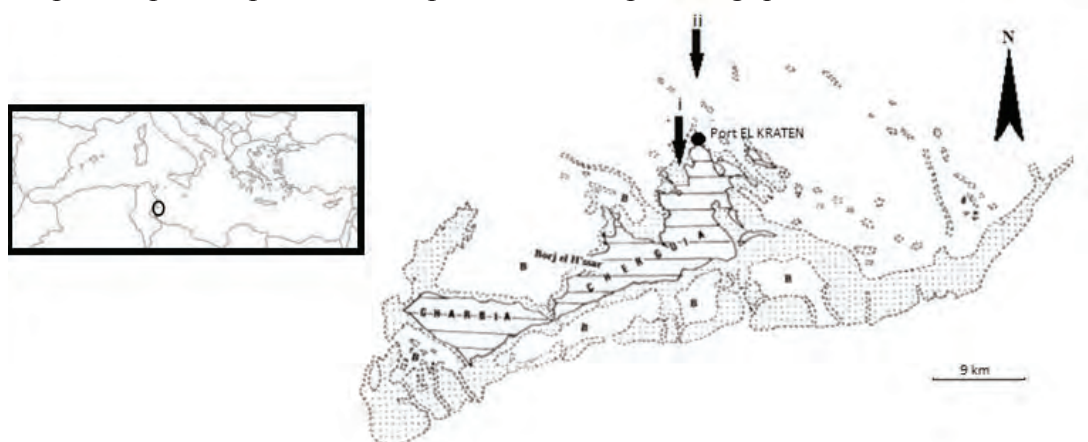
Pour mieux comprendre le déroulement du chalutage par le "kiss" et en raison de la réticence des pêcheurs à répondre aux questions à cause de la sensibilité du sujet (pratique illégale), nous avons participé à une campagne de chalutage afin de constater de *visu* les dégâts de cet engin.



L'aire concernée par l'étude de l'herbier se trouve au Nord de l'Ile Gharbi (Fig. 1). Deux sites ont été retenus pour l'évaluation de l'impact du "kiss":

- un premier site de référence (i) non impacté par la pêche au "kiss" situé à  $34^{\circ}48'35''$  de Latitude Nord et  $11^{\circ}14'14''$  de Longitude Est à 350 m du rivage et à une profondeur de 2 m. Il est localisé dans la zone concernée par le projet MedMPAnet pour la création d'une aire marine protégée. Dans cette station (i), *Posidonia oceanica* forme un herbier tigré.
- un deuxième site impacté (ii) situé à  $34^{\circ}53'53,63''$  de Latitude Nord et  $11^{\circ}15'30,19''$  de Longitude Est. Ce site (ii) se situe à 7 km au nord du port de Kraten à 3 m de profondeur où *Posidonia oceanica* forme un herbier sur mat qui est soumis à une forte pression de pêche au "kiss", particulièrement en hiver.

La localisation des stations a été déterminée à l'aide d'un GPS portable "GARMIN GPS 72 H". Pour chaque site on a relevé *in situ*, en plongée en scaphandre autonome, le recouvrement et la densité des faisceaux de *Posidonia oceanica*. Ces mesures *in situ* sont complétées par des prélèvements pour une étude phénologique.

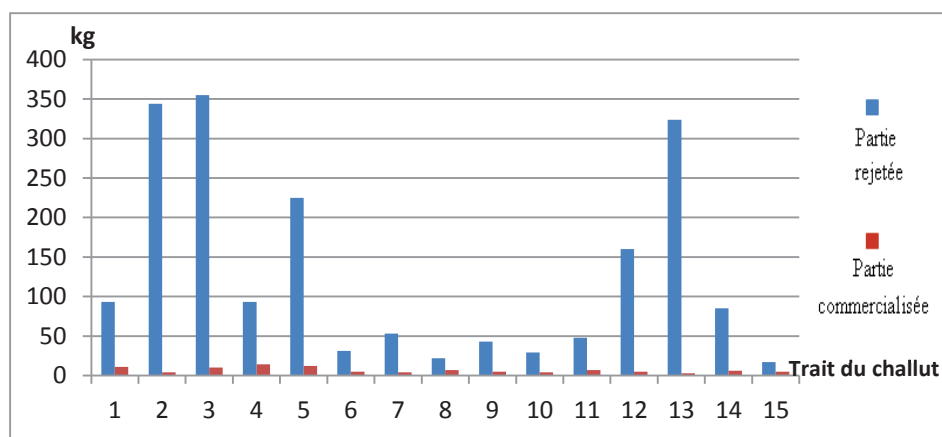


**Fig.1 : Emplacement des sites d'études (i : Site Référence ; ii : Site Impacté).**

A l'aide d'un logiciel de traitement d'image (ENVI 4.7) et une prospection sur terrain (vérité terrain), une cartographie de l'ensemble de la zone d'étude a été réalisée afin de montrer l'interférence entre les zones de présence d'herbiers à *Posidonia oceanica* et la zone de pêche au "kiss". Au niveau de la zone impactée, le nombre des traces de chalutage a été calculé le long d'une bande de 100 m de long et de 25 m de large, soit une surface de 2500 m<sup>2</sup>.

### Résultats

Les biomasses totales retenues (poids humide) dans les 15 traits de chalut effectués pendant la campagne de pêche montrent une grande différence entre la partie commercialisable, qui ne dépasse pas 15 kg par trait de chalut, et celle rejetée (atteignant jusqu'à 355 kg par trait du chalut) (Fig. 2). La partie rejetée correspond à plusieurs espèces benthiques (spongiaires, échinodermes, mollusques, etc.) mais 68% des rejets sont formés par *Posidonia oceanica* (feuilles, rhizomes, racines) qui constitue la plus grande biomasse.



**Fig.2 : Biomasses retenues dans 15 traits de chalut pendant la campagne de pêche par le "kiss" (Avril 2014).**

#### Variation du recouvrement de l'herbier à *Posidonia oceanica*

Au niveau du site de référence (site non impacté), le recouvrement de l'herbier est assez faible et ne dépasse pas 50%, du fait de la structure de l'herbier (tigré). Il est de 60%, dans le site impacté qui correspond pourtant à un herbier continu. 40% de la couverture de l'herbier est formée de mattes mortes perdues à cause des panneaux divergents utilisés pour garantir l'ouverture latérale du chalut ou "kiss". Les traces apparaissent sous forme des sillons au milieu de l'herbier qui ont les mêmes largeurs et sont d'étendues différentes. Au niveau de la zone impactée, la surface dégradée par le "kiss" est estimée à 630 m<sup>2</sup>, ce qui correspond à 25,2% de la surface totale considérée. Dans les traces du chalut, *Posidonia oceanica* tend à recoloniser la surface perdue, mais elle est en compétition avec d'autres macrophytes, dont certaines sont des espèces exotiques à caractère invasif (ex. *Caulerpa racemosa* var. *cylindracea*) ce qui risque de causer encore plus de perturbations à l'écosystème.

#### Variation de la densité des faisceaux

Pour la station de référence, la densité des faisceaux de feuilles de *Posidonia oceanica* est estimée à 594 ± 36 faisceaux.m<sup>-2</sup>. Cet herbier est classé parmi les herbiers de type II, herbier dense selon Giraud (1977) mais présente une densité « médiocre » pour cette profondeur (PNUE-PAM-CAR/ASP, 2011).

En ce qui concerne la station impactée, l'herbier présente une densité de 728 ± 38 faisceaux/m<sup>2</sup> ; il correspond également à un herbier de type I ou herbier dense selon Giraud (1977) mais sa densité est considérée comme normale (PNUE-PAM-CAR/ASP, 2011).

#### Phénologie

Les différents indices phénologiques et biométriques sont consignés dans le tableau 1.

**Tab. 1 : Indices phénologiques et biométriques de l'herbier de posidonie calculés dans le site référence et le site impacté par le chalutage benthique dans la région des îles Kerkennah (moyenne  $\pm$  intervalle de confiance).**

	Site Référence (SR)	Site Impacté par le chalutage (SI)
Nombre moyen des feuilles par faisceaux	6,1 $\pm$ 0,5 (Ad=3,2/Id=2,4)	6,2 $\pm$ 0,5 (Ad=3,4/ Id=2,6)
Longueur moyenne des feuilles adultes (cm)	40,9 $\pm$ 4,0	60,3 $\pm$ 5,1
Largeur moyenne des feuilles adultes (cm)	0,87 $\pm$ 0,01	0,97 $\pm$ 0,01
Longueur moyenne des feuilles intermédiaires (cm)	42,2 $\pm$ 5,1	65,9 $\pm$ 7,0
Largeur moyenne des feuilles intermédiaires (cm)	0,89 $\pm$ 0,02	0,94 $\pm$ 0,02
Longueur moyenne des pétioles (cm)	3,8 $\pm$ 0,3	4,6 $\pm$ 0,3
Coefficient A	51,6	43,2
LAI (m <sup>2</sup> /m <sup>2</sup> )	2,2	4,3
Biomasse foliaire totale (g,Ms.m <sup>-2</sup> )	611,4 $\pm$ 78,0	1286,4 $\pm$ 104,9
Biomasse épiphytique (g,Ms.m <sup>-2</sup> )	149,0	51,2

LAI = Indice foliaire global (Leaf Area Index) correspond à la surface foliaire par m<sup>2</sup> de l'herbier à *Posidonia oceanica* (Boudouresque, 2006).

Coefficient A = pourcentage de feuilles cassées (par l'hydrodynamisme) ou broutées (oursins et poissons principalement), (Boudouresque, 2006).

#### Cartographie des herbiers

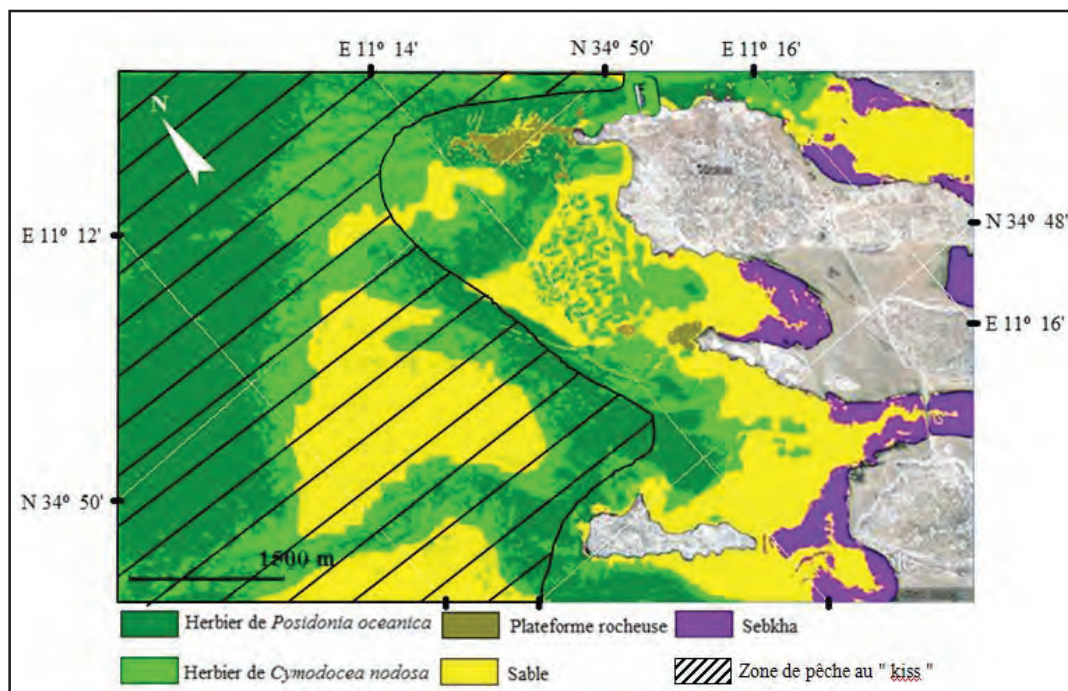
La carte des principaux peuplements et types de fonds (Fig. 3) montre l'interférence entre les zones de présence d'herbiers à *Posidonia oceanica* et la zone de pêche au "kiss". Malheureusement, une grande surface couverte par l'herbier est située dans la zone d'action du chalutage benthique par le "kiss". C'est grâce à la très faible profondeur (<1,5m à marée haute) et/ou la présence d'une plateforme rocheuse qu'on peut trouver des zones épargnées de l'action du mini chalut benthique. Ces conditions semblent être assurées dans les zones de prolifération de l'herbier tigré de Kerkennah.

#### **Discussion et conclusion**

Notre participation à une campagne de pêche au mini chalut benthique ou "kiss", nous a permis de constater que cette technique est à la fois destructive et inefficace, vue la faible production, et ne permet pas une exploitation durable des ressources de la région.

Le recouvrement et la densité des faisceaux de *Posidonia oceanica* au niveau de la station de référence montrent des valeurs relativement faibles, vraisemblablement liée au type d'herbier présent (tigré). Cependant, l'absence de matte morte dans la plupart des stations de la zone de référence indique que l'herbier à *Posidonia oceanica*, n'est pas soumis à des impacts anthropiques importants, ce qui confirme le travail de Ramos-Espla *et al.*, (2000). Mais cela n'est pas le cas pour l'herbier du site impacté où nous avons relevé une grande

fréquence des mattes mortes sous forme de traces dues au chalutage à une profondeur ne dépassant pas 3 m à marée haute.



**Fig.3 : Cartographie des principaux peuplements et types de fonds dans la région des îles Kerkennah (Avril 2014).**

Bien que les résultats de l'étude phénologique puissent paraître en discordance avec ceux du recouvrement, ils peuvent raisonnablement s'expliquer par les conditions écologiques naturelles du milieu, en effet le site de référence est très près de la côte donc il subit l'effet de la marée et de la profondeur qui vont avoir un rôle important sur les caractéristiques phénologiques de l'herbier.

Il apparaît donc que le recouvrement est le paramètre le plus significatif pour décrire les dégâts dus au passage des panneaux divergents des mini chaluts benthiques. Par contre d'après les résultats de notre étude, les paramètres phénologiques et biométriques, qui représentent des meilleures valeurs au niveau du site impacté, ne sont pas des bons indicateurs de l'impact du "kiss" sur l'herbier.

Il est vrai que cette pratique de pêche illicite réduit le recouvrement de l'herbier, mais le problème est beaucoup plus grave, parce que les traces de chalutages sont des terrains très favorables à l'installation d'autres espèces phytobenthiques et favorisent l'installation d'espèces exotiques.

Beaucoup de mesures ont été prises pour lutter contre la pratique de la pêche au "kiss" mais paraissent incapables de ralentir son évolution continue et rapide. La création d'une aire marine protégée dans la région des îles Kerkennah, la sensibilisation des pêcheurs de la région aux problèmes de l'environnement marin et leur implication dans les actions de protection et de surveillance de la future réserve marine nous paraissent des actions importantes à entreprendre d'urgence pour la protection des ressources marines de la région.

### Remerciement

Dans ce travail nous avons bénéficié de l'appui du Centre d'Activités Régionales pour les Aires Spécialement Protégées du Plan d'Action pour la Méditerranée (CAR/ASP).

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## **APPLICATION DE LA SISMIQUE UHR POUR LE SUIVI DE L'ETAT DE CONSERVATION DES HERBIERS A *POSIDONIA OCEANICA***

### **Résumé**

*La sismique réflexion Ultra Haute Résolution (UHR) est une technique récente particulièrement adaptée à la cartographie et la caractérisation des fonds marins. Elle a été utilisée avec succès par Claudio Lo Iacono et al., 2008 dans la baie de Port-Iligat (Espagne) pour cartographier la matte et l'herbier à *Posidonia oceanica* (posidonies). Cette même technique a été appliquée pour cartographier et caractériser l'herbier de posidonies et la matte morte dans l'Aire Marine Protégée (AMP) de la côte agathoise (France) en Méditerranée Nord occidentale. La sismique UHR a permis d'imager la structure et le substrat sur lequel l'herbier de posidonies se développe. L'épaisseur de la matte sous herbier, la présence et l'épaisseur de mattes mortes et le type de substrat sur lequel repose l'herbier ont été estimés à partir des profils sismiques. A l'échelle de l'AMP, la couche de matte morte est homogène et relativement fine, soit comprise entre 20 et 50 cm. La faible épaisseur de mattes mortes sur le site confirmerait les processus dynamiques rapides de développement et de disparition des herbiers. Ainsi, le rapport matte morte sur herbier vivant, bien qu'élevé sur le site, ne constituerait pas un phénomène alarmant de disparition de l'herbier, mais un développement naturel pour ce type d'herbier confronté à de forts phénomènes d'hydrodynamisme.*

**Mots clés :** Cartographie, sismique UHR, *Posidonia oceanica*, mattes.

### **Contexte**

La sismique UHR se place en complément des techniques acoustiques de cartographie sous-marine (les sondeurs mono- et multifaisceaux, les sonars latéraux) (Blouet *et al.*, 2011). La prospection sismique permet d'imager la structure de l'herbier de magnoliophytes et le type de substrat sur lequel il se développe. Basée sur l'utilisation d'un sondeur d'une résolution verticale centimétrique et d'une pénétration dans les sédiments de plusieurs mètres (en fonction de la nature des sédiments), cette méthode permet de distinguer les faisceaux de feuilles, l'épaisseur de la matte (ensemble constitué par les rhizomes, les racines et par le sédiment qui colmate les interstices) et la nature du substrat. La première application sur l'herbier de posidonies (*Posidonia oceanica*) a été réalisée par Lo Iacono *et al.* (2008) dans la baie de Port-Iligat (Espagne) afin de caractériser l'épaisseur des mattes et d'estimer la matière organique accumulée.

Cette étude a pour objectif d'associer la sismique à haute résolution (UHR) à l'ensemble de la chaîne d'acquisition acoustique afin de caractériser la matte morte de *P. oceanica* dans l'aire marine protégée (AMP) de la côte agathoise (Agde, France). Les données de hauteur de matte morte constitueront une première étape dans la compréhension du développement de *P. oceanica* sur le site mais également sur l'hydrodynamisme pouvant affecter l'état de l'herbier.

### **Méthode**

Lors de cette étude, différents instruments ont été déployés simultanément permettant la collecte de données bathymétriques et de réponse acoustique du fond marin. Toutes ces données ont une précision extrêmement fiable puisqu'elles sont géo-référencées à l'aide d'un GPS RTK



centimétrique. L'attitude du bateau, c'est-à-dire son inclinaison (lacet, roulis et tangage) est connue en permanence grâce à une centrale inertielle.

Les moyens suivants sont opérés simultanément à partir d'un mini-navire océanographique dédié à la reconnaissance des fonds marins :

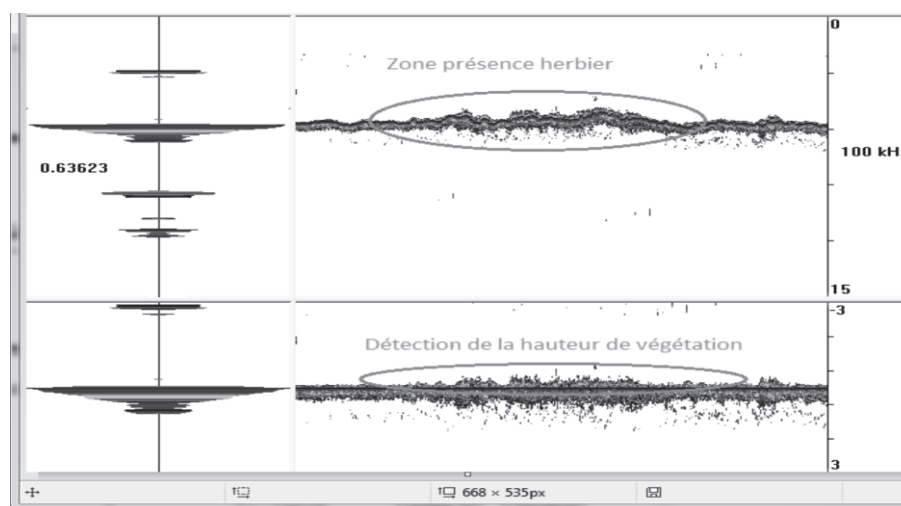
- Un sonar latéral interféromètre (GEOSWATH) à 250 kHz : il réalise l'imagerie sonar latéral des sites peu profonds en même temps que la bathymétrie multifaisceaux,
- Un sonar latéral (KLEIN 3900) à 900 kHz pour l'acquisition d'une mosaïque sonar latéral haute résolution
- Un sondeur de sédiment INNOMAR SES 2000 compact bi-fréquence (100 et 10 kHz).

Le sondeur de sédiment INNOMAR SES 2000 est fourni avec un logiciel d'acquisition permettant le pilotage de l'électronique. Il permet de régler entre autres la fréquence d'acquisition, la récurrence des émissions et permet l'enregistrement des données brutes acoustiques.

### Résultats

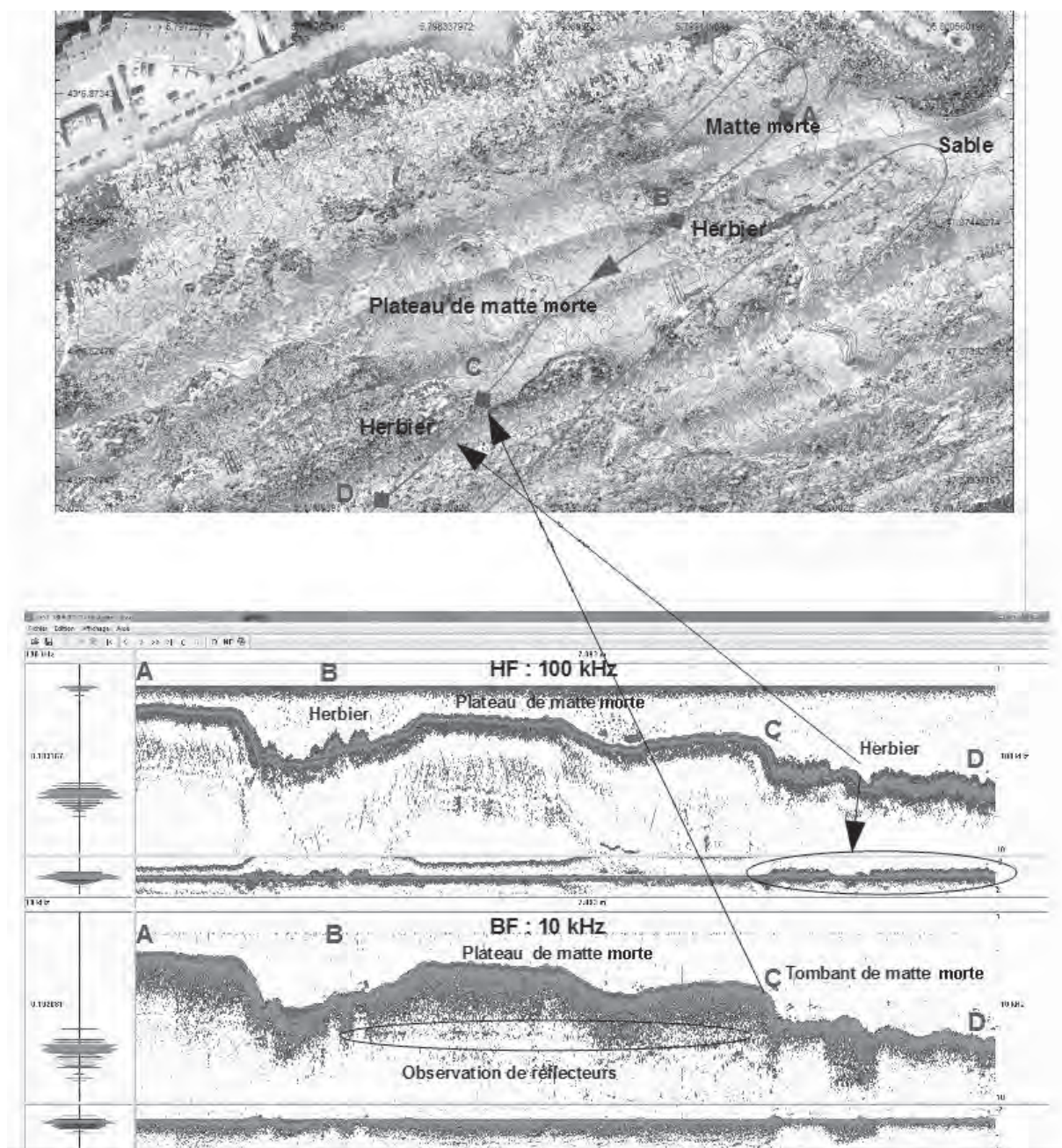
Une première phase de travail réalisée par SEMANTIC TS a consisté à développer des routines de traitement des signaux enregistrés par l'INNOMAR. Cette phase était nécessaire, dans la mesure où l'objectif était de fusionner les données sondeur de sédiments avec les données multi-capteurs acquises antérieurement. Il était donc impératif de maîtriser la chaîne de traitement et de géo-localisation des signaux d'une part, et d'autre part de disposer de fonctionnalités de graphisme et d'affichage spécifiques aux besoins de ce projet.

Le logiciel SIVA (logiciel développé par SEMANTIC TS) recalcule automatiquement la profondeur à partir des données brutes à l'aide d'algorithmes plus évolués dans le cas de l'acquisition de signaux en présence de végétation sous-marine. Un exemple (Fig. 1) met en évidence l'efficacité de la détection de la hauteur de l'herbier de posidonies à l'aide des signaux issus de la haute fréquence.



**Fig. 1 : Fenêtre de visualisation de l'historique du sondeur le long d'une radiale de mesure pour les signaux haute fréquence (100 KHZ) du logiciel SIVA. En haut : Observation de la profondeur (entre 0 et 15 m : profondeur moyenne de 6 m). En bas : Variation autour du fond (+/- 3 m).**

La méthode d'analyse des signaux du sondeur de sédiments sur de la matte morte a été testée dans un premier temps sur une zone pilote située devant le port de Sanary-sur-Mer (Provence, France). Cette zone présentait la particularité d'avoir des tombants de mattes bien marqués et donc mesurables en partie sans avoir recours à des carottages. On observe la corrélation entre les signaux du sondeur de sédiments et la vérité terrain (Fig. 2).



**Fig. 2 : En haut la vérité terrain et en bas les signaux acoustiques. Correspondance des signaux HF et de la cartographie des fonds de la zone située à Sanary-sur-Mer**

La corrélation entre les signaux du sondeur de sédiments et la connaissance du terrain (vérité terrain géo-référencée) met en évidence la capacité de la méthode utilisée pour l'observation :

A haute fréquence (HF), 100 kHz pour ces essais :

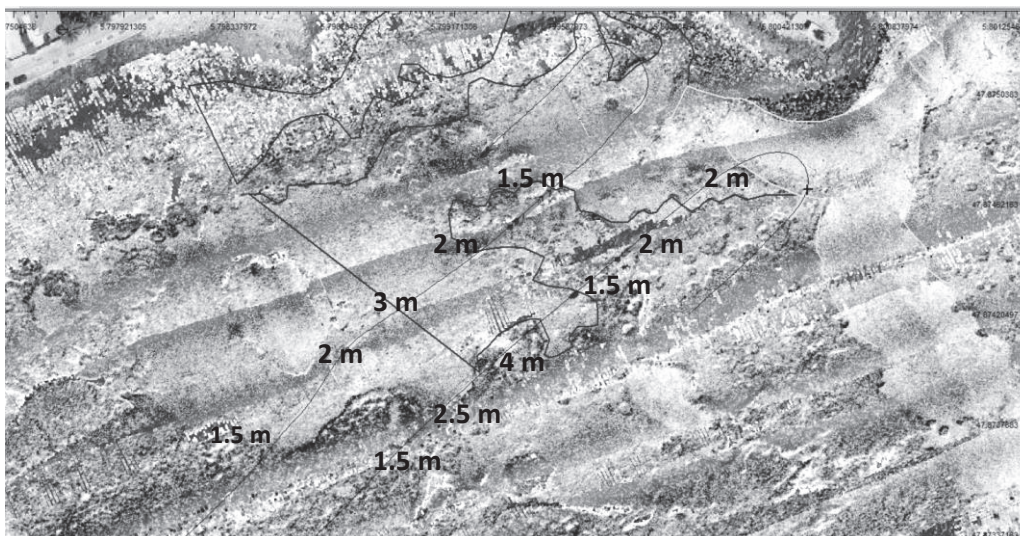
- des posidonies
- de la bathymétrie fine et donc des tombants de matte

A basse fréquence (BF), 10 kHz :

- de l'épaisseur de la couche du fond, plus épaisse dans le cas de la matte morte
- de la présence de réflecteurs dans le substrat correspondant à la base de la matte morte

La détermination de la profondeur de la matte est effectuée en traitant les signaux des sous-couches avec le logiciel SIVA. Lorsque le réflecteur est visible acoustiquement, sa profondeur peut ensuite être déterminée.





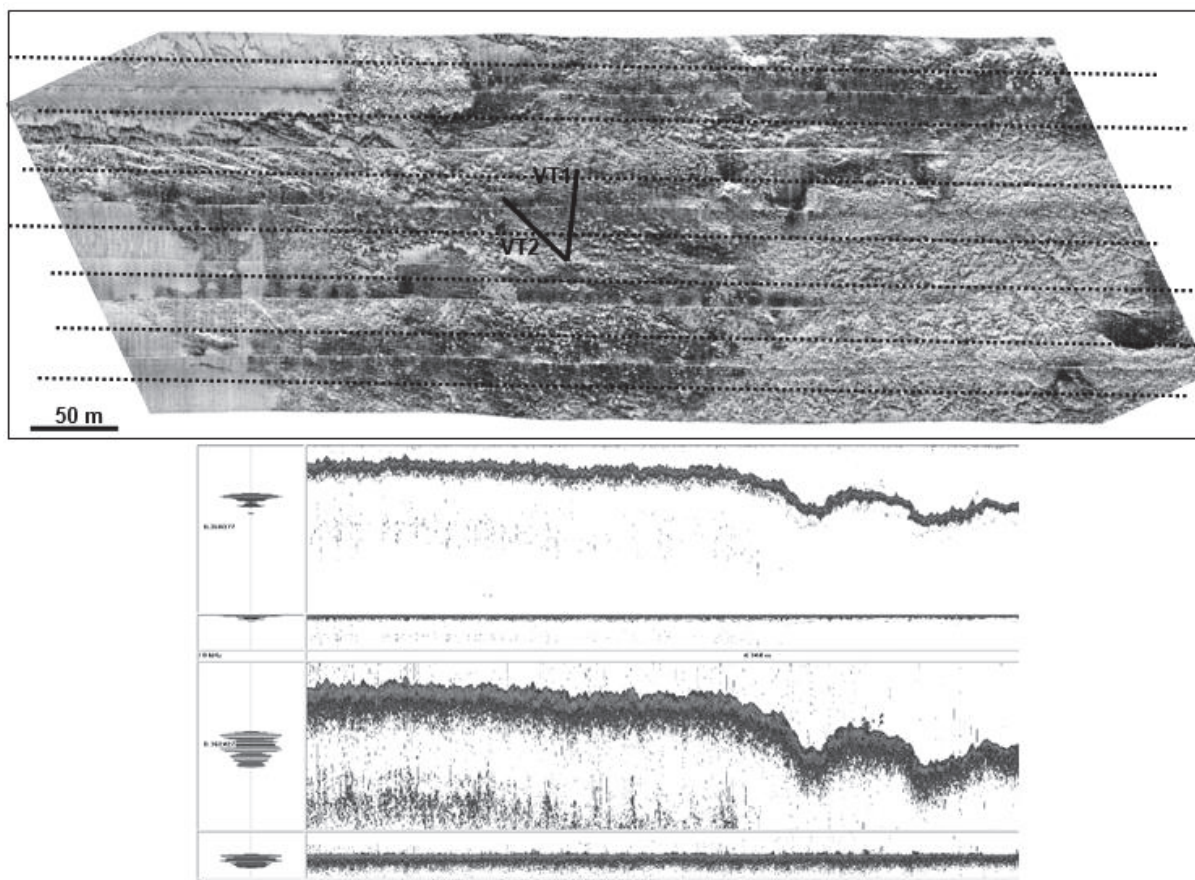
**Fig. 3 : Épaisseur estimée de la matte (en m) dans la zone étudiée à Sanary-sur-Mer**

Au lieu du point « C » (Fig. 2) où le tombant de matte est observable en plongée (hauteur observée 2 m), le système détermine acoustiquement une épaisseur de matte de 3 m sur le plateau de matte, en haut du tombant, et de 1 m au pied du tombant. Les informations du sondeur de sédiment sont cohérentes avec la connaissance du terrain.

Cette méthode a ensuite été employée sur le site de l'aire marine protégée de la côte agathoise (Agde, France) (résultats ci-dessous).

La sismique UHR a permis de visualiser la différence entre un substrat de sable et un substrat de matte, d'observer une épaisseur du substrat plus grande en présence de matte ainsi que la présence de feuilles vivantes de posidonies.

Les épaisseurs mesurées sur le terrain à l'aide d'une tige graduée en centimètres plantée dans la matte ont été comparées en deux lieux pour lesquels le sondeur de sédiments et les vérités terrain étaient proches, (à intersection de la radiale INNOMAR et des radiales de vérités terrain : VT1 et VT2 (Fig. 4)) et sont compilées dans le tableau 1. Les valeurs sont du même ordre de grandeur.



**Fig. 4 :** En haut : Lieux des vérités terrain à Agde (radiale en forme de V). Les lignes en pointillés correspondent aux radiales réalisées avec le sondeur INNOMAR.VT1 et VT2 correspondent à deux portions (2m) de vérité terrain.

En bas : Fenêtre de visualisation de l'historique du sondeur le long d'une radiale pour les signaux haute fréquence (partie haute de la fenêtre) et basse fréquence (partie basse). La profondeur moyenne est de 4 m.

**Tab 1 :** Hauteur de la matre morte estimée via le sondeur INNOMAR et en plongée (VT = vérité terrain) sur le site d'Agde

	INNOMAR	VT plongeur	
		Moyenne (Nbr=3)	Écart type
VT1	52 cm	50,7 cm	1,5 cm
VT2	29 cm	31 cm	0,6 cm

Les résultats obtenus (Tab. 1) montrent que la hauteur de la matte morte est homogène. A l'échelle de l'AMP de la côte agathoise, la couche de matte morte est également homogène et relativement fine, comprise entre 20 et 50 cm. Sa détermination n'est cependant pas possible sur tous les secteurs de matte morte. En effet, compte tenu de la résolution verticale du sondeur de sédiments qui est de 15 cm, il est difficile de mesurer de façon fiable des épaisseurs de couches inférieures à cette résolution, soit une vingtaine de centimètres.

### Discussion

Par rapport aux autres sondeurs conventionnels qui sont souvent couplés à des systèmes de classification des fonds, la sismique réflexion UHR dispose d'un faisceau d'émission très étroit (2°) ce qui lui confère une excellente résolution spatiale jusqu'à plusieurs dizaines de mètres de profondeur. De la même façon, ce type de sismique offre la possibilité de faire varier la fréquence d'émission et donc d'adapter le système en fonction de la densité de l'herbier.

Il existe un inconvénient éventuel lors de l'utilisation de cette méthode : la résolution verticale de l'appareil étant de 15 cm, ne permet pas de mesurer de façon fiable des épaisseurs de couche inférieure à cette résolution. Il est donc important d'effectuer des tests préalables afin d'optimiser la méthode et de choisir la fréquence la mieux adaptée à la problématique du site. A l'inverse, l'avantage est de pouvoir obtenir des renseignements globaux sur ce qui n'est pas accessible par les autres méthodes, hormis par plongées et carottages (hauteur des feuilles et épaisseur de la matte), qui sont, elles, ponctuelles.

En l'état actuel, nous manquons de données réelles, de type carottage, qui permettraient de connaître l'épaisseur de la matte en des lieux où cette dernière se trouve sur un substrat sableux. Ces données sont essentielles pour pouvoir comprendre les signaux acquis, calibrer la méthode et conclure sur sa faisabilité, puisque nous ne l'avons appliquée que dans une gamme d'épaisseurs assez faibles (20 à 50 cm).

Il est probable, suite à l'analyse des données, que dans certains secteurs du site, la matte morte soit située sur un réflecteur assez dur (fond de grès par exemple constitué de sable consolidé), présentant des caractéristiques variant selon son taux de consolidation entre celles du sable et celles de la roche, et empêcherait l'absorption des ondes de l'INNOMAR et donc la visualisation des réflecteurs sous le fond.

La faible épaisseur de matte morte sur le site d'Agde suggère que les processus de développement et de disparition des herbiers à *Posidonia oceanica* du site sont rapides. Ainsi, le rapport matte morte sur herbier vivant bien qu'élevé sur le site ne constituerait pas un phénomène alarmant de disparition de l'herbier mais un développement naturel pour ce type d'herbier confronté à de forts phénomènes d'hydrodynamisme.

### Remerciements

Les auteurs remercient la région Languedoc-Roussillon, l'agence de l'eau Rhône Méditerranée & Corse, le ministère de l'écologie, l'agence des aires marines protégées et la Mission Recherche et Innovation Scientifique de la DGA pour leur soutien dans cette étude, ainsi qu'un référent anonyme.

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## ***CYSTOSEIRA SEDOIDES* (DESFONTAINES) C. AGARDH DES COTES TUNISIENNES : ETAT ACTUEL DES CONNAISSANCES**

### **Résumé**

*Cystoseira sedoides* (Desfontaines) C. Agardh (Ochrophyta, fucales), est connue pour être endémique à la Méditerranée, cantonnée à la partie nord-africaine de la Méditerranée occidentale et à deux localités de Pantelleria (Sicile-Italie). La synthèse des données historiques disponibles sur la biogéographie de *C. sedoides* en Tunisie, confrontée aux résultats d'une prospection de l'infralittoral supérieur (0 - 4 m de profondeur) du littoral tunisien effectuée entre 2012 et 2014, a montré que l'espèce a connu une régression de sa distribution. La présence des peuplements de *C. sedoides* est limitée au littoral Nord de la Tunisie, entre Melloula (frontière Nord Est Tuniso-algérienne) et Raf Raf, limite Ouest du Golfe de Tunis, avec des densités moyennes variables entre 15 et 41 individus/m<sup>2</sup>. La variation des densités des peuplements semblerait être en relation avec l'importance des aménagements côtiers. La disparition de certaines stations anciennes, sur la côte Est de Tunisie, correspond en revanche, probablement ou certainement, à des erreurs de détermination.

**Key-words:** *Cystoseira sedoides*, densité, distribution, régression, Tunisie.

### **Introduction**

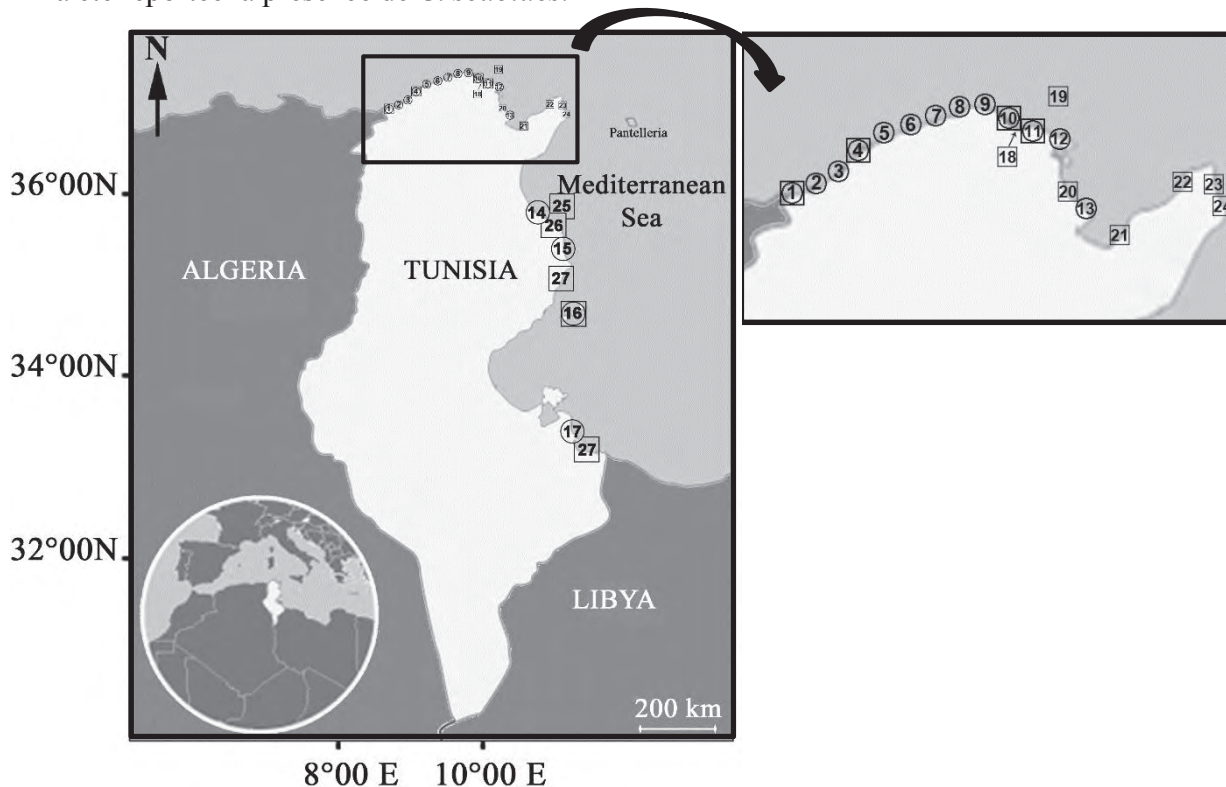
Les écosystèmes côtiers sont profondément affectés par une grande variété d'activités humaines (Arévalo *et al.*, 2007). Les assemblages de *Cystoseira* spp. représentent la communauté macrophytique dominant le paysage marin de la plupart de la Méditerranée (Boudouresque, 1972 ; Ballesteros *et al.*, 1998). La majorité des espèces du genre *Cystoseira* sont très sensibles à la pollution et à toute perturbation anthropique, les habitats qu'elles édifient sont par conséquent aussi fragilisés par l'anthropisation. C'est ainsi que des habitats à *Cystoseira* ont considérablement diminué voire même disparu au cours des dernières décennies dans de nombreuses régions de la Méditerranée (Thibaut *et al.*, 2005 ; Arévalo *et al.*, 2007 ; Sales *et al.*, 2011). *Cystoseira sedoides* (Desfontaines) C. Agardh est une espèce endémique cantonnée à la partie nord-africaine de la Méditerranée occidentale se trouve principalement en Algérie, au large de l'île Pantelleria (Italie) et en Tunisie (Guiry & Guiry, 2014). Le canal de Sicile constitue la limite orientale de son aire de répartition (Colombo *et al.*, 1982 ; Boudouresque *et al.*, 1996) formant ainsi une barrière géographique. Son aire de répartition relativement restreinte, et la rareté de ses stations, font de *C. sedoides* une espèce vulnérable justifiant la prise de mesures pour sa protection (UNEP-PAM-RAC/SPA, 2012). Cette étude vise à améliorer les connaissances sur la biogéographie de *C. sedoides* par une mise à jour de sa distribution et en statuant sur son état de conservation en Tunisie en confrontant les données historiques aux données actuelles.

### **Matériel et Méthodes**

Les données historiques disponibles sur la distribution de *C. sedoides* le long des côtes tunisiennes ont été collectées à partir de toute la documentation disponible (publications scientifiques, ouvrages spécialisés, collections historiques de G. Hamel, de J. Feldmann et de R. Lami dans le Laboratoire de Cryptogamie de Muséum National d'Histoire Naturelle de



Paris (MNHN). Les données actuelles résultent d'une prospection de 16 localités de l'infra littoral supérieur (0-4 m) représentatives de toute la côte tunisienne, réalisée entre Mars 2012 et Février 2013 (Fig. 1) et des observations effectuées, entre Mars et Avril 2014, lors d'un retour sur les localités citées par Ben Maiz *et al.* (1987) (localités 1 à 14, Fig. 1), Mhadhebi *et al.* 2011 (localité 14, Fig. 1) et par Hamza (1987) (localités 15 à 17, Fig. 1) où a été reportée la présence de *C. sedoides*.



**Fig. 1 : Distribution des localités prospectées : Côtes Nord et Péninsule du Cap Bon (1. Baie de Melloula, 2.Tabarka, 3. Barkoukech, 4. Sidi Mechreg, 5. Cap Serrat, 6. Béchateur, 7. Ras Angela, 8. Cap Blanc, 9. Bizerte et Cap Bizerte, 10. Rimel, 18. Ras Jebel, 11. Cap Zebib, 12. Raf Raf, 19. Iles Cani); Golfe de Tunis (20. La Marsa, 13. Sidi Bousaid, 21. Sidi Rais; 22. Sidi Daoud, 23. Dar Allouche, 24. Kelibia), Golfe de Hammamet (14. Monastir, 25. Iles Kuriat); Golfe de Gabès (26. Salakta, 15. Chebba, 27. Melloulèche, 16. Kerkennah, 17. Zarzis, 28. El Ketef)**  
**□: Localités prospectées entre Mars 2012 et Février 2013 ; ○ : localités prospectées entre Mars et Avril 2014**

Dans chaque localité où l'espèce est trouvée, les différentes sources de nuisance d'origine anthropiques ou naturelles pouvant perturber les écosystèmes à *Cystoseira* ont été identifiées, et l'estimation de la densité de *C. sedoides* a été faite. La densité a été déterminée par dénombrement des individus de *C. sedoides* présents à l'intérieur d'un quadrat (40 cm x 40 cm) sur 10 répétitions dans chaque station et exprimée en nombre moyen d'individus par m<sup>2</sup>. Les densités moyennes dans les différentes stations ont été comparées entre elles par une analyse de variance à une voie. Le classement des densités moyennes a été réalisé au moyen du test de Newman-Keuls (Sokal & Rohlf, 1981) au seuil de confiance p égal à 5 %.

## Résultats et discussion

### Données historiques

*Cystoseira sedoides* fût signalée pour la première fois en Tunisie sur les côtes Nord à Tabarka par Hamel en 1925 (Herbier R. Lami, exsiccata ref. PC0535386- MNHN). Elle fût

reportée entre autre à Tabarka par Pottier (1929); Feldmann (1931: Herbar J. Feldmann - Algues de la Méditerranée - exsiccata ref. PC0478032-Hamel *legit*); Hamel (1939) ; Ben Maiz *et al.* (1987). Sur les côtes Nord, elle a été signalée de même à Cap Serrat (Herbar J. Feldmann, 1931-Algues de la Méditerranée - exsiccata ref. PC0478033-MNHN); Hamel, 1931-1939 ; Meñ z & Mathieson, 1981 ; Ben Maiz *et al.*, 1987), Béchateur (Herbar cryptogamique de MNHN, Manning & family Grimm, 1973 (SOSC EGM3 - exsiccata ref. PC0535382); Ouahchi, 1977 ; Meñ z & Mathieson, 1981 ; Ben Maiz *et al.*, 1987), Cap Blanc (Zaouali, 1973), Cap Bizerte (Pottier, 1929), Bizerte (Zaouali, 1973 ; Meñ z & Mathieson, 1981 ; Ben Maiz *et al.*, 1987) et à Cap Zebib (Meñ z & Mathieson, 1981; Ben Maiz *et al.*, 1987). Dans le golfe de Tunis, elle a été reportée à Sidi Bousaid (Ouahchi, 1977 ; Ben Maiz *et al.*, 1987). Dans le golfe de Hammamet, elle fût signalée à Monastir en 1981 (Meñ z & Mathieson, 1981) et a été reportée plus tard dans Ben Maiz *et al.* (1987) et récemment par Mhadhebi *et al.* (2011). Dans le golfe de Gabès *C. sedoides* a été citée à Chebba, autour de l'archipel des Kerkennah et à Zarzis par Hamza (1987).

#### Données Actuelles

Nos prospections ont permis d'identifier *C. sedoides* uniquement sur le littoral Nord tunisien. D'ouest en est, elle est trouvée dans la Baie de Melloula à l'extrême ouest de la Tunisie, à la frontière Tuniso-algérienne, à Tabarka, Barkoukech, Sidi Mechreg, Cap Serrat, Béchateur, Ras Angela, Cap Blanc, Plage de Rimel, Cap Zebib et à Raf Raf (Fig. 1, stations 1, 2, 3, 4, 5, 6, 7, 8, 10, 11 et 12 respectivement). *C. sedoides* occupe les substrats rocheux photophiles de l'étage infralittoral supérieur. Elle forme des tâches isolées ou des ceintures discontinues caractéristiques entre 0,5 et 4 m de profondeur dans des stations exposées à un hydrodynamisme modéré à moyennement fort.

#### Densité des peuplements

La densité de *C. sedoides* est variable d'une station à l'autre mais semble être fortement influencée par l'hydrodynamisme d'une part, et par l'extension urbaine d'autre part. En effet, les stations moyennement à fortement exposées aux vagues et loin de toute influence urbaine semblent être celles où les peuplements de *C. sedoides* sont les plus denses (Tab.1) (Cap Zebib, Béchateur, Ras Angela, Raf Raf et Sidi Mechreg avec respectivement 41, 39, 38, 37 et 34 individus/m<sup>2</sup>). C'est aussi le cas de la station de Barkoukech avec une densité de 29 individus au m<sup>2</sup>, non significativement différente ( $p \geq 0,05$ ) de celles des stations suscitées. A Cap Blanc et à Cap Serrat, bien que en mode moyennement agité à agité, favorable au développement de *C. sedoides* (Hamel, 1931), les densités sont relativement faibles (28 individus/m<sup>2</sup>). Ceci ne pourrait être expliqué que par la Pression urbaine exercée sur le littoral de ces stations.

Dans la baie de Melloula, bien que la station soit loin de toute urbanisation, la densité de *C. sedoides* est parmi les plus faibles notées au Nord de la Tunisie (22 individu/m<sup>2</sup>). Cette densité serait liée au mode relativement calme de la station, peu favorable au développement de *C. sedoides*.

Les densités de *C. sedoides* les plus faibles ont été enregistrées à Tabarka et Plage de Rimel, (21 et 15 individus/m<sup>2</sup> respectivement).

A Tabarka, bien que le substrat rocheux et l'hydrodynamisme soient favorables à l'installation de *C. sedoides*, la densité de celle-ci tend à être parmi les plus faibles enregistrées sur son aire de répartition en Tunisie. L'aménagement et l'activité portuaire de la ville de Tabarka semblerait être le facteur responsable de la dégradation des peuplements de *C. sedoides* dans la zone. L'effet de l'aménagement et de l'activité portuaire est plus prononcé dans la zone de la plage de Rimel, où la densité de *C. sedoides*

est la plus faible le long des côtes Nord tunisiennes (15 individus au m<sup>2</sup> ; p < 0,05), avec la proximité des ports de pêche, de commerce et militaire de la ville de Bizerte. L'influence portuaire semblerait donc avoir le plus d'effet négatif sur les peuplements de *C. sedoides*. Le piétinement par les estivants ne constitue qu'un facteur aggravant la dégradation des peuplements, mais pas le plus déterminant.

**Tab. 1 : Densité de *C. sedoides*, description et sources de nuisances dans les stations à *C. sedoides*.**

Station	Densité*	Description de la station	Sources de nuisances
Baie de Melloula	21,87 <sup>de</sup> (6,07)	Baie peu profonde et abritée. <i>C. sedoides</i> sur roches affleurantes à complètement émergées ; Hydrodynamisme faible.	Piétinement par les estivants Surpâturage (oursins et saupes)
Tabarka	21,25 <sup>de</sup> (7,90)	Plage rocheuse. <i>C. sedoides</i> sur roches affleurantes ; Hydrodynamisme modéré	Pression urbaine Influence portuaire Piétinement par les estivants
Barkoukech	28,75 <sup>bcd</sup> (10,70)	Plage sableuse. <i>C. sedoides</i> sur roches et blocs rocheux épars ; Hydrodynamisme modéré.	Piétinement par les estivants
Sidi Mechreg	34,37 <sup>abc</sup> (7,93)	Plages situées de part et d'autre d'une petite falaise et blocs rocheux ; <i>C. sedoides</i> sur blocs rocheux ; Hydrodynamisme modéré à moyennement fort.	Surpâturage (oursins et saupes)
Cap Serrat	27,50 <sup>ed</sup> (7,34)	Bancs rocheux accidentés avec <i>C. sedoides</i> ; estran encombré d'éboulis grossiers nus ; Hydrodynamisme modéré à moyennement fort.	Surpâturage (oursins et saupes) Piétinement par les estivants Pression urbaine
Béchateur	39,37 <sup>a</sup> (11,04)	Côtes rocheuses ; <i>C. sedoides</i> en tâches sur roches éparses ; Hydrodynamisme modéré à moyennement fort.	Surpâturage (oursins et saupes) Piétinement par les estivants
Ras Angela	38,12 <sup>a</sup> (12,65)	Plage sableuse ; <i>C. sedoides</i> en tâches sur roches éparses ; Hydrodynamisme modéré à moyennement fort.	Surpâturage (oursins et saupes) Piétinement par les estivants
Cap Blanc	28,12 <sup>ed</sup> (8,96)	Eperon rocheux en continuité avec des falaises surplombant une plage ; <i>C. sedoides</i> sur blocs rocheux dispersés ; Hydrodynamisme modéré à moyennement fort.	Surpâturage (oursins et saupes) Piétinement par les estivants Pression urbaine
Plage de Rimel	15,62 <sup>e</sup> (5,31)	Plage sableuse longée par un platier rocheux large de 3m maximum, discontinu ; <i>C. sedoides</i> sur platier rocheux ; Hydrodynamisme modéré.	Influence portuaire et industrielle
Cap Zebib	41,25 <sup>a</sup> (9,86)	Côtes rocheuses ; Ceinture discontinue de <i>C. sedoides</i> sur de larges plateformes rocheuses ; Hydrodynamisme modéré à fort.	Piétinement par les estivants Surpâturage (oursins et saupes)
Raf Raf	36,87 <sup>ab</sup> (11,20)	Plage sableuse ; alternance de fonds rocheux et de fonds meubles ; <i>C. sedoides</i> sur affleurements rocheux épars ; Hydrodynamisme modéré à fort.	Piétinement par les estivants Usine textile

\* Moyennes sur 10 répétitions exprimées en individus/m<sup>2</sup> ; les valeurs entre parenthèses représentent les écart-type

Les valeurs suivies par une même lettre en exposant ne sont pas significativement différentes (P < 0,05, test de Newman-Keuls).

### Extinction de *Cystoseira sedoides* des côtes tunisiennes

Sur les côtes de Bizerte, Nord de la Tunisie (Fig.1 ; Station 9), la biomasse de *Cystoseira sedoides* était estimée à 5 360 kg/m<sup>2</sup> (Zaouali, 1973). De nos jours, *C. sedoides* a totalement disparu de Bizerte et de ses alentours (Cap Bizerte). La zone de Bizerte et une zone urbaine, portuaire (de pêche, commerciale et militaire) et un grand pôle industriel abritant entre autres une usine de sidérurgie, une raffinerie de pétrole et une unité de raffinage des huiles usagées (UNEP-MAP, 2002). Les effluents urbains et industriels excessifs de ces aménagements seraient à l'origine de la régression des peuplements de *C. sedoides* au cours de ces dernières années.

A Sidi Bousaid, les pressions urbaines, l'intense développement portuaire et les travaux d'extension du port de plaisance et l'aménagement côtier ayant pour conséquence un ensablement du littoral proche seraient les causes principales de déclin de *C. sedoides* de cette zone.

A Monastir, nos prospections n'ont révélé aucune trace de *C. sedoides*. L'absence d'échantillons de références, aussi bien relatif à la signalisation de Meñez & Mathieson (1981) qu'à celle de Mhadhebi *et al.* (2011) nous laisse suggérer deux hypothèses : soit une erreur sur l'identification de *C. sedoides*, soit une extinction des peuplements de la région. Si extinction il y a eu, elle serait due soit à une érosion côtière soit à l'expansion des villes qui constituent les principales pressions sur le golfe de Hammamet en général (UNEP-MAP, 2002).

Malgré le grand nombre d'études portant sur le genre *Cystoseira*, l'identification des espèces reste difficile en raison de la grande variabilité de la morphologie, dépendant principalement de la saison et des conditions environnementales (Roberts, 1967). *C. sedoides*, bien que facilement reconnaissable, l'erreur sur son identification par un non spécialiste demeure probable. Mhadhebi *et al.* (2011) à Monastir sont peut-être dans ce cas, mais, le cas de Hamza (1987) avec sa signalisation de *C. sedoides* dans le golfe de Gabès constitue un cas typiquement avéré, conforté par la publication d'une photo de l'échantillon attribué à *C. sedoides* ne ressemblant en aucun cas à l'espèce en question mais pouvant être *C. spinosa* déjà signalée sur les côtes sud Tunisiennes. Il ne s'agit donc pas dans ce cas, comme on le croirait, d'une extinction des peuplements de *C. sedoides* dans le golfe de Gabès, mais d'une erreur de signalisation de l'espèce en dehors de son aire de distribution géographique connue à ce jour.

### **Conclusion**

Les peuplements de *Cystoseira sedoides* présent sur la côte Nord de la Tunisie présentent un état de conservation variable. Ils semblent être bien conservés dans les zones peu anthropisées et affectés, le plus, par les aménagements portuaires et industriels, causes probables de leur extinction de la région de Bizerte (Côte Nord de la Tunisie) et de Sidi Bousaid (Golfe de Tunis). Le suivi des peuplements mérite d'être plus approfondi afin de proposer des mesures nationales de conservation respectant le développement économique et social du pays.

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## **INSIGHT INTO THE TYPOLOGY OF REEF FORMATIONS OF THE MEDITERRANEAN SEAGRASS *POSIDONIA OCEANICA***

### **Abstract**

*Reef formations of the Mediterranean seagrass *Posidonia oceanica* result from the rise of the matte towards the sea surface, when the meadow reaches the sea surface, and the tips of the leaves emerge. The process of reef formation was first described in the Bay of Port-Cros (Provence, France). Since then, the Port-Cros barrier reef has been considered as the model of *P. oceanica* reefs. In fact, there is a wide variety of *P. oceanica* reefs. Here, we outline a typology of these reef formations: fringing reefs (FR), barrier reefs with matte lagoon (BRML), barrier reefs with eroded matte lagoon (BREML), barrier reefs with a geomorphological lagoon (BRGL), fossil barrier reefs (FOBR), false barrier reefs (FABR), plateau reefs (PR), perpendicular reefs (PER) and atolls (micro- and macro-atoll, MA). It is of paramount importance to take into account this typology, which encompasses reefs that differ from the paradigmatic Port-Cros barrier reef, for the management of the coastal zone, as these reef formations have a high heritage value. In addition, these reef formations contribute greatly to ecosystem services, such as beach protection from erosion and fish nursery. The reef building process lasts centuries, or even millennia, so that their destruction is irreversible at human scale.*

**Key-words:** *Posidonia oceanica*, reef formations, natural heritage, ecosystem services.

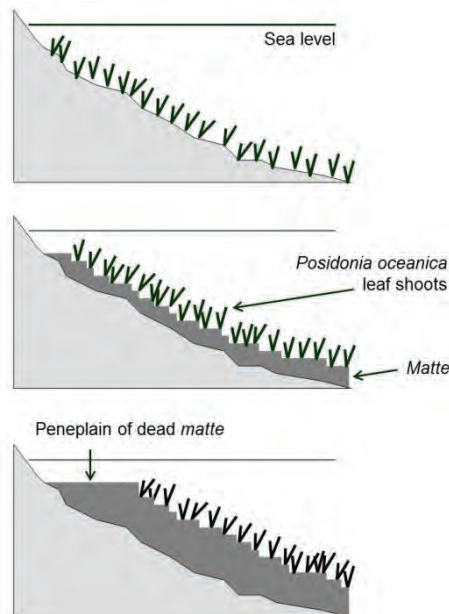
### **Introduction**

*Posidonia oceanica* (Linnaeus) Delile is a seagrass (Magnoliophyta) endemic to the Mediterranean Sea (Boudouresque *et al.*, 2012). It thrives between the sea level and 20-45 m depth, depending upon the water transparency and constitutes extensive meadows which play a paramount role in the functioning of the Mediterranean coastal ecosystems (Pergent *et al.*, 2012). Rhizomes and leaf shoots can grow horizontally, to colonize the substrate (plagiotropic rhizomes). The leaf canopy traps sediment. To resist being buried, rhizomes can also grow vertically (orthotropic rhizomes) (Molinier & Picard, 1952). The structure constituted by live and dead parts of rhizomes and roots, together with the sediment that fills the interstices, is called ‘matte’. Sediment trapping and orthotropic rhizome growth result in the rising of the sea bottom over time; the average rise ranges from 10 cm to 100 cm per century (Molinier & Picard, 1952; Mateo *et al.*, 1997; Boudouresque *et al.*, 2012). This rising can bring the meadow close to the sea surface. Under exposed conditions, the rising stops ~1 to 2 m below the sea surface. Hydrodynamism prevents the rise continuing by breaking the rhizomes and their leaf bundles (shoots). This results in the formation of a peneplain of dead matte (PDM) (Molinier & Picard, 1952) (Fig. 1). In contrast, under sheltered conditions, the rise of the *matte* can continue right up to the sea surface.

We define a *P. oceanica* reef as a formation built by the seagrass with leaf tips emerging from the sea surface and/or spreading at the sea surface, at least at low tide and in spring and early summer, when leaf length is at its maximum. Here, we propose a typology of



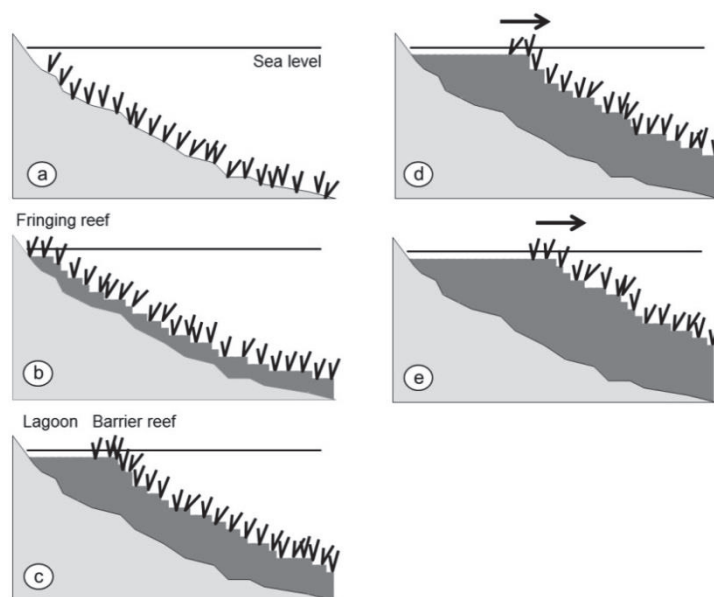
*P. oceanica* reef formations, which accounts for their real diversity, and we emphasize the role of these reefs in ecosystem services and their importance in the management of the coastal zone.



**Fig. 1: Dynamics of the *Posidonia oceanica* meadow under exposed conditions and the formation of a peneplain of dead matte (PDM). Light grey: initial substrate. Dark grey: the matte (dead or alive).**

### Typology of reef formations

In the first stage, the emersion of leaf tips happens close to the coast. This formation is known as a **fringing reef (FR)**. Then, the continuing rise of the *matte* widens the fringing reef. Within the fringing reef, the leaf canopy and the shallowness hamper water circulation. The water temperature may go below (in winter) and above (in summer) the limits of *P. oceanica* tolerance. The case is similar with salinity. As a result, between the coast and the *P. oceanica* emersion front, leaf shoots die, and a **lagoon** is formed (Fig. 2).

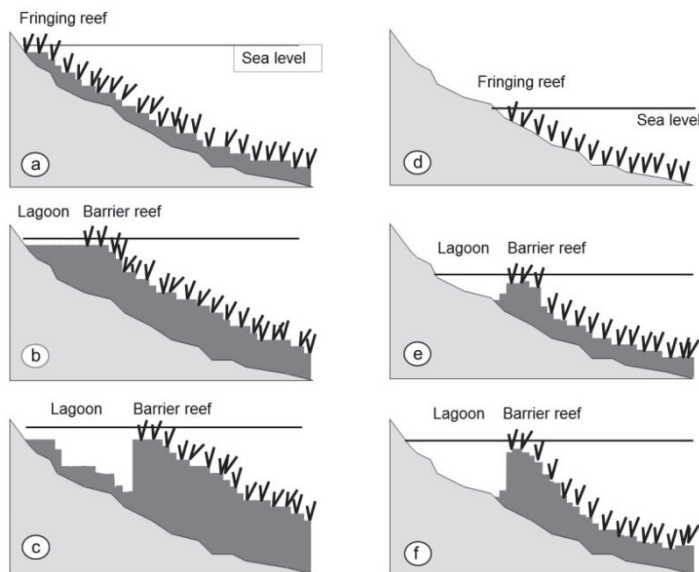


**Fig. 2: Dynamics of the *P. oceanica* meadow under sheltered conditions: process of building and evolution of a fringing reef and a barrier reef (a through e). The matte rises and reaches the sea surface. It first forms a fringing reef (b) then a barrier reef separated from the coast by a lagoon (c). Over time, the barrier reef expands seaward (arrows). Light grey: initial substrate. Dark grey: the matte (dead or alive). From Boudouresque *et al.* (2012), modified.**

The *P. oceanica* emersion front thus constitutes a **barrier reef (BR)**. With time, the barrier reef moves seawards and the lagoon grows (Fig. 2d-e). In the lagoon, two other species of seagrasses can establish, *Zostera noltei* Hornemann and *Cymodocea nodosa* (Ucria) Ascherson. The process of building of *P. oceanica* fringing and barrier reefs was initially described by Molinier & Picard (1952), from the Bay of Port-Cros (Port-Cros Island, Provence). For this reason, the Port-Cros reef was considered as the type, a sort of paradigm, of this kind of formation, despite its poor state of conservation (Goujard *et al.*, 2010), and little attention was paid to the diversity of the *P. oceanica* reef formations. The barrier reef described in the Bay of Port-Cros is referred to here as a **Barrier Reef**

with **Matte Lagoon (BRML)**. The lagoon is shallow (rarely more than 1 m depth). Beneath a thin layer of sediment, the dead *matte* is present. The depth of the lagoon, slightly greater than that of the emerging reef, can be due to the partial decomposition of the *P. oceanica* remains and the resulting packing down of the dead *matte*.

A **barrier reef with eroded matte lagoon (BREML)** is similar to a BRML. The difference is that the dead *matte* of the lagoon has been eroded, so that the depth of the lagoon can be greater (several metres) (Fig. 3c). This erosion can be natural or due to human dredging, in order to allow the mooring of small boats. The BR of La Madrague-de-Giens (Gulf of Giens, Provence) belongs to this type of *P. oceanica* reef formation.



**Fig. 3: a-c. Building of a BREML (barrier reef with eroded matte lagoon). d-f. Building of a BRGL (barrier reef with geomorphological lagoon). Note the relative sea level (RSL) rise over time. Light grey: initial substrate. Dark grey: the *matte* (dead or alive).**

A **barrier reef with a geomorphological lagoon (BRGL)** is a BR whose lagoon is not only relatively deep but is also devoid of dead *matte*. It results from the rise of the relative sea level, either due the actual rise of the sea level, or to the subsidence of the shelf. An initial fringing reef (Fig. 3d) rose on the spot and turned into a barrier reef (Fig. 3e-f). Within the lagoon, *P. oceanica* was never present. If erosion of the dead *matte* within the lagoon of a BREML is total, it can be misinterpreted as a BRGL. The presence of BRGLs is unlikely in areas where recent subsidence of the shelf is absent.

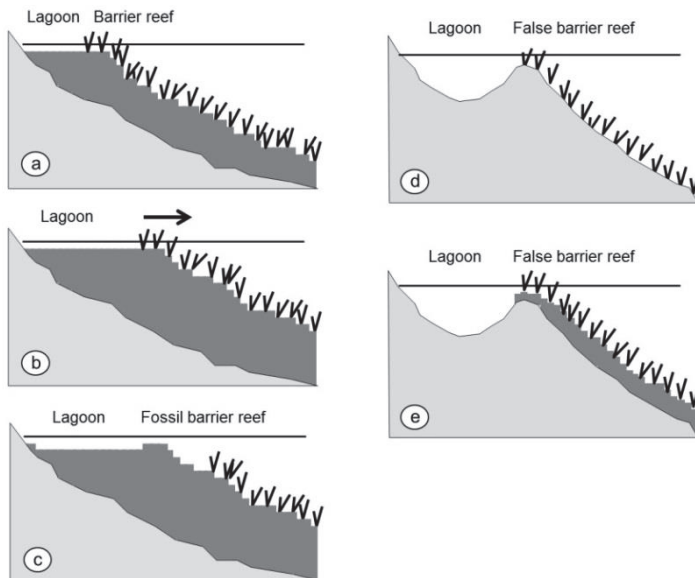
A **fossil barrier reef (FOBR)** is a former barrier reef (BRML) whose leaf bundles at the emersion front subsequently withered and died. However, the profile, with a shallow emersion front and a slightly deeper lagoon, has been preserved (Fig. 4c). Of course, the seaward progression of the emersion is stopped. Over time, the emersion front, which is no longer protected by leaves and no longer traps particles, will be eroded by hydrodynamism, so that the formation will resemble a peneplain of dead *matte* (PDM).

A **false barrier reef (FABR)** corresponds to a lagoon of geomorphological origin, separated from the open sea by a shallow rocky ridge. *P. oceanica* established on the seaward side of the rocky ridge, mimicking a *P. oceanica* barrier reef (BR) (Fig. 4d-e).

In the San-Fiorenzu Gulf (Saint-Florent, northern Corsica), a unique *P. oceanica* formation has been described, the **plateau reef (PR)** (Boudouresque *et al.*, 1985; Pasqualini *et al.*, 1997). It consists in a triangle of shallow dead *matte*, fringed with living *P. oceanica* (Fig. 5). Its genesis probably resulted from alternate and/or opposite currents running

to the two external sides of the triangle; two barrier reefs, back to back, progressed seaward and generated a kind of lagoon in the form of a triangle.

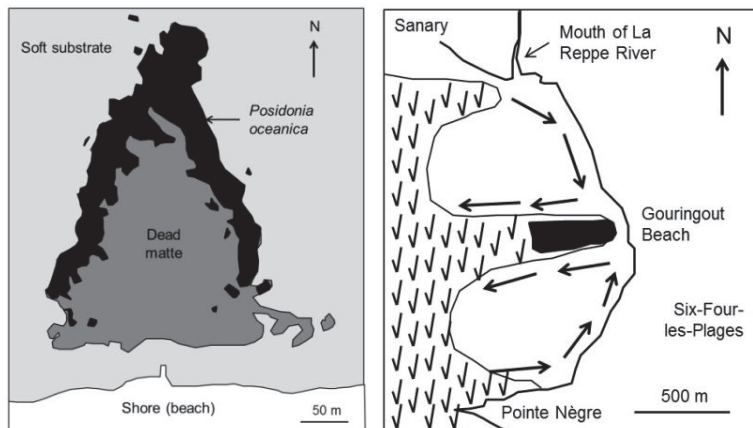
At Gouiringout beach (Sanary, eastern Provence, France), a *P. oceanica* reef perpendicular to the coast (**perpendicular reef**, PER) is probably due to a couple of opposite current cells (Fig. 5). PERs possibly correspond to a reduced, or an initial, form of PR.



**Fig. 4:** a-c. Origin of a fossil barrier reef (FOBR). d-e: False barrier reef (FABR). Light grey: initial substrate. Dark grey: the mat (dead or alive).

Finally, **atolls** of *P. oceanica* (micro- and macro-atolls, MA), where leaves are emergent at low tide, could be considered as belonging to the family of the *P. oceanica* reef formations (Fig. 6). The San Fiorenzu MA is a circle of living *P. oceanica*, 10-12 m in diameter (micro-atoll), the centre of which is occupied by

dead *matte*; plagiotropic (i.e. horizontal) rhizomes develop at the edge of the MA, in a centrifugal manner, whereas the shoots die in the central part of the MA. Near the mouth of the Ain Al-Ghazala Lagoon (Libya), large atolls of *P. oceanica*, 20-70 m in diameter, called ‘macro-atolls’, have been described by Pergent *et al.* (2007).



**Fig. 5:** Left. A map of the plateau reef (PR) of San-Fiorenzu, northern Corsica. Redrawn from Pasqualini *et al.* (1997). Right. The perpendicular reef (PER) of Gouiringout beach (eastern Provence). v: non-emergent *P. oceanica* meadow. Black: PER. Arrows: dominant currents.

### Ecosystem services and natural heritage value of *Posidonia oceanica* reefs

The *P. oceanica* meadows are at the origin of a number of ecosystem services of major importance (Fig. 7). In addition to the overall services provided by *P. oceanica* meadow, reef formations enhance the protection of beaches against erosion (service 7) and harbour nurseries of coastal ‘fish’ (teleosts) of economic interest (service 2).

*P. oceanica* reef formations are highly vulnerable to human activities, e.g. coastal development, establishment of port facilities, small pleasure boats which unwittingly plough furrows, at low tide, to cross the reef, deliberate dredging of channels to allow boats to moor within the lagoon and artificial beaches (Boudouresque *et al.*, 2012). Reefs have

probably been destroyed, millennia or centuries ago, because they were located in bays that have been made into ports, for example the Marseille' Lacydon (Greek name of the modern 'Vieux Port'), the bay of Toulon (eastern Provence), etc. (Boudouresque *et al.*, 1985). More recently, the Bandol BRML was buried beneath land reclamation. Nowadays, many *P. oceanica* reef formations are in a poor state of conservation, such as the BRMLs and BREMLs of Port-Cros Bay, Le Bruscat and La Madrague de Giens (eastern Provence). Some of them are used as harbours for small leisure boats, the barrier reef acting as a natural jetty, such as La Madrague de Giens and Le Pilon reefs (eastern Provence); passes were opened through the reef and the lagoon was deepened. Once the regression has started, stopping it proves to be very difficult, if not impossible, as illustrated by the barrier reef (BRML) of Port-Cros Bay (Goujard *et al.*, 2010). Management by a 50 year-old National Park, steady monitoring by scientific teams and the Scientific Council of the National Park, the prohibition of boating and mooring in the vicinity of the reef and within the lagoon, and well implemented regulations have not succeeded in stopping the decline. At the scale of the Mediterranean Sea, *P. oceanica* meadows may be more resilient to human disturbance than previously thought (Leriche *et al.*, 2006). However, as regards *P. oceanica* reef formations, the picture is probably worse than we usually think. All *P. oceanica* reefs, whatever their type, therefore deserve particular attention and protection.

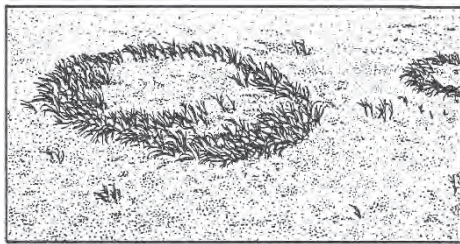


Fig. 6: A micro-atoll of *P. oceanica* (MA) in Turkey. From S. Cirik in Boudouresque *et al.* (1990).

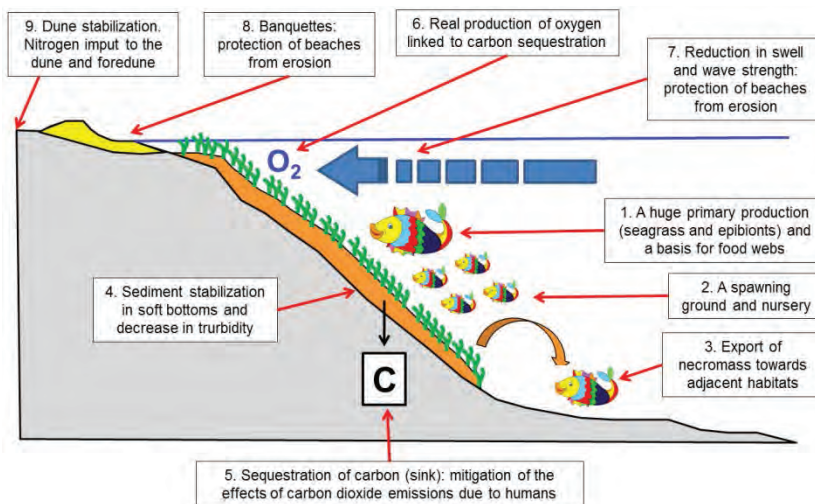


Fig. 7: Ecosystem services (1 through 9) generated by the *P. oceanica* ecosystem. Reef formations, where present, enhance the protection of beaches from erosion (service 7), while lagoons play an important role as 'fish' (teleosts) nurseries (service 2).

## Conclusion

More than sixty years ago, the first *P. oceanica* barrier reefs were described. Subsequently, a few other reef formations were described, namely fringing reefs, plateau reefs and atolls. Here, we have proposed a comprehensive typology of these formations, with their formal description, together with a number of new previously unrecorded reef types. Taking into account this typology, it may be expected that a number of *P. oceanica* reefs,



that did not match the paradigmatic structure of the Port-Cros barrier reef (now: barrier reef with matte lagoon; BRML), will attract researchers' attention. In addition, reef formations from southern and eastern Mediterranean (e.g. Greece, Turkey, Tunisia and Algeria) were poorly documented here, in the framework of a 6-page article. They obviously should require further investigations.

At the scale of the Mediterranean Sea, *P. oceanica* reef formations may be less uncommon than previously thought. However, their state of preservation seems to be generally poor. Reef building requires centuries or millennia and their regression or loss is irreversible at human scale. They therefore merit a high level of protection.

**Acknowledgements.** The authors acknowledge with Michael Paul for improvement of the English text and valuable comments from an anonymous reviewer. This work was funded by the *Parc national de Port-Cros* (PNPC) (partnership between PNPC and GIS Posidonie).

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## **ANALYSE CRITIQUE DE L'ÉVALUATION DE L'ÉTAT DE CONSERVATION DE L'HERBIER DE POSIDONIE DANS LE CADRE DU PROGRAMME DE CARTOGRAPHIE DES HABITATS MARINS – CARTHAM**

### **Résumé**

*Le programme de cartographie des habitats marins (CARTHAM) mis en place par l'Agence des aires marines protégées, a permis d'évaluer l'état de conservation des herbiers de Posidonie et sa surface au sein de plusieurs aires marines protégées (AMP) dont les sites Natura 2000. CARTHAM a permis une cartographie quasi continue des biocénoses marines côtières.*

*L'analyse critique porte sur la méthode d'évaluation de l'Etat de Conservation (EC) de l'herbier de Posidonie proposée dans le cadre du programme CARTHAM.*

*La méthode d'évaluation s'appuie sur plusieurs paramètres comme la surface de l'habitat, « structure et fonctionnalité » ou encore « menace et pression ». Pour encadrer ces travaux, conduits sur 26 AMP et par 8 opérateurs, le Muséum National d'Histoire Naturelle (MNHN) a identifié un ensemble de descripteurs servant de base commune pour apprécier l'EC.*

*L'évaluation de l'EC de l'herbier de Posidonie, malgré cette recherche d'harmonisation reste délicate. L'avis d'expert est renforcé par les données CARTHAM, mais est indispensable à l'analyse. Le niveau d'investissement et l'effort d'échantillonnage ont été proposés par les opérateurs et n'ont pas été être encadré par des documents techniques. L'analyse de l'EC n'est pas aujourd'hui structurée pour répondre à une évaluation multi-échelle.*

**Key-words:** cartographie, état de conservation, Posidonie

### **Introduction**

Le programme de cartographie des habitats marins (CARTHAM) a été mis en œuvre par l'Agence des aires marines protégées (AAMP) sur demande du Ministère chargé de l'environnement. Ce programme vise, notamment à répondre aux engagements communautaires et à la stratégie nationale création et de gestion d'aires marines protégées (AMP). Il a été lancé en 2010 sur les trois façades maritimes françaises.

CARTHAM concerne, pour la façade Méditerranée, 23 sites Natura 2000, un parc naturel marin et deux parcs nationaux, soit 12 lots suite au regroupement de plusieurs AMP (Fig. 1). Il répond à deux objectifs principaux : (i) établir un état initial biologique de l'ensemble des AMP ; (ii) évaluer l'Etat de Conservation (EC) des habitats d'intérêt communautaire. Il a été encadré scientifiquement par le Muséum National d'Histoire Naturelle (MNHN) et un collègue d'experts issus des conseils scientifiques régionaux du patrimoine naturel (CSRPN).

L'étude des lots CARTHAM de Méditerranée a été confiée à 8 opérateurs. Les travaux ont été relus par les CSRPN et les données brutes ont été contrôlées par les géomaticiens de l'AAMP. L'EC, au sens de la Directive Habitats, Faune et Flore (DHFF), est évalué comme étant « favorable », « défavorable inadéquat » ou « défavorable mauvais ». Les 4 paramètres retenus pour cette évaluation sont l'aire de répartition, la surface, les « structures et fonctions » et les perspectives futures de conservation. Les avis d'expert

sont repris quand les données factuelles sont insuffisantes pour statuer. Lorsque l'information disponible est insuffisante pour statuer, il est possible de rapporter l'état de conservation comme "inconnu".

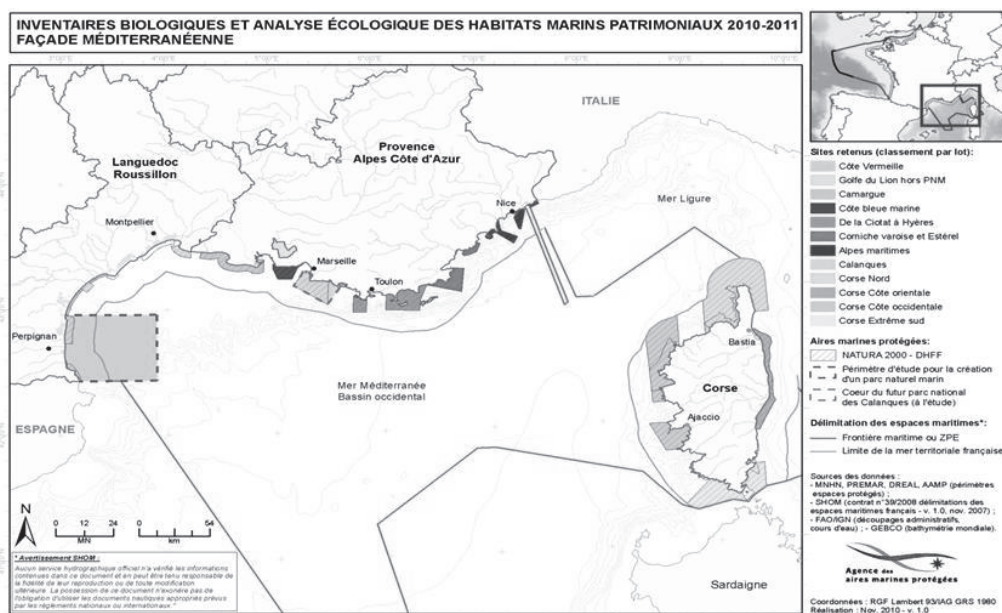


Fig. 1 : Carte des 12 lots CARTHAM de Méditerranée (source: AAMP)

L'objectif est de traiter les éléments liés aux descripteurs de l'EC de l'herbier de Posidonie et à l'estimation de sa surface utilisés pour CARTHAM sur les lots méditerranéens, d'en faire une analyse critique et de rapporter les interrogations soulevés notamment sur l'évaluation de l'EC d'un habitat à différentes échelles.

### Matériels and méthodes

Sur la base d'un état des connaissances des données existantes les opérateurs CARTHAM ont proposé des investigations complémentaires faisant appelés à différentes méthodes d'acquisition connues et complémentaires (Fig. 2). La mise en œuvre de ces méthodes a été hétérogène d'une AMP à une autre, dépendant des offres retenues suite à une mise en concurrence des opérateurs possibles. Toutes les offres retenues ont proposées de renseigner les données liées aux herbiers de Posidonie.

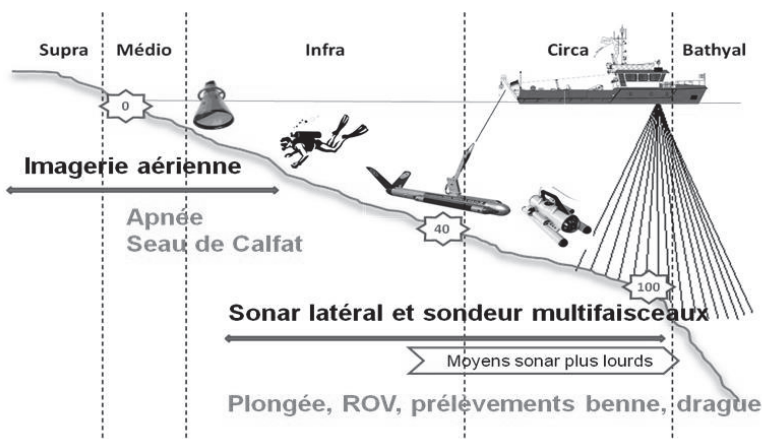
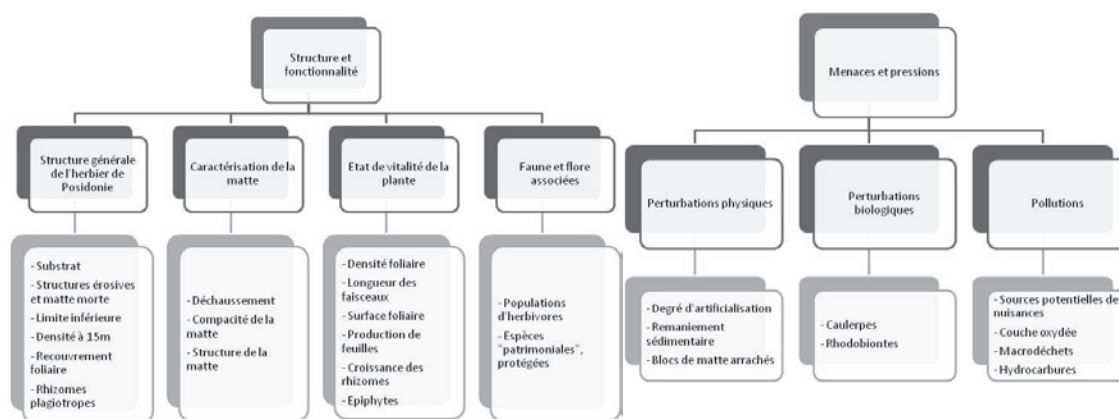


Fig. 2 : Méthodes d'acquisition de terrain des données utilisées dans pour CARTHAM (d'après Clabaut & Corman, non publié)

Pour encadrer et harmoniser au mieux le travail des opérateurs le MNHN a proposé une grille de descripteurs d'état, une métrique (unité de collecte du descripteur), ainsi qu'une méthode et une échelle de collecte. Pour les informations biologiques, une stratégie de non prélèvement sur l'herbier a été adoptée afin de ne pas dégrader l'habitat, stratégie compatible avec les statuts de protection nationale de la Posidonie

Pour l'herbier de Posidonie, 26 descripteurs d'état ont été identifiés. Ils sont regroupés en deux paramètres permettant l'évaluation de l'état de conservation de l'herbier, « structures et fonctionnalités », ainsi que « menaces et pressions » (Fig. 3).



**Fig. 3 : Descripteurs des paramètres « structure et fonctionnalité » et « menaces et pression » en haut, au milieu les critères et en bas les descripteurs d'état (d'après Lepareur, 2011).**

Ces descripteurs d'état ont été sélectionnés par le MNHN en concertation lors de groupes de travail composés des experts français de la façade Méditerranée (CSRPN). Ils ont été sélectionnés sans regard particulier sur leur faisabilité ou leur priorisation de mise en œuvre, mais pour cadrer le travail des opérateurs.

### Résultats

CARTHAM a permis une cartographie des biocénoses, sur la base des habitats d'intérêt communautaire, quasi continu de la bande côtière française.

Les **descripteurs utilisés pour qualifier l'EC de l'herbier de Posidonie** n'ont pas été imposés aux opérateurs qui se sont appuyés sur leur propre expérience, les réseaux de surveillance et la liste exhaustive du MNHN. Le renseignement de chaque descripteur a été contraint par des niveaux d'investigation variables dépendant principalement de l'offre retenue dans le cadre de la mise en concurrence des opérateurs. L'offre retenue correspondant généralement au meilleur rapport qualité-prix : qualité sur les réponses apportées par rapport au cahier des charges CARTHAM et coût par rapport à l'enveloppe globale du programme.

L'utilisation des descripteurs est la suivante (en pourcentage de sites où le descripteur est renseigné) :

**[100%]** : substrat, structures érosives, limite inférieure, espèces « patrimoniales », blocs de matte arrachés, caulerpes, macrodéchets ;

**[+50%]** : densité à -15 mètres, recouvrement foliaire, déchaussement, degré d'artificialisation, remaniement sédimentaire, sources de nuisances potentielles, hydrocarbures ;

**[-50%]** : rhizomes plagiotropes, compacité de la matte, densité foliaire, longueur moyenne des faisceaux, Rhodobiontes, couche oxydée ;

[0%] : structure de la matre, surface foliaire par faisceau, production de feuilles, croissance des rhizomes.

Les descripteurs de « menaces et pressions » ont été davantage renseignés que les descripteurs de « structure et fonctionnalité ».

Les descripteurs du paramètre « menaces et pressions » ont été renseignés selon les recommandations des grilles de descripteurs d'état du MNHN. Ils permettent d'interpréter des tendances ou évolutions visibles de l'herbier. Il s'agit pour certains d'une interprétation qualitative permettant l'interprétation des résultats ouvrant alors la possibilité d'une étude plus approfondie d'un site dans le cas de problème particulier.

Pour certains descripteurs du paramètre « structure et fonctionnalité », les opérateurs ont mis en œuvre d'autres méthodes, répondant mieux aux moyens financiers et techniques liés à l'offre. Les descripteurs concernant la matre morte de l'herbier ont été visuellement appréciés ou non renseignés. Les opérateurs ont essayé au maximum de répondre aux recommandations du MNHN. Les méthodes non développées sont généralement celles les plus lourdes, nécessitant de nombreux traitements coûteux ou des prélèvements.

Au final, l'interprétation des informations s'appuie sur l'analyse des experts, qui les rassemblent afin de qualifier l'EC de l'habitat.

Concernant le paramètre de **superficie de l'habitat**, il a été systématiquement renseigné. Les surfaces obtenues s'appuient donc sur des données historiques (généralement moins de 10 ans) et complémentaires (Fig. 4).

## Discussions

Le cahier des charges national élaboré pour répondre à CARTHAM n'a pas permis d'avoir un effort d'investigation homogène sur les 26 aires marines protégées concernées pour la Méditerranée.

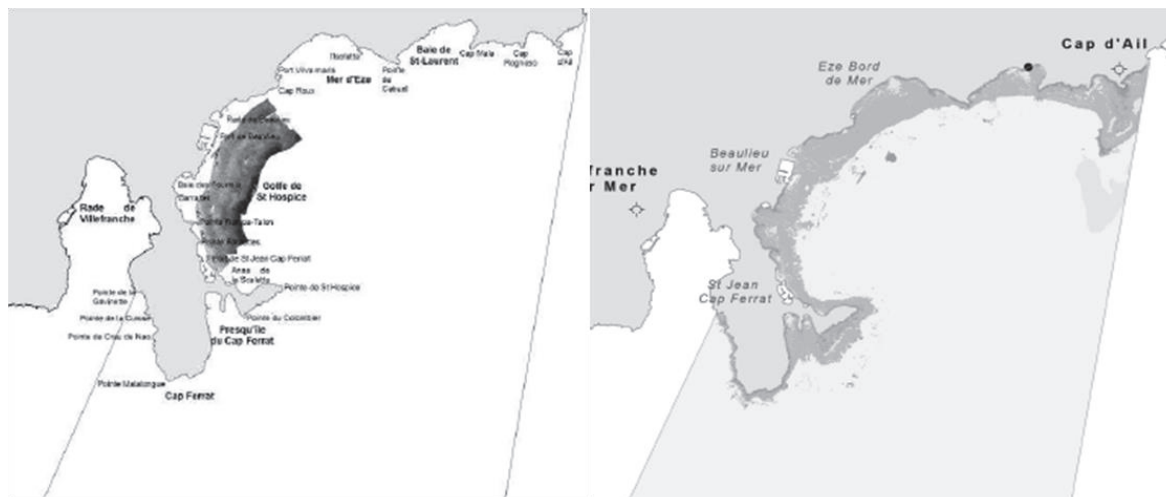
La grille et les descripteurs permettent d'avoir une vue cohérente des différences d'acquisition de données entre les opérateurs. Ils ont également l'intérêt d'objectiver le dire d'expert en précisant les interprétations. A ce stade le dire d'expert reste l'unique moyen d'avoir une interprétation agrégative des différents descripteurs relevés.

↳ **La mise en œuvre d'une approche écosystémique pour évaluer l'état de l'herbier à *Posidonia oceanica* basé sur l'EBQI : ecosystem-based index of the quality of its functioning (Personnic *et al*, 2014) permettrait de s'affranchir d'une partie du dire d'expert.**

Certains descripteurs ont été relevés sans méthode spécifique, basés sur l'appréciation de l'observateur. La méthode ici est donc peu reproductible.

↳ **Les différentes méthodes d'acquisition doivent être explicitées et reproductibles pour permettre une analyse comparative.**

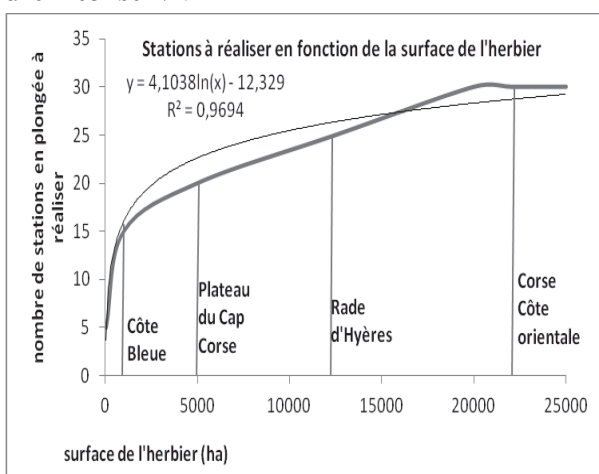
La stratégie d'échantillonnage n'a pas fait l'objet de préconisation particulière. Pour suivre les changements de l'herbier il est proposé d'effectuer une station de suivi *in situ* par hectare d'herbier de Posidonie (Montefalcone *et al.*, 2013). Une telle préconisation est économiquement insoutenable. En effet à titre d'exemple la surface de l'herbier de la côte orientale de la Corse est d'environ 22 000 hectares, ce qui impliquerait un suivi sur 22 000 stations. Les herbiers de Posidonie ont des superficies très variables selon les AMP. En compilant le nombre de stations échantillonnées en plongée en fonction de la surface d'herbier considéré, il est possible de créer une courbe indicatrice du nombre de stations à échantillonner suivant la surface d'herbier (Fig. 6).



**Fig. 4 : [Gauche] extraction de la carte mosaïque sonar réalisée en 2010 pour CARTHAM et [droite] cartographie de l'herbier de Posidonie d'une partie du site Natura 2000 du Cap Ferrat (Alpes maritimes)**

La stratégie d'échantillonnage n'a pas fait l'objet de préconisation particulière. Pour suivre les changements de l'herbier il est proposé d'effectuer une station de suivi *in situ* par hectare d'herbier de Posidonie (Montefalcone *et al.*, 2013). Une telle préconisation est économiquement insoutenable. En effet à titre d'exemple la surface de l'herbier de la côte orientale de la Corse est d'environ 22 000 hectares, ce qui impliquerait un suivi sur 22 000 stations. Les herbiers de Posidonie ont des superficies très variables selon les AMP. En compilant le nombre de stations échantillonnées en plongée en fonction de la surface d'herbier considéré, il est possible de créer une courbe indicatrice du nombre de stations à échantillonner suivant la surface d'herbier (Fig. 5).

↳ **Une courbe nombre de station de suivi/surface de l'herbier, consolidée et partagée avec les experts, apporterait un élément intéressant pour évaluer l'effort et le coût d'un tel suivi.**



**Fig. 5: Courbe du nombre de stations à échantillonner en plongée en fonction de la surface de l'herbier. En noire, la courbe logarithmique des tendances.**

D'autres questions sont soulevées ici :

- Comment choisir une station (de manière aléatoire, en fonction des pressions et des menaces, une station déjà suivie, *etc.*) ?
- A quelles profondeurs doivent se trouver ces stations ? Doit-on comme pour la Directive Cadre sur l'Eau (DCE) choisir des stations une profondeur de 15m ?
- Quel est la périodicité d'échantillonnage ?



Le paramètre de superficie de l'habitat ne permet pas une analyse fine et localisée de l'herbier. Ce paramètre a été renseigné par des acquisitions de données étalées dans le temps. Pour suivre ce paramètre et avoir une vraie analyse évolutive de l'herbier, il est nécessaire avoir une acquisition sonar sur une période restreinte (quelques semaines) pour pouvoir reproduire la même opération dans quelques années. Pour évaluer l'évolution de la surface de l'herbier dans six ans (évaluation imposée par la Commission Européenne pour le rapportage de l'article 17 de la DHFF), seules les données récentes pourront faire l'objet d'une comparaison.

Sur un sonogramme, en conditions favorables, la limite de détection de l'herbier apparaît pour un recouvrement de 25% (Philippe Clabaut, com.pers.). Il est donc difficile de mettre en évidence des zones de progression, due à la faible vitesse de croissance des rhizomes. Considérant cela, il est difficile de délimiter un herbier dans le cas de limite diffuse et donc d'en définir sa superficie. Une variabilité acceptable devrait être définie statistiquement et associée à ce paramètre. Les outils géomatiques permettent aujourd'hui de faire de comparaison de surface précise et d'identifier finement les secteurs sous pression.

↳ **Le paramètre de la superficie de l'habitat doit être évalué à la hauteur de sa sensibilité et de son coût d'acquisition élevé. Nous estimons que ce paramètre pourrait être relevé tous les 6 à 10 ans.**

Le rapportage de l'EC peut se faire à différentes échelles géographiques :

- Suivi d'une pression, d'une menace sur un habitat (quelques dizaines de mètres carrés) ;
- Suivi d'un habitat dans une aire marine protégée (plusieurs dizaines d'hectares) ;
- Suivi d'un habitat à l'échelle d'une façade nationale (plusieurs centaines de kilomètres) ;
- Suivi d'un habitat à l'échelle du bassin Méditerranéen (plusieurs centaines de kilomètres carrés).

Or, il n'existe pas de méthode pour définir si un état favorable de conservation sur plusieurs AMP peut correspondre à un état favorable de la façade.

↳ **Dans une recherche de rationalisation des moyens et donc une réduction des coûts liés aux suivis des herbiers de Posidonie, la démarche d'évaluation à multi-échelle est à construire.**

Il ne faut cependant pas oublier que d'autres habitats doivent être suivis parallèlement à l'herbier, ce qui implique une réflexion plus poussée et générale sur la surveillance des habitats et son coût.

## Remerciements

P. Clabaut, C.F. Boudouresque (MIO), F. Lepareur (MNHN), S. Personnic (MIO), R. David (IMBE), G. Pergent (Université de Corse) ainsi que l'ensemble des membres des CSRPN de Méditerranée pour leur implication sur CARTHAM.

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## ***POSIDONIA OCEANICA* MEADOWS IN GREEK SEAS: LOWER DEPTH LIMITS AND MEADOW DENSITIES**

### **Abstract**

*The current study aims to present the main characteristics (depth of lower limit and shoot density at a fixed depth of 15m) of *Posidonia oceanica* meadows in Greek coasts. All sampling sites were chosen at pristine (undisturbed) areas and are equally distributed geographically (10) in each one of the three main divisions of the Greek seas: North Aegean Sea, South Aegean Sea and Ionian Sea. In total, *P. oceanica* meadows were studied in 30 sampling sites. Based on our results, the mean value of the lower depth limit of *P. oceanica* meadow in the North Aegean Sea was  $26.3 \pm 6.44$  meters, while the mean value of the shoot density was  $343.1 \pm 104.58$  shoots/m<sup>2</sup>. In the South Aegean Sea, the mean values of the lower depth limit and shoot density of *P. oceanica* meadow were slightly higher,  $30.0 \pm 5.75$  meters depth and  $430.2 \pm 87.97$  shoots/m<sup>2</sup> respectively. In the Ionian Sea, the mean values of lower depth limit of *P. oceanica* meadow ( $35.4 \pm 4.95$  meters depth) and shoot density ( $470.19 \pm 69.33$  shoots/m<sup>2</sup>) were higher than the corresponding values of the Aegean Sea. Since all of our sampling sites correspond to undisturbed conditions the above mentioned values of lower depth limit and shoot density of each division of the Greek seas (North Aegean Sea, South Aegean Sea, and Ionian Sea) could be used for the calculation of reference conditions values for the estimation of the ecological status of all Greek seas' meadows.*

**Key-words:** *Posidonia oceanica*, lower depth limit, shoot density, Mediterranean, Greece

### **Introduction**

Being perennial sessile organisms, marine angiosperms respond directly to abiotic and biotic environmental changes and thus, represent sensitive indicators of the coastal ecosystem status (Pergent *et al.*, 1995; Romero *et al.*, 2007; Montefalcone, 2009). A typical representative of marine angiosperms is the Mediterranean endemic species *Posidonia oceanica* (Linnaeus) Delile which constitutes a key species for the Mediterranean ecosystem (Pergent, 1992). *P. oceanica* is a very well-studied seagrass in the Western Mediterranean Sea, while in the case of the Greek seas there is rather sparse and limited information.

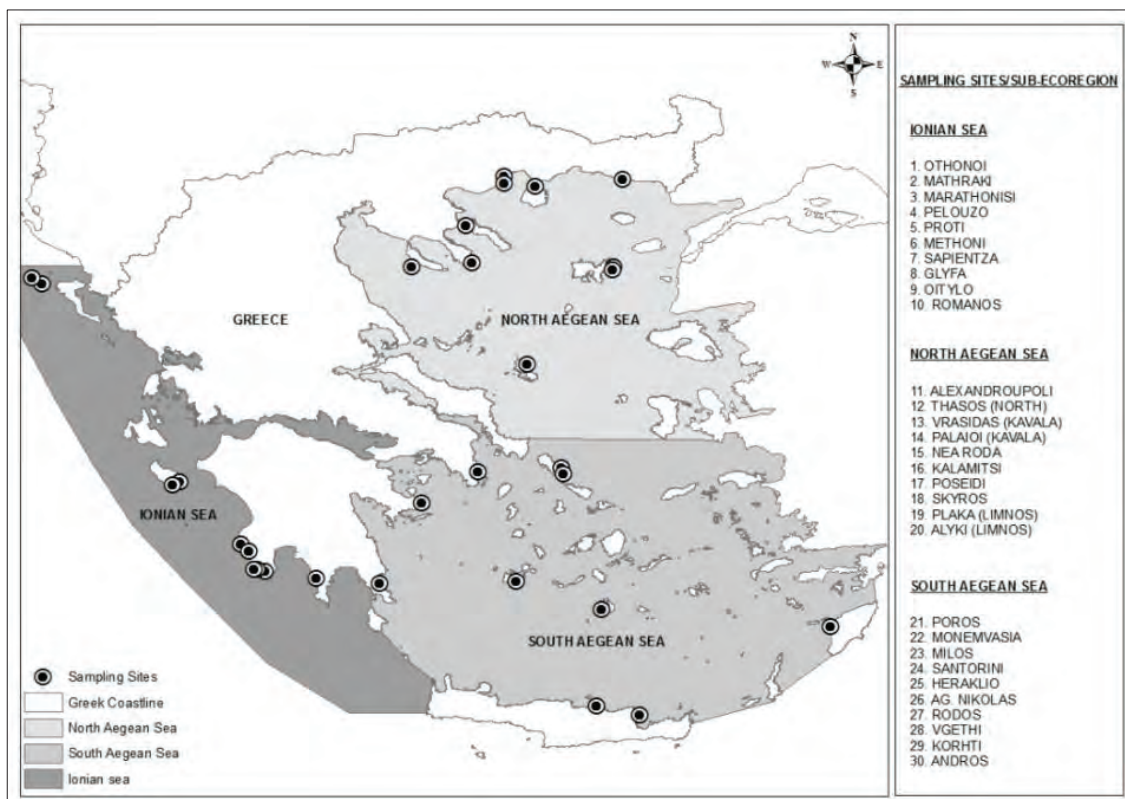
In Greece the seascape of the Aegean and Ionian Sea encompasses a large variety of hydrological, oceanographic and biogeographic features. The Aegean Sea in particular, displays a complex hydrographic and ecological structure due to its geographical position between the Black Sea, the Ionian Sea and Levantine Sea (Zervakis *et al.*, 2004). The Aegean Sea is separated by the Cyclades plateau into two sub-basins, the North Aegean and the South Aegean, with different hydrographic characteristics due to the influence of Black Sea waters and Levantine Sea waters respectively. Consequently, the following sub-ecoregions are adopted for the Greek seas: a) the North Aegean Sea, with its coastal water masses being affected by the extending continental shelf zone, the transboundary rivers of northern Greece and the Black Sea waters, thus, showing trends of natural

eutrophication. b) the South Aegean Sea, with its coastal water masses being affected by the extending continental shelf zone of the Cyclades plateau and the Asia Minor stream. Here, the open coastal waters are typically oligotrophic, while phenomena of anthropogenic eutrophication may occur in enclosed embayments; and c) the Ionian Sea, being characterized by a narrow shelf zone with steep slopes, resulting in mostly deep and oligotrophic coastal water masses.

The aim of this study is to describe two of the main characteristics of *P. oceanica* meadows in each one of the sub-ecoregions of the Greek seas: Lower Depth Limit and meadow density, which are two of the most commonly used metrics in *P. oceanica* monitoring. In addition, we propose a preliminary definition of Reference Conditions (RC) in the Greek sub-ecoregions, based on a large *P. oceanica* meadows dataset, extracted through a long sampling effort along the Greek coasts for the implementation, evaluation and intercalibration of the proposed *Posidonia* indices (POMI; PREI; Valencia CS) (Romero *et al.*, 2007; Fernandez-Torquemada *et al.*, 2008; Gobert *et al.*, 2009).

### Materials and methods

*Posidonia oceanica* meadows that were subject only to minor human impact were studied in 30 sites along the Greek coasts (Fig. 1) during the last three years (2012-2014).



**Fig. 1: The sub-ecoregions of Ionian Sea, North Aegean Sea, South Aegean Sea and the sampling sites of *P. oceanica* meadows.**

At each sub-ecoregion ten (10) geographically distributed *P. oceanica* meadows were studied. Both metrics, the lower depth limit and shoot density (at a constant depth of 15m), were recorded and measured *in situ* through scuba diving. These metrics were selected because they provide information on the vitality of the meadow (at population level) for several disturbances (water transparency, sedimentary dynamics, trawling) commonly met

in the Mediterranean Sea. Reference values were defined for each metric in each sub-ecoregion on the basis of spatial data (i.e. existing sites in undisturbed conditions). In more detail, the reference value for lower depth limit was defined by the maximum value in each sub-ecoregion. For shoot density, the reference value was calculated as the mean of the three higher values of the considered metric in order to buffer possible outliers.

For both metrics, the statistical significance of differences between the sub-ecoregions was evaluated. Since the conditions of normality and homogeneity of distribution were not met, the non-parametric Kruskal-Wallis test was applied. The differences between sites were tested using the non-parametric Mann-Whitney U-test. All differences were considered significant at  $p < 0.05$ .

## Results

Based on our results (Tab. 1), the mean value of the lower depth limit of *P. oceanica* meadow in the North Aegean Sea was  $26.3 \pm 6.44$  meters, while the mean value of the shoot density was  $343.1 \pm 104.58$  shoots/m<sup>2</sup>. In the South Aegean Sea, the mean values of the lower depth limit and shoot density were not found to be significantly different from the above mentioned values of North Aegean Sea (Tab. 2), despite being a bit higher:  $30.0 \pm 5.75$  meters depth and  $430.2 \pm 87.97$  shoots/m<sup>2</sup> respectively. In the Ionian Sea, the mean values of lower depth limit ( $35.4 \pm 4.95$  m) and shoot density ( $470.2 \pm 69.33$  shoots/m<sup>2</sup>) were significantly higher than the corresponding values of the Aegean Sea. In addition, Reference Conditions values for each sub-ecoregion are shown in Table 1.

**Tab 1. Mean values and Reference Conditions (RC) of each *Posidonia* meadow metric at each sub-ecoregion of Greece.**

Sub-Ecoregion	Lower depth limit (m)		Shoot density (shoots/m <sup>2</sup> ) (at 15m depth)	
	mean $\pm$ SD	RC	mean $\pm$ SD	RC
North Aegean Sea	$26.3 \pm 6.44$	33	$343.1 \pm 104.58$	456
South Aegean Sea	$30.0 \pm 5.75$	38	$430.2 \pm 87.97$	506
Ionian Sea	$35.4 \pm 4.95$	44	$470.2 \pm 69.33$	553

**Tab. 2: Significant differences of *Posidonia* Lower Depth Limit (m) and Shoot Density (shoots/m<sup>2</sup>) between sub-ecoregions of Greece ( $p < 0.05$ ). ns = non-significant.**

Sub-Ecoregions	Lower Depth Limit (m)	Shoot Density (shoots/m <sup>2</sup> )
Ionian Sea - North Aegean	0.003	0.011
Ionian Sea - South Aegean	0.043	ns
South Aegean - North Aegean	ns	ns

## Discussion and conclusions

The lowest mean values of shoot density and lower depth limit at the North Aegean Sea sub-ecoregion (Tab. 1), in comparison with the other sub-ecoregions, indicate that *P. oceanica* meadows of the North Aegean Sea are clearly linked with the main hydrographic and ecological characteristics of the area: the nutrient rich, low saline, Black Sea water occupying the surface water layers (Stergiou *et al.*, 1997; Lykousis *et al.*, 2002), the river flows that locally enhance the productivity of the coastal zone (Karageorgis *et al.*, 2003), and the extending continental shelf. At the South Aegean Sea sub-ecoregion, the shoot density and lower depth limit values (Tab. 1) are higher than those recorded in



North Aegean Sea due to the different hydrographic characteristics of the South Aegean: the surface layers consist of highly saline waters of Levantine origin, the limited river flows, and the narrower shelf zone. The same applies for the sub-ecoregion of the Ionian Sea, whose characteristics (deep oligotrophic waters and narrow shelf zone with steep slopes) (Ramfos *et al.*, 2006) contribute to the presence of *P. oceanica* denser meadows and greater deeper limits (Tab. 1).

Finally, since all of our sampling sites correspond to undisturbed conditions the extracted Reference Conditions (RC) values for each Greek sub-ecoregion can be used for the estimation of the ecological status of all Greek seas' meadows in terms of the Water Framework Directive (2000/60/EC).

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## **ARE MEDITERRANEAN MPAs PROTECTING MARINE FORESTS?**

### **Abstract**

*In the Mediterranean Sea, Marine Protected Areas (MPAs) are nearly 700, covering approximately 5% of the sea surface, but merely 0.1% of the Mediterranean's total surface is included in no-take zones. Mediterranean MPAs are often established according to political or socio-economic criteria more than nature conservation aspects, and only less than half of them have a management plan or have evaluated the status and the distribution of marine habitats. Results from our literature-based research highlight that scientific studies are abundant only in few Mediterranean MPAs, generally the biggest and the long-established ones. Usually, it is often the case that on land the vegetation has a primary role in establishing protected areas. By contrast, in marine systems, the vegetation has a secondary role or in some cases is absent in the establishment of protected areas. Furthermore, in the most studied Mediterranean MPAs, there have been limited scientific research on the status of large brown algae (i.e. *Cystoseira* and *Sargassum*) forests. As a result of this lack of information, marine forests are generally not included in Mediterranean MPA management plans, making it difficult to assess their evolution and understand the potential role of MPAs in the conservation of marine forests. With this contribution, we would like to remark the importance of conducting research on marine forests of *Fucales* in MPAs that may represent priority sites for the conservation of healthy forests and for the recovery of degraded ones.*

**Key-words:** *Cystoseira*, marine protected areas, marine forests, algae, Mediterranean Sea

### **Introduction**

Since many centuries Mediterranean populations have exploited coastal ecosystems for their development. In the last decades this phenomenon intensified with increased pollution, fishing, tourism and an uncontrolled coastal urbanisation (Caddy, 1993; Airoidi, 2003; Ludwig *et al.*, 2009), so that important changes have occurred on coastal ecosystems (Airoidi & Beck, 2007) but, unluckily, they are difficult to quantify due to the lack of historical data. As a result, a set of conservation measures have been conceived at the regional, national or international level to protect this biodiversity (for a review see Micheli *et al.*, 2013). Nowadays, nearly 700 Marine Protected Areas (MPAs) have been established in the Mediterranean Sea, covering nearly 5% of its surface (including the Pelagos Sanctuary, representing alone 4% of the surface), while merely 0.1% of the Mediterranean total surface is included in no-take zones. Such percentages are even smaller if we consider only the 161 MPAs of national status that cover 0.73% of the Mediterranean Sea (Gabrié *et al.*, 2012). The 33 SPAMIs (SPA/BD Protocol) usually overlap with national MPAs and therefore in the following text, we include them in the general term MPAs. However, MPAs are often established more according to political or socio-economic choices than conservation aspects (Leenhardt *et al.*, 2013), and only less than half of them have a management plan or have evaluated the status and the distribution of marine habitats, among which algal forests. Indeed, contrarily to what generally happens on land, where vegetation has a priority role in the establishment of protected

areas (e.g. many forest-reserves were established in the world to reduce deforestation), marine forests are not considered in the creation of MPAs and they are generally not targeted in monitoring programs or in the evaluation of MPA efficacy. An exception is the CARLIT index, applied in the North-Western Mediterranean and in the Adriatic Seas in the framework of the Water Directive 2000/60/EU (Ballesteros *et al.*, 2007; Mangialajo *et al.*, 2008), that use the distribution of very shallow *Cystoseira* forests to assess the ecological status of coastal shallow waters. It is now widely accepted that marine forests of large brown seaweeds (represented by the genus *Cystoseira* and *Sargassum* in the Mediterranean Sea) are some of the most important marine habitats, forming extended canopies comparable to land forests and providing refuge and subsistence for many organisms, including fish (Jones *et al.*, 1994; Ballesteros *et al.*, 1998). However, many large brown forests are considered threatened worldwide and several studies described the loss of *Cystoseira* forests in the Mediterranean Sea (for a review see Airoidi *et al.*, same issue). Only few events of natural recovery have been reported at present, and especially in MPAs (Bonaviri *et al.*, 2009; Hereu & Quintana, 2012; author's personal data). MPAs could have a strong potential for conservation of large brown algae forests because they may guarantee protection from several kinds of direct (i.e. coastal development and destructive fishing) and indirect (cascade effects of overfishing) human impacts that are the major causes of loss of these forests (Gianni *et al.*, 2013). The aim of this work was to quantify the contribution of scientific research to the study of large brown algae forests in the Mediterranean MPAs in order to estimate the degree of knowledge of this habitat compared to other major habitats and/or species.

### Materials and methods

We identified Mediterranean MPAs using the MedPAN list (see Gabrié *et al.*, 2012). In our work we considered only 113 national MPAs out of 161, because we excluded coastal protected areas mainly characterised by wetlands or land, with no evidences for a real protection of the marine environment. We also considered 32 SPAMIs out of 33: we excluded the Pelagos Sanctuary because it is mostly characterized by pelagic waters. Natura 2000 sites were not considered in this work because most of them are still in the establishment process or lack a management plan, including a specific regulation for tourism or marine resources exploitation. We searched ISI Web of Science in order to quantify the number of international scientific papers produced on each MPAs. In this way we can obtain replicable results, although, unfortunately, grey literature or national papers are not considered. Papers were found searching the crossed topics:

- "name of the MPA" and
- "\* protected area\*" or "park" or "marine reserve\*" or "nat\* reserve\*" or "monument" or "Mediterranean".

We selected all studies performed in the MPAs focused on the marine environment, whether or not the authors considered the protection of the site as mandatory for their study. In addition, we evaluated the number of papers on algae, on Fucales, on seagrasses and on fish combining to the previous search the following topics, respectively:

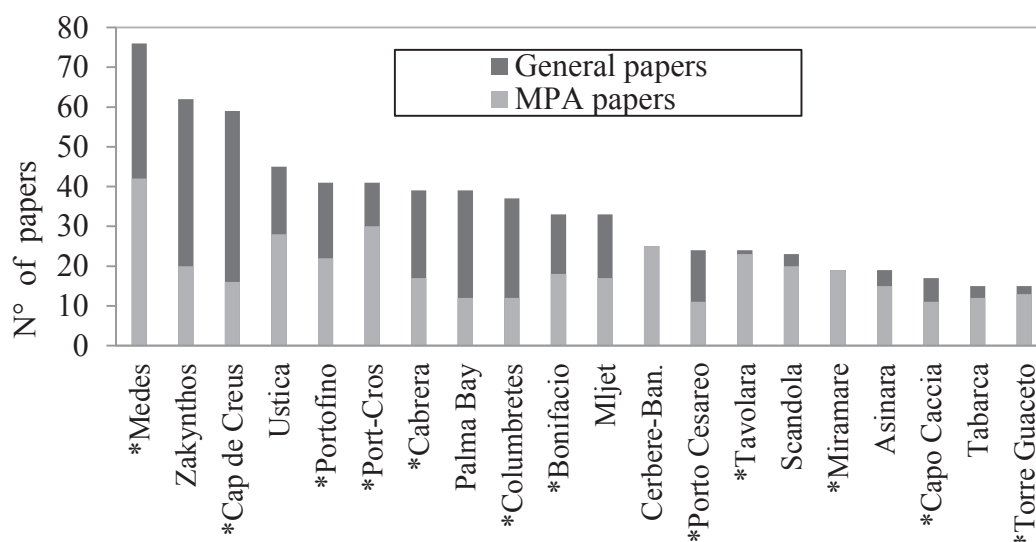
- "\*alga\*" or "seeweed"
- "Fucales" or "furoids" or "*Cystoseira*" or "*Sargassum*"
- "seagrass" or "*Posidonia*" or "*Cymodocea*" or "*Zostera*"
- "fish\*"

In this study, we considered papers with algae, seagrasses or fish as main subject, but also papers focused on other topics and reporting some analyses or observations on algae,

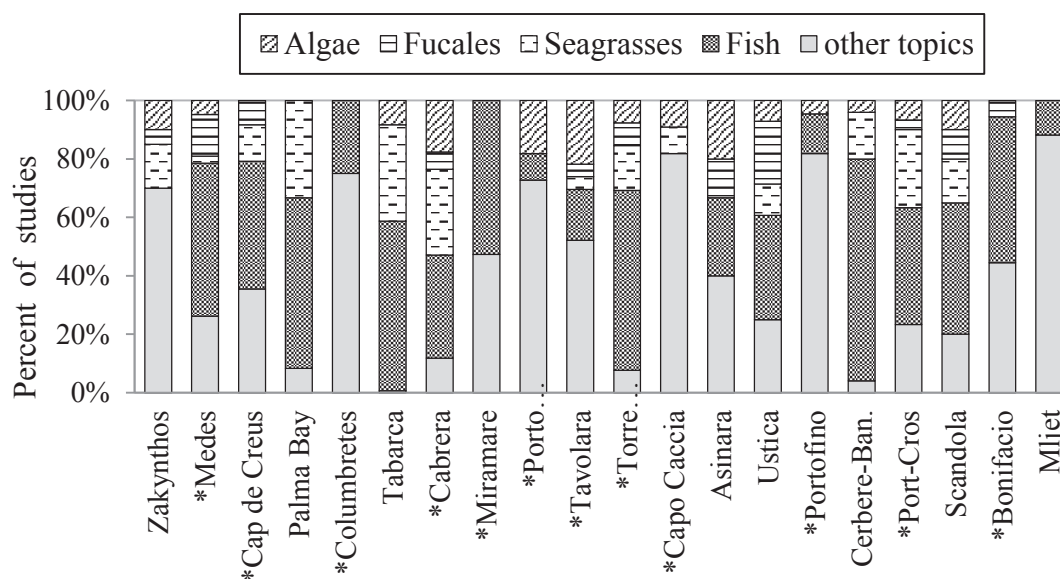
seagrasses or fish. All our searches are updated to July 2014. For the entire dataset and for each MPA, we identified the papers considering the protection as mandatory for the study (hereafter MPA papers) in contrast to the general studies not considering the protection (hereafter General papers) and we calculated the relative proportions of studies on algae, Fucales, seagrasses and fish.

## Results

In total, 1500 studies were carried out in the 113 MPAs (512 of which in the 32 SPAMIs), but merely 38% of them were specifically performed in those sites because they were protected (MPA papers). Studies on marine vegetation (algae and seagrasses) represented 15% of all studies and 26% of MPA papers. Papers on algae represented 9% of all studies and 14% of MPA papers (40% of which centred on Fucales). Studies on seagrasses represented about 6% of all studies (mostly on *Posidonia oceanica*) and 12% of MPA papers. Studies on fish assemblages represented 22% of all studies and 36% of MPA papers. Such percentages were coherent with the ones calculated only on SPAMIs. Only 20 out of the 113 Mediterranean MPAs with national status had at least 10 MPA papers published in international journals (henceforth it will be mentioned as Highly studied MPAs), 12 of which are also SPAMIs. In Figure 1 the amount of MPA and General papers are reported for the Highly studied Mediterranean MPAs. The totality of studies performed at Miramare and Cerbère-Banyuls MPAs were focused around the protection effect (MPA papers). A great percentage of MPA papers (> 70%) was also found for Tabarca, Tavolara-Punta Coda Cavallo, Torre Guaceto, Asinara, Port-Cros and Scandola MPAs. On the contrary, in Zakynthos, Cap de Creus, Palma Bay and Columbretes islands only 30% of studies was performed there because of the protected status of the site. The relative proportion of papers on algae (excluding Fucales), on Fucales, seagrasses and fish respect to the number of MPA papers carried out at the highly studied MPAs is reported in Figure 2.



**Fig 1: Amount of papers considering the protection as mandatory for the study (MPA papers) and of papers not considering the protection as mandatory (General papers) in highly studied (at least 10 MPA papers) MPAs. MPAs with an asterisk are also SPAMIs.**



**Fig. 2: Percentages of studies on algae (excluding Fucales), Fucales, seagrasses, fish and “other topics” in MPA papers (considering the protection as mandatory for the study) for highly studied MPAs (at least 10 MPA papers). MPAs with an asterisk are also SPAMIs.**

The highest percentage of studies on algae, including also Fucales, was done in the protected areas of Ustica, Asinara and Tavolara (30-40%). In almost all other MPAs, papers considering algae represented less than 20%. The percentage of studies on Fucales was also lower in all MPAs (< 15%), except for Ustica (21%). Papers on seagrasses were mostly carried out in Palma Bay, Tabarca island, Cabrera archipelago and Port-Cros, where they represented nearly 30% of MPA papers. In our search we did not find any study on marine vegetation of the protected areas of Columbretes, Miramare and Mljet. In contrast, the percentages of papers on fish were almost always greater than the ones on marine vegetation (> 30%). Torre Guaceto and Cerbère-Banyuls were the MPAs with the highest percentage of studies on fish (> 60%).

## Discussion

According to the Convention of Biological Diversity (reviewed in 2010), by 2020 10% of the Mediterranean Sea’s surface should be protected. In order for this to be a reality, more MPAs have to be established. However, as we showed in this work, in many national MPAs a big amount of research did not consider the protection as mandatory (General papers). This is partially due to the fact that data previous MPA establishment are lacking, so it is often difficult to assess the effect of protection on some habitats (i.e. marine vegetation). Only 20 out of 113 MPAs can be considered as highly studied MPAs. Such MPAs include the biggest and/or the long-established ones and most of them belong to France, Italy and Spain, the Mediterranean countries where most of marine research published on international journals is carried out. For instance, many North-African and Middle-East MPAs were established more than 30 years ago, but few international studies are available. Information on marine forests potentially exists in these MPAs, but it is hardly accessible, consisting in grey literature or being written in other languages than English. The highest percentage of studies carried out in Mediterranean MPAs is focused on fish assemblages, the compartment usually more facilitated by the protection (generally fishery limitations). Seagrasses, and in particular *Posidonia oceanica* meadows, were object of several studies in different MPAs, since their

ecological role is widely recognised (Personnic *et al.*, 2014). The amount of studies on *P. oceanica* was comparable to the amount of studies on all algae. Indeed, we found a low percentage of studies focused on *Cystoseira* and *Sargassum* and they were astonishingly almost absent in many well-managed MPAs where it is still possible to find dense marine forests (e.g. Tavolara-Punta Coda Cavallo, Columbretes). Ustica MPA had the higher number of papers on *Cystoseira* forests, because many studies investigated the cascade effects of sea urchins proliferations on benthos and highlighted the loss and then a gradual recovery of macroalgal assemblages (among others: Gianguzza *et al.*, 2006; Bonaviri *et al.*, 2009).

Although *Cystoseira* species are listed in the annexes of some European Conventions (Barcelona Convention, 1976 and Bern Convention, 1979), they are still unprotected and little studied. In order to ameliorate the conservation of marine forests in the Mediterranean Sea, the implementation process of MPAs has to be improved and the management has to be planned on the base of a complete and detailed habitat mapping of marine vegetation. The establishment of Natura 2000 marine sites is often accompanied by the cartography of habitats (e.g. in France), following the Habitat Directive annexes (Directive 92/43 EEC). *Posidonia oceanica*, priority species for this Directive, is therefore well documented in Natura 2000 sites and we have now good information on its health and evolution. Unluckily, large brown seaweeds (i.e. *Cystoseira* species), although mentioned in the previous Conventions, are not included in the Habitat Directive, so that the cartography done in the Natura 2000 sites usually reports merely “photophilous algae on rocky bottom”, without any specification if macroalgal communities are deserts of encrusting corallinales, filamentous algae, shrubs of photophilous algae or forests of large brown algae. In conclusion, results of our work highlighted a lack of information on marine forests in Mediterranean MPAs, especially compared to other communities (i.e. fish assemblages or *P. oceanica* meadows). Although grey literature and old papers were excluded by our search, we are confident that the results would have been comparable to the ones obtained with the search on ISI Web of Science.

MPAs can theoretically have an important role for marine forests conservation. Unluckily, the current state of knowledge does not allow assessing if present regulations in MPAs are protecting healthy marine forests and/or enhancing natural recovery of the degraded ones (Sala *et al.*, 2012). Protection of Mediterranean coastal ecosystems should be evaluated with a constant monitoring of the distribution and status of marine forests and we suggest that future research priority should be the assessment of the conservation status of Fucales in particular in the MPAs, in order to better understand the real role of present protection rules in the conservation of healthy forests and, potentially, in the restoration of damaged ones.

### Acknowledgments

Authors want to thank Claudia Scianna that helped to list Mediterranean MPAs. The work was realized within the framework of the project MMMPA-Training Network for Monitoring Mediterranean Marine Protected Areas, funded by the European Community's Seventh Framework Programme (FP7/2007-2013) [grant n° 290056].

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## DEVELOPMENT OF THE NATIONAL MONITORING PROTOCOL FOR *POSIDONIA OCEANICA* MEADOWS IN CROATIA: A PILOT PROJECT

### Abstract

*In the framework of the MedMPAnet Project, we aimed at developing a monitoring protocol for the habitat type 1120 Posidonia beds to assist Croatia in fulfilling the requirements of the Habitats Directive. Field activities for assessing the status of Posidonia oceanica meadows were carried out in five sites within the Primorje-Gorski Kotar County (Northern Adriatic Sea). At each site, the survey was performed at nine stations and three different depths. Non-destructive sampling was done using traditional descriptors widely applied in nearly all Mediterranean P. oceanica monitoring programmes. Shoot density and the surface of different substrate types were assessed in the field; percentage covers of live P. oceanica and dead mat were then used to calculate the Conservation Index (CI). Values of density and CI were averaged for each station and each bathymetric range, then plotted to obtain an overview of the status of the investigated meadows. Meadows were then classified according to the values of density and the descriptors of lower limit (depth, type and coverage) following standard thresholds used in the Mediterranean to get information on conservation status of the meadows.*

*Posidonia oceanica meadows showed clear signs of deterioration in three sites, two of them being close to direct sources of anthropogenic pressures. The collected data are preliminary and could serve as a baseline for future 6-year surveillance cycles within the framework of national monitoring programme to report status of this priority habitat according to the provisions of the EU Habitats Directive.*

**Key-words:** Monitoring, *Posidonia oceanica*, Croatia, Northern Adriatic, SCUBA methods.

### Introduction

*Posidonia oceanica* (L) Delile (Magnoliophyta, Posidoniaceae) is present on almost all coasts of the Mediterranean (Boudouresque *et al.*, 2006); under optimal conditions of water transparency it forms extended meadows up to over 40 m deep. Because of their ecological functions and the socioeconomic relevance (Boudouresque *et al.*, 2006) as well as their vulnerability and decline in many areas of the Mediterranean (Boudouresque *et al.*, 2006; Di Carlo *et al.*, 2011), *P. oceanica* meadows are protected under the Habitats Directive 92/43/EU (priority habitat in Annex I, Natura 2000 Code 1120) and other international agreements (e.g. Barcelona and Bern conventions).

Although *P. oceanica* meadows are common along the Croatian coast (Benacchio, 1938; Bakran-Petricioli, 2007; 2011; Gamulin-Brida, 1967; Zavodnik, 1983; Zavodnik *et al.*, 2005), they are insufficiently studied and there is no precise information about their distribution and condition (Kružić, 2008; VV.AA., 2012; Zavodnik *et al.*, 2005).

Recently, under the WFD (2000/60/EC) the *Posidonia oceanica* Multivariate Index (POMI, Mascarò *et al.*, 2012; Romero *et al.*, 2007) has been applied for monitoring the environmental quality of coastal waters in several sites throughout the Croatian coasts (Mascarò *et al.*, 2012). However, this index is designed to provide indications on the

ecological status of coastal waters and is based on the combination of multiple descriptors of the meadows to be measured and/or assessed at 15 m depth (Romero *et al.*, 2007). Hence, its application does not provide any information on other parts of the meadows (e.g. the shallower areas and the lower limit) that are particularly vulnerable (Boudouresque *et al.*, 2006).

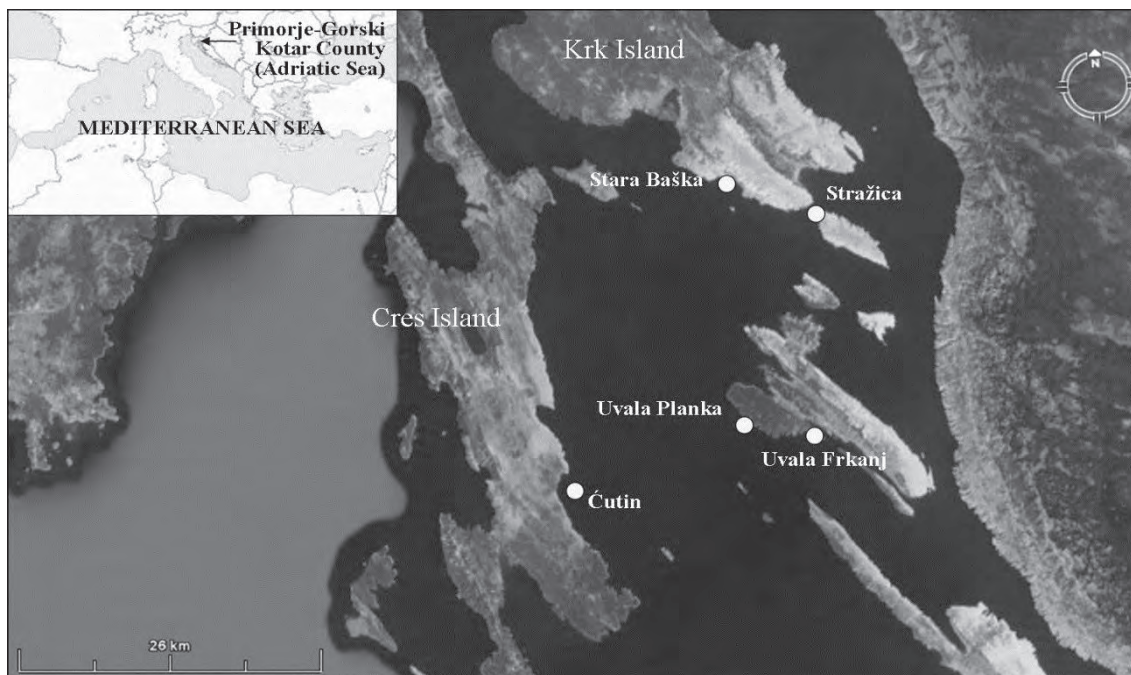
A monitoring protocol addressed to gather data useful to assess the conservation status of *P. oceanica* meadows in Croatia has been developed within the MedMPAnet Project. The protocol is based on non-destructive approaches and uses the most fundamental descriptors that are present in programmes to measure conservation status of *P. oceanica* in nearly all Mediterranean countries (Pergent-Martini *et al.*, 2005). This paper reports the results of a pilot activity addressed to assess the conservation status of *P. oceanica* meadows in five sites of Northern Adriatic Sea.

### **Material and methods**

The study was carried out in September 2013 in five meadows within the Primorje-Gorski Kotar County (Northern Adriatic Sea, Fig. 1): Stara Baška (Krk Island), Ćutin (Cres Island), Uvala Planka (Rab Island), Uvala Frkanj (Rab Island) and Stražica (Prvić Island). At each meadow, general information on the features of nearby coasts and potential human pressures were recorded; nine monitoring stations were selected at three depth ranges: shallow ( $8\pm 2$  m), intermediate ( $15\pm 2$  m) and close to lower limit.

For each meadow, we assessed the most fundamental descriptors used in programmes to evaluate the structure of *Posidonia oceanica* meadows and their conservation status (Buia *et al.* 2004; Marcos-Diego *et al.*, 2000; Pergent-Martini *et al.*, 2005; Pergent *et al.*, 1995; UNEP-RAC/SPA, 2011). Surveys were carried out by means of SCUBA diving without removal of biological material. Shoot density was detected by counting the number of leaf shoots within eight random replicated quadrats (40 x 40 cm); percentage of seabed covered with live plants of *P. oceanica* compared to that non-covered and consisting of dead matte, sand, or rock (Buia *et al.*, 2004) was assessed using the Line Intercept Transect technique (Bianchi *et al.*, 2004; Montefalcone *et al.*, 2007), four transects in each station. In addition, we recorded the depth of upper and lower limits and the limit typology as progressive, sharp, erosive, sparse or regressive (Montefalcone, 2009; Pergent *et al.*, 1995; UNEP-RAC/SPA, 2011).

Shoot density, percentage cover of live *P. oceanica* and dead matte and Conservation Index (CI, Moreno *et al.*, 2001), were calculated for each replicate. Values of density and CI were averaged for each station and each bathymetric range, then plotted to obtain an overview of the status of the investigated meadows. Since there is no baseline for Croatia, the density and the descriptors of lower limit (coverage, type and depth) were used to classify the meadows into five classes of conservation status (high, good, moderate, poor, bad) according to the standardized scale suggested by UNEP-RAC/SPA (2011).



**Fig. 1: Study area and monitoring sites.**

## Results

A total of 42 monitoring stations were surveyed; in Stara Baška only shallow and intermediate depth ranges were investigated because the deep meadow was very sparse, with small patches on dead matte and single shoots sparse until 26 m.

Table 1 summarizes, for each meadow, the information on the characteristics of the adjacent coast, the potential human pressures documented during the fieldwork and some meadow features.

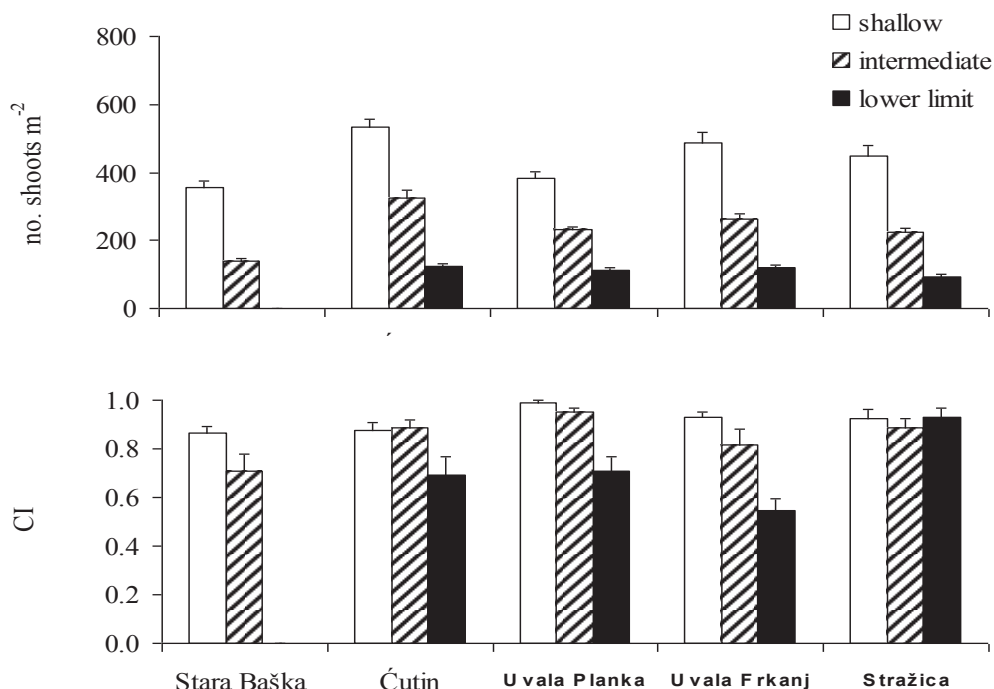
**Tab. 1: Coast features, potential human pressures and meadow features in the five studied sites (nr = not recorded).**

Site	Stara Baška	Čutin	Uvala Planka	Uvala Frkanj	Stražica
<b>coastal features</b>	rocky shore with coves and beaches exposed to SW	small rocky islands exposed to I and II quadrants	rocky coast exposed from SE to NW	rocky coast exposed from SE to W	rocky coast exposed to W-SW and E-NE
<b>Potential human pressures along the coast</b>	anchoring, mooring, wastewater, camping area, recreational beaches	no	boat anchoring	anchoring, mooring, tourist structures, Rab town about 1 NM away	Baška town about 2 NM away
<b>general description of the meadow</b>	flat and discontinued meadow	flat, continuous meadow	steep, continuous meadow	steep meadow, patchy at shallow stands	steep meadow, upper limit on rock and dead matte
<b>main substrate</b>	sand, matte	rock, sand	rock, sand	matte, sand, sparse rocks	rock, matte, sand
<b>upper limit, depth</b>	6 m	6 m	6-12 m	5-13 m	5-9 m
<b>lower limit, <i>P. oceanica</i> cover</b>	nr	56%	57%	53%	53%
<b>lower limit, type</b>	regressive	regressive	progressive	regressive	progressive
<b>lower limit, depth</b>	26 m	26.5 m	27.8 m	26 m	28 m



Meadow density is provided as mean values ( $\pm$ SE) between the stations at each depth (Fig. 1). The lowest densities were detected in Stara Baška ( $355\pm 22$  and  $141\pm 8$  shoots  $m^{-2}$  at shallow and intermediate stations, respectively) and in Stražica ( $92\pm 10$  shoots  $m^{-2}$  at the lower limit); the highest values in Čutin at all three depth ranges:  $534\pm 22$  (shallow),  $324\pm 23$  (intermediate) and  $124\pm 9$  shoots  $m^{-2}$  (lower limit).

At shallow and intermediate depths, values of CI (mean values between the stations at each depth  $\pm$  SE) were lower in Stara Baška ( $0.86\pm 0.03$  and  $0.71\pm 0.07$ , respectively) and higher in Uvala Planka ( $0.99\pm 0.01$  and  $0.95\pm 0.02$ ); at the lower limit they varied from  $0.55\pm 0.05$  in Uvala Frkanj to  $0.93\pm 0.04$  shoots  $m^{-2}$  in Stražica (Fig. 1).



**Fig. 2: Mean values ( $\pm$ SE) of the shoot density (above) and CI (below) of each depth range in each meadow.**

## Discussion

Although interpretation of data limited to one occasional survey can provide uncertain and tentative information, the findings of the field work have provided an initial insight on the current status of the meadows within the Primorje-Gorski Kotar County.

At all sites, at least two of the descriptors used to assess the conservation status of the meadows were found negative (bad or poor, Tab. 2). *Posidonia oceanica* shows clear signs of deterioration in Stara Baška and Uvala Frkanj that are the most impacted sites among the regions investigated. Unexpected results were found for the meadow of Čutin where, the abundance of dead matte was high and the lower limit was recorded shallow and regressive, despite the absence of evident anthropogenic pressures. It should be noted that Čutin showed the highest values of density and moderate conservation status at intermediate stations; strong hydrodynamics in the shallow and over-sedimentation in the depth could account for the unfavourable conditions of this meadow.

The meadows in Uvala Planka and Stražica have the deepest lower limit and both are progressive; low abundance of dead matte was recorded and, as consequence, the highest



values of CI were calculated. On the contrary, shoot density was low for both meadows at all the investigated depths.

**Tab. 2: Conservation status of investigated meadows based on the interpretation of descriptors as proposed by UNEP-RAC/SPA (2011); B = bad, P = poor, M = moderate, G = good, H = high, nr = not recorded.**

Site	Stara Baška	Ćutin	Uvala Planka	Uvala Frkanj	Stražica
density (shallow stations)	P	M	P	M	M
density (intermediate stations)	P	M	P	M	P
density (lower limit)	nr	P	P	P	P
lower limit, depth	P	P	M	P	M
lower limit, type	B	B	H	B	H
lower limit, % cover <i>P. oceanica</i>	nr	H	H	H	H

These results confirm what is well known for the Northern Adriatic about the lack of good conditions for plant growth due to low water transparency (Bakran-Petricioli *et al.*, 2006; Gamulin-Brida, 1967). However, since data are not enough to assess unambiguously the ecological status of the meadows, field status should be reviewed over additional monitoring cycles and future surveillance programmes. Actually, the Habitats Directive requires the development of monitoring programs aimed to evaluate the conservation status of *P. oceanica* meadows and to detect changes over time and, in agreement with its Art.17, only at the end of the 6-year cycle the condition of the habitat type in the whole biogeographical region, could be defined as favourable or unfavourable.

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## **ECOLOGY AND PERTURBATIONS OF MEDITERRANEAN DEEP-WATER ALGAL COMMUNITIES: LINKING POPULATION BIOLOGY AND COMMUNITY ECOLOGY FOR CONSERVATION**

### **Abstract**

*Mediterranean deep-water algal communities of the genus *Cystoseira* are highly endangered, and in some places have become totally extinct. Furthermore, their distribution is largely unknown, even in the most studied sites, such as MPAs, where their presence has been described only recently. These populations of *Cystoseira* create a complex spatial structure that allows the coexistence of many associated species, thereby resulting in highly diverse communities. Deep-water algal communities show low dynamics and can therefore easily be affected by disturbances even inside MPAs. Consequently, the status of these communities can be an excellent indicator of anthropogenic disturbances such as trampling (by fishing nets, anchoring or diving), or competition by introduced invasive species. Moreover, measuring the impact of disturbances on the population structure and dynamics of these habitat-forming species allows impact assessments in the whole community.*

*Here we present the results of several years of research on the distribution, composition and dynamics of deep-water algal communities along latitudinal gradients and under different perturbation regimes. Their response to different anthropogenic impacts and the conservation strategies to diminish the effect of these disturbances is discussed.*

**Key-words:** *Cystoseira zosteroides*, Population dynamics, Perturbations, Resilience

### **Introduction**

Deep water algal assemblages constitute highly structured and diverse communities, characterized by low dynamics and low resistance to perturbations (Feldmann, 1937; Ballesteros 1992; 2006). Despite their widespread distribution and their floristic, ecological and biogeographical interest, deep-algal communities are poorly known (Ballesteros *et al.*, 2009). Human perturbations such as overfishing, habitat destruction, invasive species and global warming undermine the resilience of benthic communities. Although studies have been centered in describing the impacts and consequences of global change on benthic communities, few have focused on algal communities, and specially studying their resistance to perturbations and their recover capability.

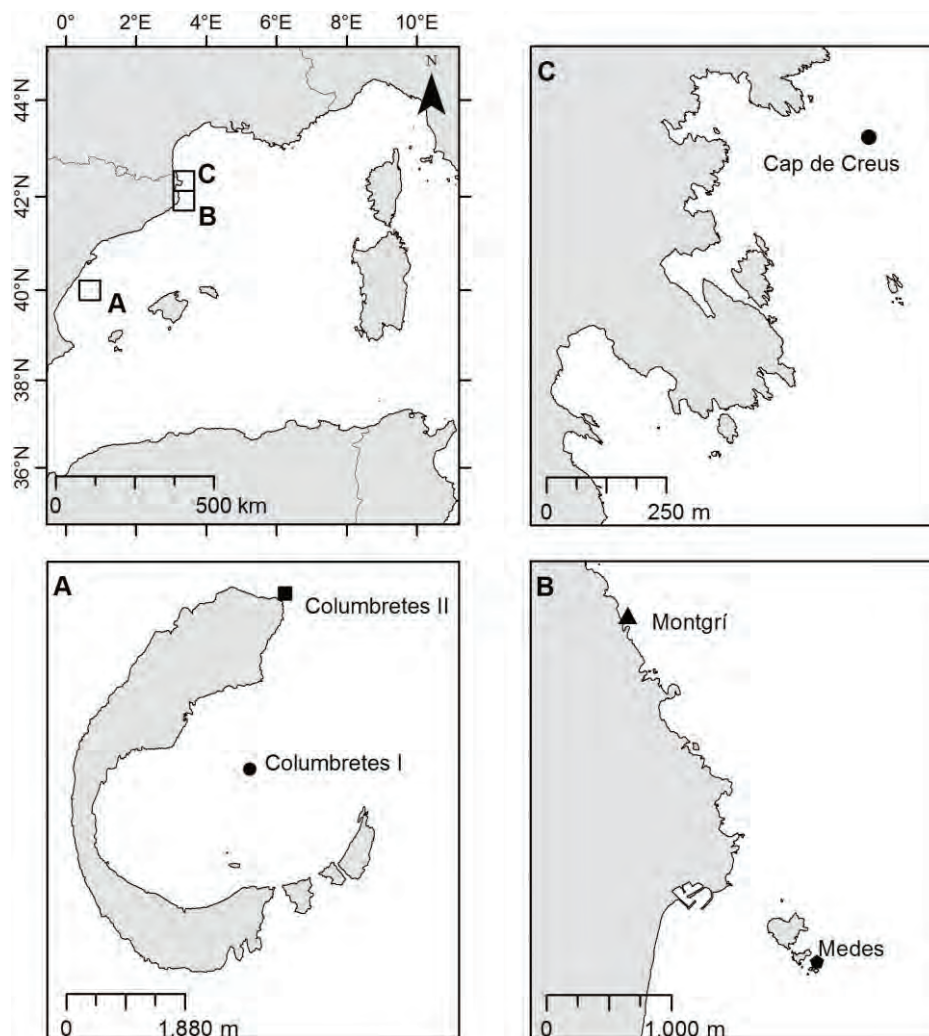
In the Mediterranean coralligenous assemblages are highly diverse and exhibit great structural complexity. Structural animal species such as gorgonian and corals have received special attention during last decades due to the occurrence of mass mortality events and other disturbances associated to global change. Nevertheless, the ecology and the effects of global change on erect algae communities, such as *Cystoseira* spp. communities, are poorly known (Ballesteros *et al.*, 2009).

*C. zosteroides* is vulnerable against anthropogenic disturbances. Evidences show that *C. zosteroides* assemblages are in decline in some Mediterranean areas, where their abundance and distribution has declined in the last years (Thibaut *et al.*, 2005; Serio *et al.*, 2006).

Some studies describe small-scale patterns such as composition and population structure, revealing a high spatial variability in the composition and structure of deep-water *Cystoseira* communities (Hereu *et al.*, 2008). However, little is known about their dynamics although Ballesteros *et al.* (2009) estimated very low growth rates ( $0.5 \text{ cm year}^{-1}$ ) and high longevity (more than 50 years) for *C. zosteroides*. Thus, populations and assemblages dominated by *C. zosteroides* can potentially be highly determined by large-scale, low-frequency catastrophic episodic events, as suggested by Ballesteros *et al.* (2009). These unusual events would lead to punctual high mortality rates of old plants, clearing the canopy and, in turn, allowing intense recruitment. Similar dynamics have been hypothesized to occur in other algal assemblages (Schiel and Foster, 2006), although direct evidences are lacking. In this work, several *Cystoseira zosteroides* populations have been studied following a latitudinal gradient in the western Mediterranean, with the aim to describe their demographic characteristics, their population dynamics and the effect of different perturbations on their viability.

### Material and methods

In order to estimate recruitment, mortality and growth of individuals over time, permanent transects were installed at three locations in the Catalan coast in summer 2008 in the



**Fig. 1: Map of the study sites in the NW Mediterranean. A. Columbretes Islands Marine Reserve (Castelló, Spain). B. Medes Islands Marine Reserve and Montgrí coast (Catalunya, Spain). C. Cap de Creus Marine Reserve (Catalunya, Spain).**

following protected areas: Medes Islands, Montgrí and Cap de Creus, and two locations in 2010 in Columbretes Islands Marine Reserve (Fig. 1). At each site three transects within the same population were installed with permanent marks. Each transect was 1 m wide and 5 m long, and was partitioned in a grid of 50 cm x 50 cm quadrats where each individual was mapped and length of the main axis measured. The axis is a measure of the perennial part of the plant, thus an excellent indicator of size of each plant (see Ballesteros *et al.*, 2009; Navarro *et al.*, 2011).

Sampling was performed in August when *C. zosteroides* attains its highest seasonal biomass (Ballesteros, 1990). Transects were re-visited yearly during three and four consecutive years in order to measure density and size structure, and obtain the demographic values (growth, mortality and recruitment) of the studied populations comparing measures of each plant and quadrat from successive years. Recruits were not considered in the growth comparison between populations as recruitment occurs in spring, and censuses were performed in summer and the growth of recruits did not account for a whole year. Mortality was calculated as the loss of individuals between years and recruitment was estimated as the number of new individuals (i.e. less than 0.5 cm high).

## Results

Size structure of *C. zosteroides* populations showed striking differences. In Columbretes and Montgrí, the population was dominated by medium size individuals, with low mortality rates and low recruitment (Fig. 2), while in Medes and Cap de Creus, populations were dominated by young individuals. Moreover, two punctual episodes caused changes in the structure of Medes and Montgrí. First, a huge storm in December 2008 caused high erosion in benthic communities due to the water flow and the movement of sediments and rocks. This storm affected dramatically with values of 50% to 80% of adult mortality in Montgrí and Medes respectively. After this storm event, a huge recruitment episode occurred in Medes, with an increase of 15% of the population density due to the arrival of new individuals.

Another perturbation occurred in Montgrí in 2009. An abandoned fishing net caused a high mortality by abrasion and plucking individuals that were trapped in the net. Following this mortality, a new huge recruitment episode occurred at the following season (Fig. 2).

Recruitment showed a clear negative correlation with adult density (Spearman rank correlation  $\rho = -0.52$ ;  $p < 0.001$ ; Fig. 3). At stable and dense populations, such as Columbretes, recruitment was practically zero, while populations where adult densities were reduced by a perturbation, such as the exceptional storm in the Catalan coast or the effects of an abandoned net, recruitment was very high.



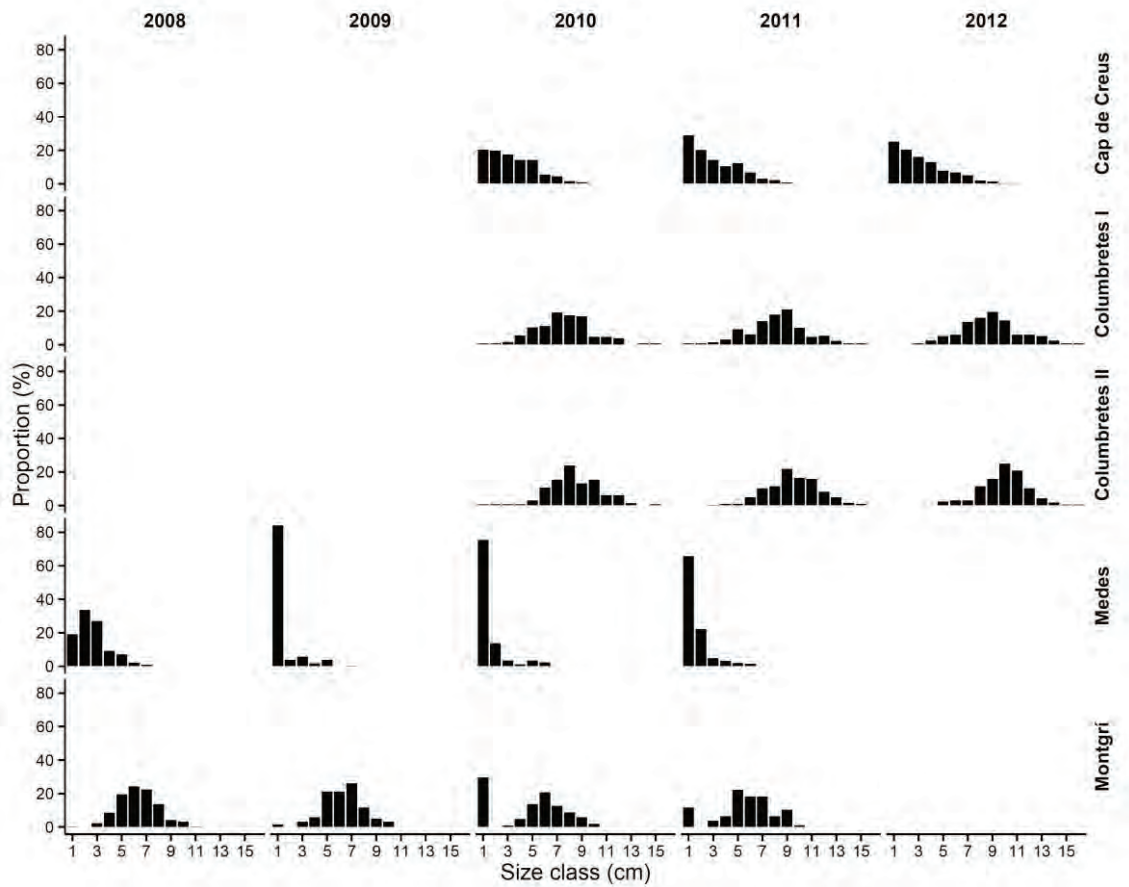


Fig. 2. Size distribution frequencies of the length of the main axis of *Cystoseira zosteroides* populations in the study sites between 2008 and 2012.

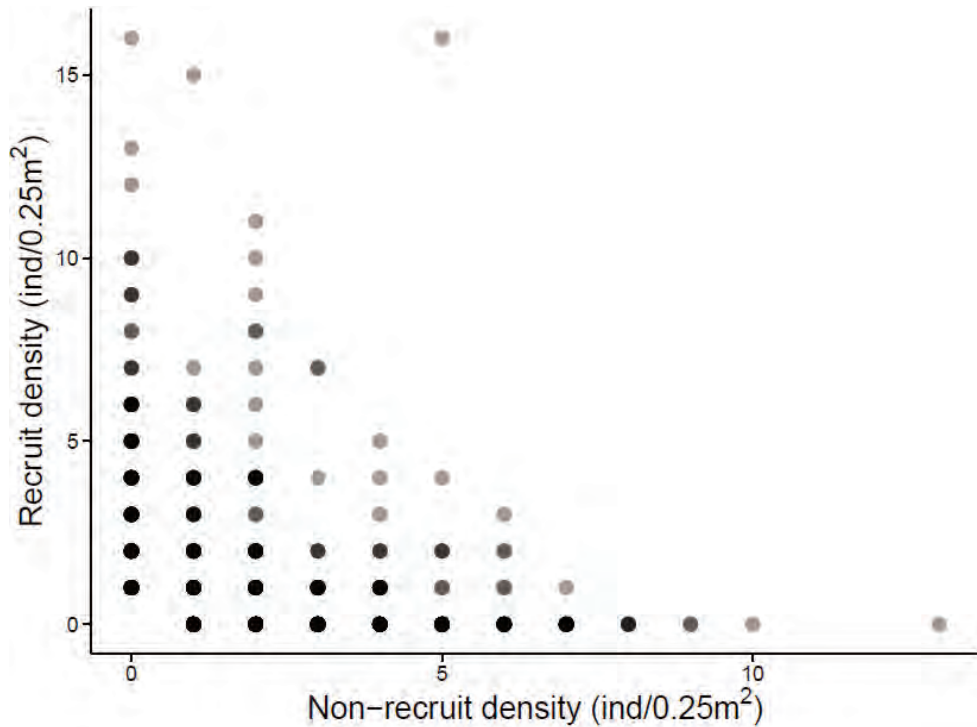


Fig. 3. Correlation between number of recruits and number of non-recruits per quadrat at each sampled site.

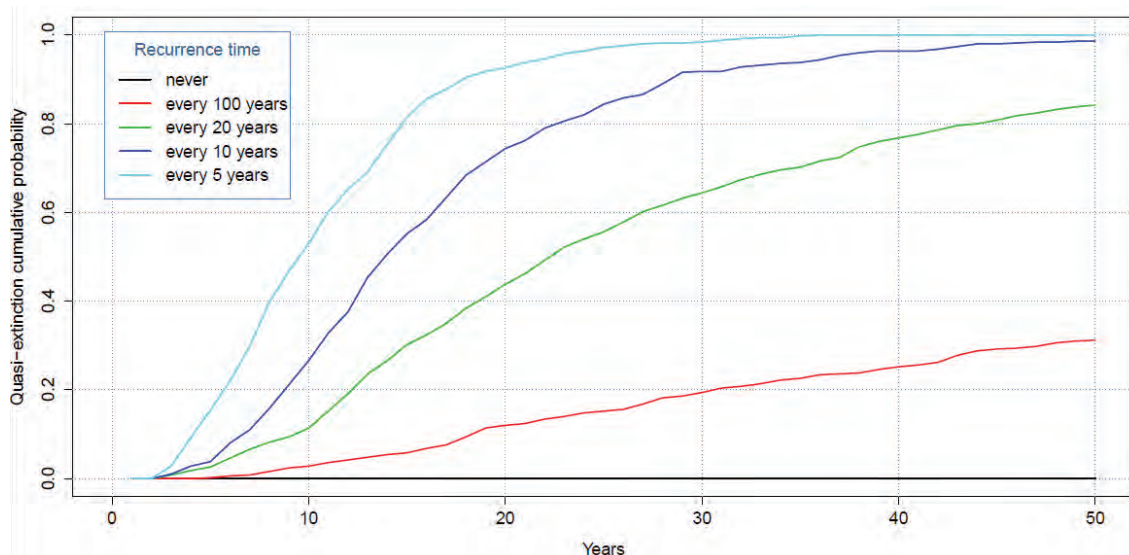
### Discussion and conclusions

This long-term study confirms that *Cystoseira zosteroides* populations are formed by long-live individuals with a very slow growth and low mortality, displaying low recruitment rates in dense populations (Ballesteros *et al.*, 2009; Navarro *et al.*, 2011). Nevertheless, when punctual catastrophic episodes occur and there is a high adult mortality, the recruitment drastically increases. Thus, low frequency episodes may have a great influence in the population structure of *C. zosteroides* stands. Fishing nets seem to be a major pressure causing the progressive disappearance of such stands along the Mediterranean as suggested by Thibaut *et al.* (2005).

Another important issue is the mechanism of recruitment in the dynamics of such communities. While populations are well conserved, recruitment plays a little role in the dynamics and structure of population. Nevertheless, when a high adult mortality occurs, recruitment is important, showing a clear density-dependence, and probably being a mechanism of resilience for such populations. This finding agrees with the hypothesis that deep-water *Cystoseira* forests should be driven by episodic disturbances, after-disturbance recruitment pulses, and long periods of steady growth (>10 years) (Ballesteros *et al.*, 2009).

In a whole, the combination of short-term acting factors that ensure long periods of steady growth, with episodic disturbances that can determine populations at long temporal scale, together with the variability associated to these factors, should be responsible for the complex, almost unpredictable, distribution and size structure of deep-water *Cystoseira* stands.

Next steps may enlarge spatial scales and model populations with the aims not only to understand the relative importance of each demographic parameter, but also to predict future scenarios of viability of these populations under different perturbations. Preliminary results (Fig. 4) showed that *C. zosteroides* is an excellent species to model in order to obtain highly useful information to establish adequate management plans for Mediterranean benthic communities conservation.



**Fig. 4.** Output of a preliminary demographic model, projecting the probability of quasi-extinction of *Cystoseira zosteroides* populations under different scenarios of recurrent perturbations.

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## **ASSESSING *POSIDONIA OCEANICA* BEDS REGRESSIONS USING ANTHROPOGENIC PRESSURES MAPS ALONG A FRENCH COASTAL REGION**

### **Abstract**

*During the last half century, the development of coastal anthropogenic activities is at the origin of increasing pressures on marine coastal ecosystems. The management of those multiple and simultaneous threats moreover requires reliable and precise data on the distribution of the pressures and of the most sensitive ecosystems.*

*Posidonia oceanica beds (Tracheophyta) are encountered between the sea surface and a depth of 30-40 meters. Despite their important ecological and economical roles, they are threatened by many human activities. Regressions can be mapped on the basis of the visible areas of dead mattes and historical data. We used Random Forest algorithm to make predictions of *P. oceanica* beds regression data according to anthropogenic pressures along a Mediterranean French region (Provence-Alpes-Côte-d'Azur). Pressures tested appealed to reclamations from the sea, coastal population, aquaculture, wastewater effluents, anchoring areas, coastal land cover, and stream effluents data as a function of bathymetry and currents. Model performances were particularly good for extreme ecological states (classification error rates of 5% and 27% respectively for regression rate < 10 % and > 90 %). Stream effluents (approximated by siltation) was the major parameter forcing the distribution of *P. oceanica* dead mattes, while sketches of ecological thresholds regarding aquaculture, anchoring and coastal infrastructures are proposed.*

**Key-words:** human impacts; seagrass distribution; resilience; CART (classification and regression trees)

### **Introduction**

Coastal anthropogenic activities have presented worldwide, and particularly in developed areas, a strong increase in the last half century, increasing in the same fashion the pressures on marine coastal ecosystems (Millennium Ecosystem Assessment MEA 2005). The management of those multiple and simultaneous threats moreover requires reliable and precise data on the distribution of the pressures and of the most sensitive ecosystems (Halpern *et al.* 2008; Micheli *et al.* 2013). In Mediterranean Sea, the seagrass *Posidonia oceanica* constitutes a particularly sensitive ecosystem (Boudouresque *et al.* 2012). It forms dense meadows of leaf shoots, rhizomes and roots. Rhizomes, roots and the trapped sediments are called “matte”. This ecosystem, encountered between 0 and 40 meters depth provides numerous ecosystem services (Borum *et al.* 2004). *P. oceanica* beds are however threatened by many human activities such as coastal constructions, trawling, anchoring, or wastewater releases, involving modifications in the local environmental conditions as for example the sedimentation regime. As a result, numerous destroyed meadows i.e. dead mattes are encountered today along the Mediterranean coasts (Boudouresque *et al.* 2009).

In this study we apply the Random Forest algorithm, an advance in the classification and regression trees approach, to better understand the relationship between human coastal

activities, modeled according to the distance to the source of the pressure driver, and the ecological status (alive / dead matte) of the *P. oceanica* meadows.

### Materials and Methods

The studied area ranges between Ramatuelle Cape and the boundary France-Italy (around 150 km). The last maps concerning the marine habitats covering the seabed are available on DONIA® Expert ([www.Medtrix.fr](http://www.Medtrix.fr)). They provide the spatial distribution of ten different habitats including alive *Posidonia oceanica* meadows and dead matte. Between 0 and -15 m, these maps are completed with historical data (aerial pictures of 1920's) in order to identify potential dead matte that could be invisible (covered by sand for example) on present maps (see Deter *et al.* 2013 for the methodology). This permits to consider the temporal dimension of the meadows at their upper limits: alive = stable / dead matte or past meadow invisible on present maps = regression. The bathymetric data (horizontal resolution of 25 m) come from the French hydrographic office "SHOM" completed by our own data acquired with a multi-beam GeoSwath sounder.

The main coastal human activities are listed and mapped according to six main categories: reclamations from the sea (Medam 2014), the coastal population (INSEE data), the aquaculture, the wastewater effluents (localized deeper than -15 m) and the anchoring areas weighted as a function of bathymetry and currents (Anthropo-map data available on [www.medtrix.fr](http://www.medtrix.fr)), and the land cover observed (urbanization and agriculture in a radius of respectively 10 and 5 km) along the coast (Corine Land Cover dataset). Those different activities are then transformed into a human pressure index, ranging between 0 (no pressure), and 20 (intense human pressure on the site). The index value is interpolated using a function of decreasing weight with the distance from the source of the pressure. One environmental parameter is used as a proxy of the stream effluents pressure on the coastal ecosystems: siltation as a sludge percent cover (ranging between 0 and 100 %). It was sampled on the basis of photographic quadrats taken on 32 coralligenous reefs localized within the study area (data available within the project RECOR on [www.medtrix.fr](http://www.medtrix.fr), see Deter *et al.* (2012) and Andromède océanologie (2013) for the methodology).

Data are mapped on ArcGIS using the French Lambert 93 projection. The different layers are rasterized using three different pixel resolutions: 200 m x 200 m, 100 m x 100 m, and 20 m x 20 m, to query the spatial scale of *P. oceanica*'s ecological response. The ecological status of *P. oceanica* is calculated, for the 200 m and 100 m resolution datasets, as a rate (%) of surface regression per pixel, using the following formula: regression rate (=R) = regression surface / pixel total surface area x 100. This regression rate is then subdivided into four categories for classification: pristine for  $R < 10$ , acceptable for  $10 \leq R < 50$ , threatened for  $50 \leq R < 90$  and critical for  $R \geq 90$  %. This *P. oceanica* regression variable is however kept binary for the 20 m resolution layer, because of computational limitations. The data on *P. oceanica*'s ecological status, bathymetry, siltation, and human induced pressure are finally sampled from the raster layers.

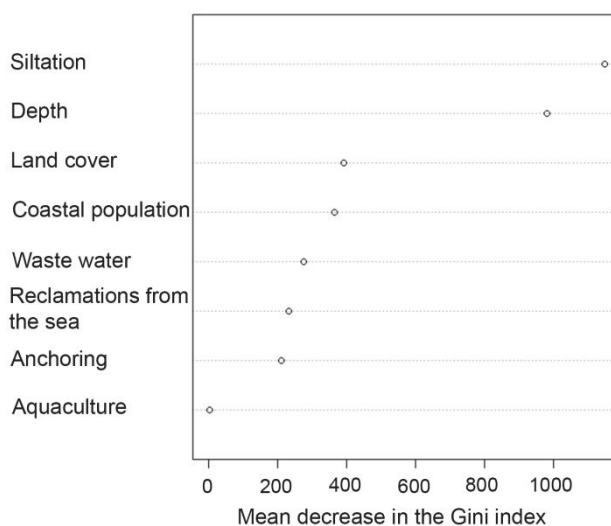
For the modeling approach, we use the software R and the Random Forest algorithm (RF) in order to predict the *P. oceanica* ecological status with the different descriptor variables. Just as traditional classification methods, RF uses a set of descriptor variables to predict one variable of interest according to classification trees. RF however combines many trees together in a "forest", thereby improving the efficiency of the model (Breiman, 2001). The Random Forest algorithm can also be used in statistical regression mode. Its functions according to the following steps: bootstrapping (classification: the Gini Index is



computed for each splitting point on each variable and the best split is kept for the node), error estimation (averaging the number of misclassifications), estimation of the importance of the variables, prediction. RF is built using 1000 trees, and testing randomly 3 potential splitting variables at each node for the 200 m and 100 m resolution datasets. Because of computational limitations, RF is built using 250 trees and testing 3 potential splitting variables at each node for the 20 m resolution dataset. We use the Gini index in order to measure the inequality among the trees. For classification, the final class will be the class having the most votes, whereas for statistical regression, the final value will be an average over all the trees. We test the prediction capacity of the model (the algorithm runs the dataset down each tree in the forest) by building a prediction forest on 80% of the dataset. A prediction test is then performed on the 20% remaining of the cases, in order to analyze the correspondence between observed and predicted *P. oceanica* regression values.

## Results

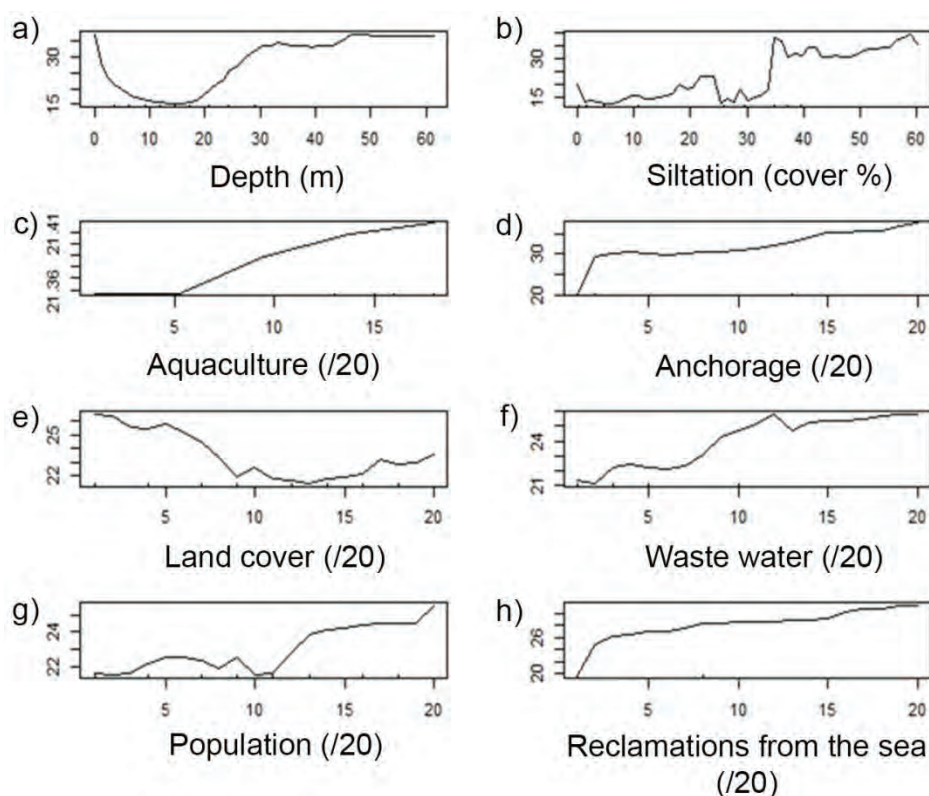
The Random Forest algorithm appears particularly accurate for the classification of pristine areas (error rate = 5 %) and areas in critical ecological state (error rate = 27 %). Model performances compared by resolution show that the 100 m resolution dataset performs the best: Pearson correlation between observed and predicted *P. oceanica* regression values = 0.81, Error rate = 21.93. The siltation appears to be the most important descriptor followed by the bathymetry (Fig 1). The pressure from the aquaculture appears on the other hand not to be of great importance in the classification but this pressure is quiet weak in the study area in comparison with other Mediterranean areas (Pergent-Martini *et al.* 2006).



**Fig. 1: Importance of each variable (siltation, depth, land cover, coastal population, waste water effluents, reclamations from the sea, anchoring and aquaculture) in the classification model, in terms of mean decrease in the Gini index. The higher is the value of the Gini index, the greater is the importance of the variable for the regression rate of *P. oceanica* meadows.**

The prediction test shows a good correlation ( $r=0.81$ ) between observed and predicted data especially for extreme ecological conditions. The partial dependence plots between the predicted regression rates of *P. oceanica* beds and each descriptor used in the study implied interesting ecological considerations (Fig. 2). The ecological depth optimum for

*P. oceanica* is found to be between 5 and approximately 20 m, around 15 m deep (Fig. 2a). The partial dependence plot with respect to the siltation percentage cover shows a progressive doubling of the regression rate between 0% and 60% sludge cover (siltation), with a not explicated low regression frame around 30% of sludge cover (Fig. 2b). The response to aquaculture is not significant with a mean predicted regression varying of only 0.1%, but the slope of this response is positive with respect to the pressure intensity, with a breaking point at a pressure value of 5/20 (Fig. 2c). *P. oceanica* regression seems very sensitive to anchoring with a steep increase in regression from the lowest pressure values, and a doubling of the mean predicted regression rate between anchoring pressure values of 0 and 20 (Fig. 2d). The dependence on the land cover pressure shows a decrease in regression up to a pressure of approximately 13/20, followed by a final increase of regression (Fig. 2e). This unexpected behavior should be further investigated by analyzing the raw human activity data transformation into a pressure index. The effect of wastewater and population is described by a regular increase of the regression rate from approximately 21 to 26%. Low regression rates are also observed for a population pressure value around 10 (Fig. 2f, 2g). The regression increase is moreover important for low infrastructure pressure, with an overall increase of 10% between 0 and full infrastructure pressure (Fig. 2h).



**Fig. 2:** Partial dependence plots between the mean predicted *P. oceanica* regression rate (%) and a) depth (m), b) siltation (cover %), c) aquaculture (note /20), d) anchorage (note /20), e) land cover (note /20), f) wastewater effluents (note /20), g) coastal population (note /20) and h) reclamations from the sea (note /20)

### Discussion

The analysis of the response of *P. oceanica* to anthropogenic coastal pressures highlights first of all the evidence that the spatial resolution of *P. oceanica* ecological response to

human stress was of the order of 100 meters or lower, as suggested already for marine coastal ecosystems in general (Parravicini *et al.* 2012). This observation moreover shows the potential of amelioration of the model when considering a lower resolution. In this case, the training dataset should however be reduced to a smaller area for computational considerations.

Concerning the model explaining *P. oceanica* regression rates between 0 and -15m, the two most important descriptors are finally two environmental ones (siltation and depth) commonly known to influence the growth of *P. oceanica* (Boudouresque *et al.* 2012). The optimal depth observed with the partial dependence plots well corresponds with the values from the literature (Boudouresque *et al.* 2012). The effect of water turbidity will be more precisely tested with a larger data set based on Previmer data ([www.previmer.fr](http://www.previmer.fr)). The two next more important drivers are land cover (urbanization and agriculture) and coastal population. These are certainly the primary factors explaining all the following drivers tested as waste water effluents and reclamations from the sea are dependent on the population and urbanization. Indeed, urbanization involves the presence of consumers, so an increased need in resources (water, energy, raw material) and in natural areas for recreational activities, but also the emission of varied discards in waters, soils and air. Here wastewater effluent show a relatively weak effect on meadows certainly because we only consider the upper limits (0 to -15 m) while the effluents are mostly localized deeper.

The partial dependence also shows interesting results: the breaking points highlighted must be analyzed in comparison with the raw human activity dataset, in order to answer crucial issues like for example to which minimal distance waste water effluent or an aquatic farm must be built from a *P. oceanica* meadow in order not to impact it.

## Conclusion

This study develops a model between the main human coastal pressures on the French Mediterranean coast and the ecological response of the most important ecosystem in the region. The classification algorithm used shows good performances. The application of this model will be broaden to a bigger area and with more descriptors. The only data necessary to run the prediction for ecological status once the algorithm is well trained are in fact the cartography of the ecosystems and of the different pressures. The minimum surface needed to train efficiently the algorithm should moreover also be investigated.

## Acknowledgments

This study benefited of a financing from the French Water Agency (Agence de l'eau Rhône-Méditerranée-Corse). Florian Holon received a PhD grant (2013-2016) funded by LabEx CeMEB and Andromède Océanologie.

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## **FIRST CONTINUOUS SEABED MAP IN FRANCE USED FOR THE CREATION OF A MANAGEMENT TOOL PROTECTING *POSIDONIA OCEANICA***

### **Abstract**

*The Mediterranean Sea is a biodiversity hotspot facing numerous threats: high population density, high urbanization rate, coastal erosion, overexploitation of marine resources and spread of invasive species. Maps of marine habitats are an essential tool in order to appreciate the ecological and spatio-temporal heterogeneity of the environment, potential and real distribution of species, identify corridors but also propose relevant management measures and evaluate their effects. Different studies allowed the realization of high scale marine habitat maps. It resulted in a patchwork of more or less connected maps. Through the DONIA program we reviewed and completed 30 years of work in order to obtain a continuous map of the French coastline: 1 700 km and 47 water bodies.*

*We obtained a 1:5 000 continuous map of the seabed habitats using ten classes. The results are freely available (with login) on [www.medtrix.fr](http://www.medtrix.fr) in DONIA®expert. Besides the complete French Mediterranean coast, several Mediterranean islands are concerned: the Galite Archipelago in Tunisia, Zembra Island in Tunisia and Tavolara - Punta Coda Cavallo in Sardinia (Italy). A simplified database called DONIA® intended for the general public is also freely available on Medtrix without any login. The same regions are concerned but habitats were simplified into four classes (seagrass, dead matte, sand, rock) instead of ten. This simple dynamic map is freely accessible to all through the DONIA® application (App Store and Google play); it helps boats to safely anchor outside of sensitive habitats. The application opens new perspectives in terms of management of marine protected areas by facilitating the communication between site managers and visitors and providing an active management of mooring.*

**Key-words:** anchoring management; marine habitat; cartography; seagrass conservation

### **Introduction**

The Mediterranean Sea is a biodiversity hotspot facing numerous threats: a very high population density, high urbanization rate, coastal erosion, overexploitation of marine resources and spread of invasive species (PNUE/PAM-Plan Bleu, 2009; Chevaldonné *et al.*, 2010). A key-ecosystem particularly mistreated despite its protected status is the *Posidonia oceanica* seagrass meadow (Boudouresque *et al.*, 2009). Indeed, its ecological preferences (shallow and clear waters, low current) and biological characteristics (very slow growth) makes this species very vulnerable to anthropogenic pressures (Boudouresque *et al.*, 2012). For example, every day, by anchoring, thousands boat users may irreversibly destroy small pieces of meadow because they do not know this protected species and are not aware of its spatial distribution.

Maps of marine habitats are an essential tool in order to appreciate the ecological and spatio-temporal heterogeneity of the environment, potential and real distribution of species, identify corridors but also propose relevant management measures and evaluate their effects (Boström *et al.*, 2011). Several high scale maps of marine habitat are



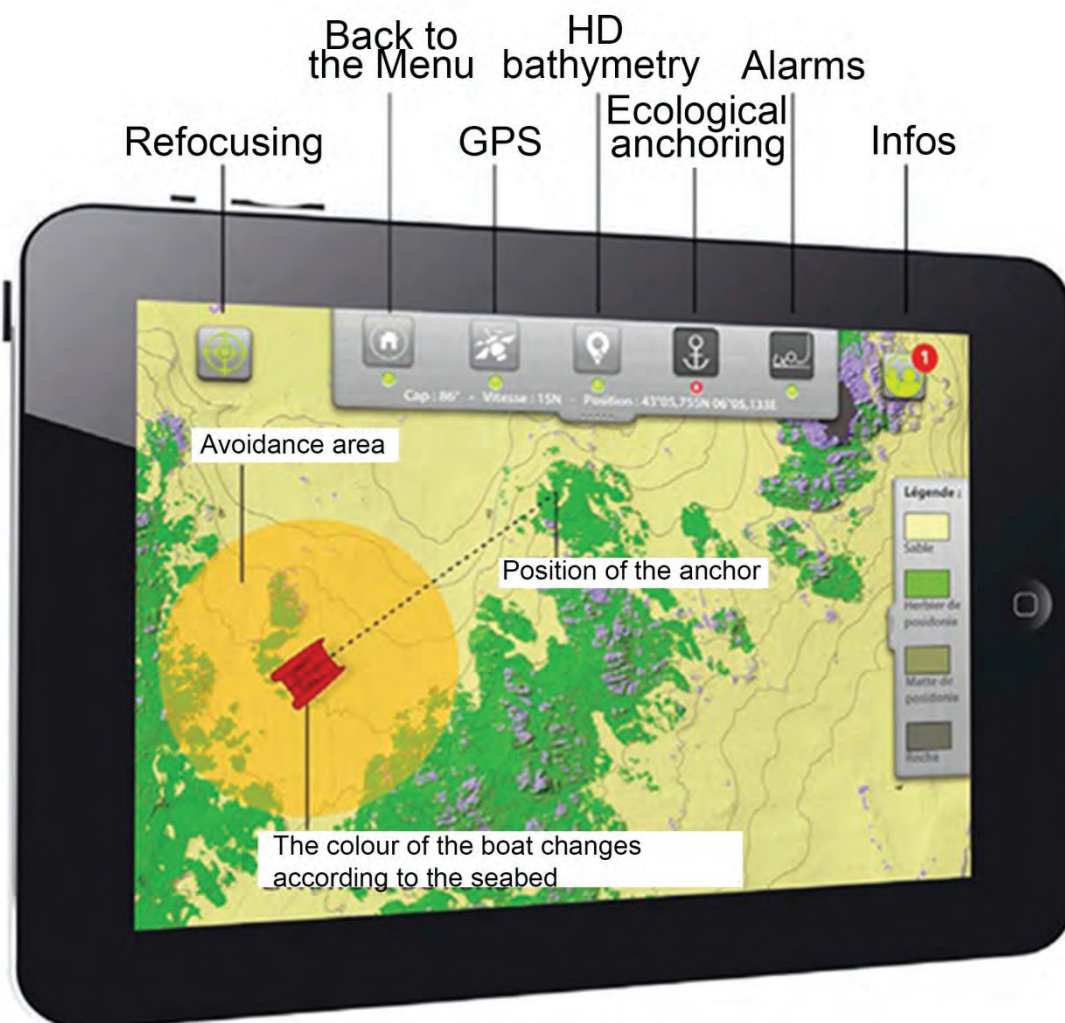
produced each year. The outcome is a patchwork of more or less connected maps. Through the DONIA program, we reviewed and completed 30 years of work in order to obtain a continuous map of the entire French Mediterranean coast and of 47 water bodies. We then used this map for the development of a free mobile application helping boat owners to avoid anchoring within seagrass and thus preserve *Posidonia oceanica* beds.

### Materials and Methods

After a bibliographic synthesis, we pooled and homogenized the habitat maps built in the framework of numerous programs: DONIA (Andromède Océanologie), CARTHAM - AAMP (G.I.S. Posidonie, Sintinelle, Stareso, Evemar, Andromède Océanologie), PIM (Conservatoire du littoral, Agence de l'eau RMC), MEDBENTH (IFREMER), EGIS EAU, Aire marine protégée de la côte agathoise (site Natura 2000), DREAL PACA, Ville de St Cyr-sur-Mer and Ville de Saint Raphaël. Campaigns were led between 2010 and 2014 using aerial photography (in order to see the spatial extent of the habitats in shallow waters), a multi-beam echo-sounder survey (to draw the bathymetry), a side-scan sonar survey (used in more turbid and deeper waters), sonar information post-treatment (in order to determine potential presence and covering of underwater habitat representation), production of a preliminary cartography which is then validated by 'ground truth' diving sessions (field work). The methodology is available at Andromède Océanologie (2014). A final continuous habitat map was realized considering ten habitat classes. This map was secondarily simplified (four classes) in order to be understood by the general public. A Smartphone / Tablet Application was then developed in partnership with the French company 3Liz in order to help yachtsmen and yacht captains to check the nature of the seabed and find a suitable location for anchoring.

### Results

We produced a 1:5000 continuous map for seabed habitats using ten classes: *Cymodocea nodosa* seagrass, *Posidonia oceanica* seagrass, dead matte association, infralittoral shingle association, infralittoral soft bottoms, infralittoral algae, coralligenous habitat, offshore rocks, circalittoral soft bottoms, artificial habitats. The results are freely available (with login) on [www.medtrix.fr](http://www.medtrix.fr) in DONIA®expert. Besides the complete French Mediterranean coast (Languedoc-Roussillon, Provence and French riviera, Corsica being completed), several Mediterranean islands were concerned: the Galite Archipelago in Tunisia, Zembra Island in Tunisia and Tavolara - Punta Coda Cavallo in Sardinia (Italy). A simplified database (four classes = seagrass, dead matte, sand, rock) called DONIA® intended for the general public is also available on the so-called [www.donia.fr](http://www.donia.fr) website and on [www.medtrix.fr](http://www.medtrix.fr) without any necessary login. This simplified dynamic map is freely accessible to all through the DONIA® application (App Store and Google play), helping boats to anchor safely outside of sensitive habitats. Once the chosen maps are downloaded on a smartphone or a tablet, DONIA® is able to position the boat and show the nature of the underwater seabed. The boat appears colored in green or red depending if anchoring should be preferred or avoided (Fig. 1). The application also provides a prepaid panel of anti-skid and anti-collision anchoring alarms which increase safety onboard, even detecting the crossing of chains and preventing all risk of entanglement. DONIA® also offers users the ability to acquire high precision bathymetry 3D maps to accurately visualize the underwater topography of several sites and prepare for scuba dives for example. Officially launched at the 2013 Paris Boat Show, DONIA® had more than 1 000 users (and 8 113 views) in July 2014.



**Fig. 1: Main window of the DONIA® application (currently only available in French). The legend displays color correspondence for the habitats: yellow = sand, green = *Posidonia oceanica* meadow, brown = dead mat and violet = rocky habitat.**

### Discussion

We present here a very important step forward in marine ecology: the first continuous map of marine habitats along the French Mediterranean coasts. A fine knowledge of the presence, distribution and conservation status of the habitats of each area, and in particular of *Posidonia oceanica* meadows, is the first essential step for an appropriate and effective long-term managing of marine resources. Moreover, these data are now available to the general public and more importantly to sea users. Free and easy to use, DONIA® is a resounding success. It received two awards in 2013: “Companies and biodiversity” from the French Ministry of Environment and “Bateau Bleu” from the French nautical industries.

A management tool named “Donia Manager Module” is under progress. The objective is for managers of marine protected areas to control and limit the impacts of tourism in their area, especially of leisure boating. This interactive interface will allow a real-time monitoring of the boat flow through the sending and receiving of information between

managers and users, such as on anchoring, possible marine pollution, or on the presence of protected species.

### Conclusion

The application opens new perspectives in terms of management of marine protected areas by facilitating the communication between site managers and visitors and providing an active management of mooring. It will also raise awareness and call for sustainable practices associated with open anchoring in sites of community interest.

The objectives of this tool are to decrease the impact of anchoring in marine protected areas by 50 %. This innovative tool is expected to widen its geographical scope and to be exported, first in Mediterranean countries such as Italy and Spain, then in all parts of the world where the impact of recreational boating on the marine environment is a major challenge for the conservation of marine biodiversity.

### Acknowledgments

This study received a financing from the French Water Agency (Agence de l'Eau Rhône-Méditerranée-Corse). Maps available with DONIA® were created from data collected by Andromède océanologie; Agence de l'Eau RMC; Conservatoire du Littoral; DREAL PACA; EGIS EAU; ERAMM; GIS POSIDONIE; IFREMER; Institut océanographique Paul Ricard; Nice Côte d'Azur; TPM; Programme CARTHAM - Agence des Aires Marines Protégées & Andromède océanologie, ASCONIT Consultants, COMEX-SA, EVEMAR, GIS Posidonie, IN VIVO, Sintinelle, Stareso; Programme MEDBENTH; Université de Corse (EQEL); Ville de St Cyr sur mer; Ville de Cannes; Ville de Marseille; Ville de St Raphaël; Ville de St Tropez. . We thank all the people who participated in field work and/or map making in the framework of these programs. Florian Holon received a PhD grant (2013-2016) funded by LabEx CeMEB (Laboratoire d'excellence "Centre méditerranéen de l'environnement et de la biodiversité") and Andromède Océanologie. We thank Sibylle Cazacu for improving the English version of the manuscript.

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## **DISTRIBUTION AND COMPOSITION OF *CYSTOSEIRA* STANDS ALONG THE WEST ISTRIAN COAST (NORTHERN ADRIATIC, CROATIA) AND COMPARISON WITH HISTORICAL DATA**

### **Abstract**

*The northern Adriatic represents the northernmost biogeographic sector of the Mediterranean. Additionally, due to north Italian rivers runoff, it represents one of the most eutrophicated regions of the Mediterranean. Differently to other north Adriatic areas, the sea bottom along the west Istrian coast is mainly rocky, i.e. particularly adapted to harbor Cystoseira (Phaeophyceae, Fucales)-dominated assemblages. The west Istrian coast spans approximately 100 km straight from north to south. This particular orientation produces large scale gradients of oceanographic variables and wave action intensities. A series of Cystoseira demography surveys were conducted during the last 10 years at different depths. From 0 to 1 m depth Cystoseira stands were mainly composed of Cystoseira amentacea, which attained maximum abundances along southern sectors of the coast. Around 1 m depth stands were dominated by C. crinita or C. barbata. The composition of these stands likely depends on local gradients of wave action and anthropogenic pressure. From 4 m depth to the rocky bottom edge (usually between 10 and 20 m) mixed stands were dominated by C. foeniculacea or C. corniculata. In some sites C. spinosa was a major component of these mixed stands. C. compressa and C. humilis were found along the whole coast at all depths as components of mixed stands. Monospecific stands of C. compressa were noticed only in moderately polluted sites. Comparison with historical data revealed long term changes of Cystoseira spp. abundance. A phase of regression, occurring during the last decades of the past century, was followed by a phase of recovery. At present, the composition of stands in sites not directly subjected to urban pollution might be considered similar to that assessed during the 1950s.*

**Key-words:** *Cystoseira*, mixed stands, long-term changes, eutrophication, rocky bottoms, northern Adriatic

### **Introduction**

Almost 30 species of the genus *Cystoseira* C. Agardh are present in the Mediterranean (Ribera *et al.*, 1992). Different *Cystoseira* species can be found from the infralittoral to the upper circalittoral zone (Giaccone & Bruni, 1973). Usually they are large and long-lasting canopy forming macroalgae. The complex structure of their thalli forms particular habitats suitable for the settlements of other algal and animal species. Thus, they play an essential role in sustaining biodiversity levels of Mediterranean coastal ecosystems (Ballesteros *et al.*, 1998; 2009). Anthropogenic pressure (eutrophication, urban and industrial pollution) and sea urchin grazing can negatively affect *Cystoseira* species (Thibaut *et al.*, 2005). According to these issues, *Cystoseira* spp. have been proposed as biological indicators of coastal waters' ecological quality status for the implementation of the European Water Framework Directive (Ballesteros *et al.*, 2007).

The aim of this study is to summarise results of several surveys of *Cystoseira* species distribution conducted along the west Istrian coast in the period 2003-2013 and to compare these results with historical data. Along the west Istrian coast *Cystoseira* spp.



were studied since the end of the XIX century (Munda, 2000). Historical most detailed surveys are those for 1950 (Ercegović, 1952) and for the period 1967-1970 (Munda, 1979). The west Istrian coast is located in the northern Adriatic, which represents the northernmost biogeographic sector of the Mediterranean (Bianchi & Morri, 2000). Because of its specificity in oceanographic conditions (Degobbi *et al.*, 2000), it can be expected that patterns of *Cystoseira* stands' composition differ from those in other Mediterranean regions.

### Materials and methods

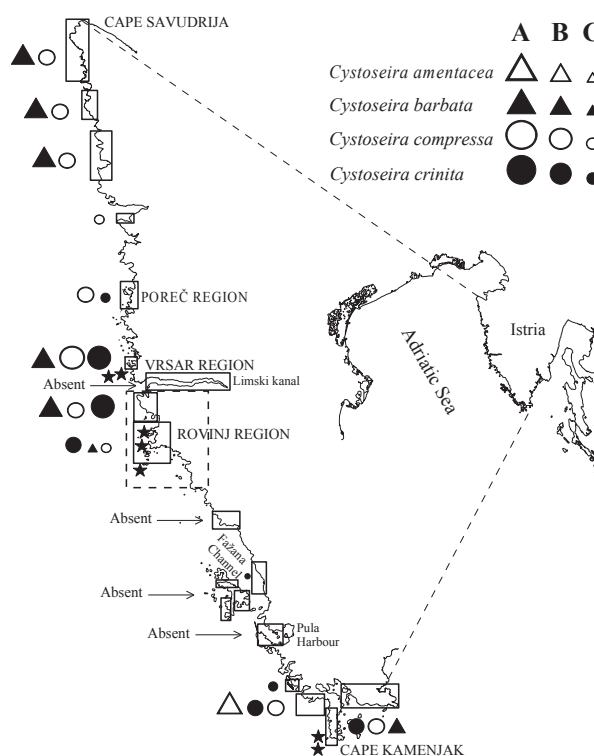
*Cystoseira* species distribution and abundance were estimated during four surveys including visual observation and collection of samples in randomly chosen quadrats (SCUBA diving).

(1) In 2003-2004 the abundance of *Cystoseira* spp. was assessed in 10 randomly chosen locations in the region of Rovinj from 0 to 5 m depth. *Cystoseira* spp. abundance was assessed in the laboratory as biomass (wet weight).

(2) In spring 2009, 12 locations scattered from Cape Savudrija to the entrance of the Channel of Fažana were sampled from 4 to 12 m depth. *Cystoseira* abundance was assessed in the laboratory as biomass (wet weight).

(3) In 2011-2013, *Cystoseira* spp. distribution was estimated in sectors of the west Istrian coast ranging from several km to ten or more km according to the CARLIT methodology (Ballesteros *et al.*, 2007).

(4) Locations sampled in 1950 (Ercegović, 1952) (Fig. 1) and in the period 1967-1970 (Munda, 1979) (region of Rovinj, Fig. 1) were visually inspected by SCUBA diving in spring 2003 and 2009.



**Fig. 1:** Abundance of *Cystoseira* spp. assessed with the CARLIT method. Rectangles show inspected sectors. A = continuous stands, B = larger patches and C = sparse patches. Locations sampled by Ercegović are indicated with ★. The area sampled by Munda (region of Rovinj) is marked with dashed rectangle.



## Results

*Cystoseira* taxa recorded historically and in the period 2003-2013 are shown in Table 1. In total, 12 *Cystoseira* taxa (9 species and 3 forms) have been recorded for the west Istrian coast. All these taxa were found in samples collected during the period 2003-2013 but *C. dubia*. Recent composition patterns of *Cystoseira* stands in the study area are presented below.

**Tab. 1: *Cystoseira* taxa recorded along the west Istrian coast. 1950: Taxa recorded in the sector between the Vrsar region and Cape Kamenjak (Fig. 1) (Ercegović, 1952). 1967-1970: Taxa recorded in the region of Rovinj (Munda, 1979; 2000). 2003-2013: Taxa recorded along the west Istrian coast during this study.**

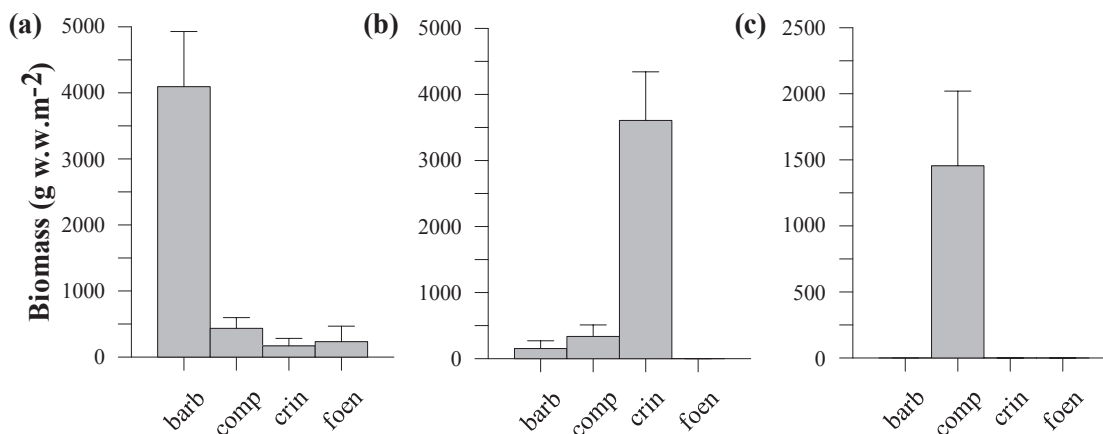
<i>Cystoseira</i> taxa	1950	1967-1970	2003-2013
(1) <i>C. amentacea</i> var. <i>spicata</i> (Ercegović) G. Giaccone	+	+	+
(2) <i>C. barbata</i> (Stackhouse) C. Agardh	+	+	+
(3) <i>C. compressa</i> (Esper) Gerloff & Nizamuddin	+	+	+
(4) <i>C. compressa</i> f. <i>plana</i> (Ercegović) Cormaci, G. Furnari, Giaccone, Scammanca & D. Serio	-	+	+
(5) <i>C. compressa</i> f. <i>rosetta</i> (Ercegović) M. Cormaci, G. Furnari, G. Giaccone, B. Scammacca & D. Serio	+	+	+
(6) <i>C. corniculata</i> (Turner) Zanardini	+	+	+
(7) <i>C. crinita</i> Duby	+	+	+
(8) <i>C. dubia</i> Valiante	-	+	-
(9) <i>C. foeniculacea</i> (Linnaeus) Greville	+	+	+
(10) <i>C. foeniculacea</i> f. <i>latiramosa</i> (Ercegović) A. Gómez Garreta, M.C. Barceló, M.A. Ribera & J. Rull Lluç	-	+	+
(11) <i>C. humilis</i> Schousboe ex Kützing	+	+	+
(12) <i>C. spinosa</i> Sauvageau	+	+	+

### Rovinj region (sampled 2003-2004 from 0 to 5 m depth)

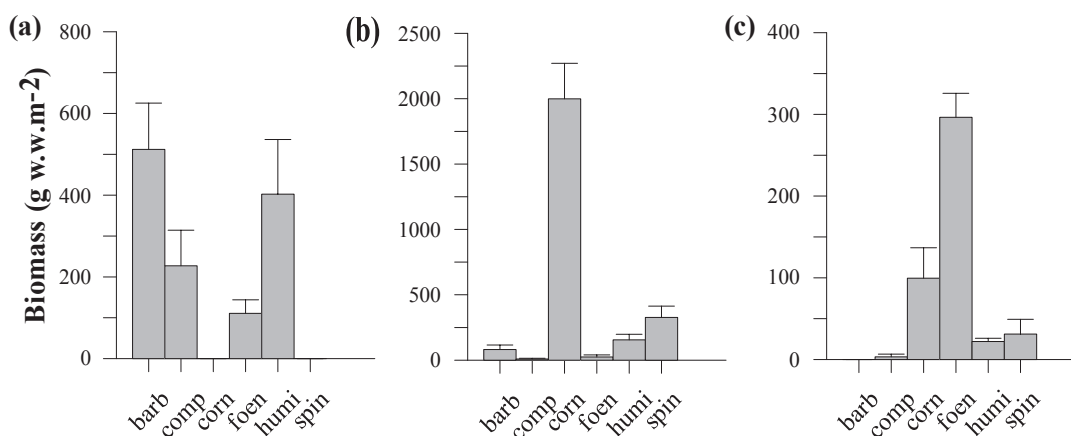
*Cystoseira* spp. formed mixed stands that could be subdivided in three types. (a) *C. barbata* stands were found in unpolluted locations not exposed to wave action. Low amounts of *C. compressa*, *C. crinita* and *C. foeniculacea* were present in these stands (Fig. 2a). (b) In intermediately exposed location, the dominant species was *C. crinita*. *C. compressa* and *C. barbata* were present in low amounts (Fig. 2b). (c) In locations subjected to urban pollution, only *C. compressa* was present (Fig. 2c).

### Northern and central west Istrian coast (sampled 2009 from 4 to 12 m depth)

*Cystoseira* spp. formed mixed stands that could be subdivided in three types. (a) *C. barbata*-*C. humilis* stands were characteristic for some very sheltered locations of the Poreč-Vrsar region on the gently sloping rocky bottom. Relevant quantities of *C. compressa* and *C. foeniculacea* were also present (Fig. 3a). Sparse small thalli of *C. compressa* f. *plana* and *C. foeniculacea* f. *latiramosa* were found in the samples. (b) *C. corniculata*-dominated stands were found on steep rocky bottoms from exposed and intermediately exposed habitats. Other species composing these stands were *C. barbata*, *C. compressa*, *C. foeniculacea*, *C. humilis* and *C. spinosa* (Fig. 3b). (c) *C. foeniculacea*-dominated stands were found on the gently sloping rocky bottom of exposed and intermediately exposed habitats. *C. compressa*, *C. corniculata*, *C. humilis* and *C. spinosa* were present in these stands (Fig. 3c).



**Fig. 2:** Examples of *Cystoseira* mixed stands found along the west Istrian coast between 0 and 5 m depth. (a) *C. barbata* dominated stand (45° 07' 12" N, 13° 36' 87" E). (b) Stand dominated by *C. crinita* (45° 00' 09" N, 13° 43' 87" E). (c) Stand dominated by *C. compressa* (45° 05' 84" N, 13° 38' 11" E). Abbreviations: barb, comp, crin and foen = *C. barbata*, *C. compressa*, *C. crinita* and *C. foeniculacea*, respectively. Data are means and SE of 15 replicated measurements.



**Fig. 3:** Examples of *Cystoseira* mixed stands found along the west Istrian coast between 4 and 12 m depth. (a) *C. barbata*-*C. humilis* stand (45° 10' 02" N, 13° 35' 49" E). (b) Stand dominated by *C. corniculata* (45° 17' 54" N, 13° 35' 05" E). (c) Stand dominated by *C. foeniculacea* (45° 00' 28" N, 13° 42' 45" E). Abbreviations: corn, humi and spin = *C. corniculata*, *C. humilis* and *C. spinosa*, respectively; other abbreviations as in Fig. 2. Data are means and SE of 18 replicated measurements.

CARLIT survey of the west Istria coast (2011-2013)

*Cystoseira barbata* was very abundant in the northern parts of the coast. Sparse patches of *C. compressa* were present. In the central sector characteristic species were *C. crinita* and *C. barbata*. Sparse thalli of *C. amentacea* were found in islets (e.g. Isle Sv Katerina, 45° 04' 34" N, 13° 37' 39" E; Isle Lunga, 45° 08' 31" N, 13° 34' 53" E). In locations subjected to urban pollution, dense settlements of *C. compressa* were found. The size of thalli was very large (total length up to 1 m). In more exposed locations *C. compressa* f. *rosetta* was mostly present. The southern sector was characterised by continuous belts of *C. amentacea*. Relevant patches of *C. crinita* and *C. compressa* were also found.

Usually, in very exposed locations *Cystoseira* spp. were not found (e.g. Isle Sv Ivan, 45° 02' 44" N, 13° 37' 28" E; Marine protected area Brijuni, 45° 53' 37" N, 13° 44' 48").

The deleterious effect of sea urchins grazing was particularly evident at locations Limski kanal and Pula Harbour.

#### 2003 and 2009 surveys of historically sampled locations

Both 2003 and 2009 visual surveys revealed the presence of all *Cystoseira* taxa in all locations surveyed in 1950 (Fig. 1; Ercegović, 1952) except for location 148 (Isle Sv Katarina) which delimit Rovinj's south harbour. *C. barbata* and *C. spinosa*, reported to dwell at 1 m depth during the 1950 survey, were not found in this location. This fact could be related to an enhanced anthropogenic pressure in the harbour area. Visual surveys of locations sampled in 1967-1970 in the region of Rovinj (Munda, 1979; 2000) gave similar results. *C. barbata* and *C. spinosa* which were present in the area of the port of Rovinj in 1967-1970 were not found. In locations outside the port, all historically recorded taxa were found but *C. dubia*. This species was not recorded in any 2003-2013 survey.

#### **Discussion and conclusions**

The depth range between 0 and 3 m depth comprises the infralittoral fringe and the upper infralittoral zone. The distribution of *Cystoseira* spp. was mainly regulated by the exposition to S and SW winds (i.e. wave action). *C. amentacea* was found in the infralittoral fringe of the south Istrian coast and in the coast of the islands exposed to wave action. In the upper infralittoral *C. barbata* and *C. crinita* were the dominant species. The abundance of *C. barbata* decreased in the direction from the north to the south. *C. crinita* was most abundant in the central part of the coast. Mixed stands composed of both species were frequent in locations intermediately exposed to wave action. *C. compressa* was found in both exposed and sheltered habitats; however *C. compressa* f. *rosetta* was particularly abundant in exposed locations. In intermediately polluted habitats only *C. compressa* was recorded. *C. barbata* and *C. crinita* disappeared from these habitats likely because of anthropogenic pressure's deleterious effects. In the medium-lower infralittoral zone (4-12 m depth), three different types of *Cystoseira* mixed stands were found: (a) *C. barbata*-*C. humilis* stands in very sheltered locations, (b) stands dominated by *C. foeniculacea* and (c) stands dominated by *C. corniculata*. *C. spinosa* was a major component of the last two types. Since the medium-lower infralittoral represents the majority of the rocky bottom of the west Istrian coast, *C. foeniculacea* and *C. corniculata* could represent the most ecologically important species of the study area. Both species are adapted to dwell in habitats with high sedimentation rates (Ercegović 1952).

Distribution patterns of *Cystoseira* spp. can be considered similar to those observed in 1950 (Ercegović, 1952) and in the period 1963-1970 (Munda, 1979; 2000), which suggests a good preservation status of *Cystoseira* populations in the study area. However, in the period 1978-1983 *Cystoseira* spp. were in a phase of regression in the region of Rovinj. *C. amentacea* became rare and was found only in unpolluted locations. *C. barbata* was not found in the area. *C. compressa* disappeared from most locations where it was present in 1967-1970. *C. corniculata*, *C. crinita* and *C. spinosa* became rare. Sparse patches of these three species were present only in islets. *C. foeniculacea* completely disappeared from the vegetation (Munda, 2000). The regression phase lasted until mid 1990s. The *Cystoseira* spp. decline was likely caused by a direct discharge of urban sewage, an increased eutrophication of the northern Adriatic and sea urchins grazing (Munda, 2000; Hanel 2002). In the northern Adriatic, eutrophication levels change at a long-term scale in relation to variations of anthropogenic nutrient load, primarily due to the Po River runoff and mutable oceanographic conditions (Degobbis *et al.*, 2000). The

assessed recovery of *Cystoseira* spp. along the west Istrian coast was likely driven by a marked reduction of eutrophication levels of the northern Adriatic in the period 2000-2009 (Đakovac *et al.*, 2012). Additionally, starting with 1990s the density of sea urchin has decreased, enhancing *Cystoseira* re-colonisation of barrens (Hanel, 2002).

### Acknowledgements

This research was supported by the Ministry of Science, Education and Sport of the Republic of Croatia (projects 0098111), projects Western Istrian COastal Sea (WICOS) and Monitoring of coastal and transitional waters in the Adriatic Sea (Croatian Water Agency)

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## **THE IMPORTANCE OF GENETIC MAKE-UP FOR RESTORATION SUCCESS – A CASE STUDY OF THE SEAGRASS *ZOSTERA NOLTII* HORNEM IN A MEDITERRANEAN LAGOON**

### **Abstract**

*Seagrass meadows are one of the most threatened ecosystems on earth. In order to restore seagrasses and maintain the ecosystem services they provide, transplantation and restoration of meadows is performed for several seagrass species and on different continents. Several studies have highlighted the importance of genetic diversity and make-up for transplantation success in seagrasses, but genetic diversity is still rarely taken into account in transplantation trials. To restore ecosystem functioning in the Mediterranean saline Berre lagoon (France), transplantation of *Zostera noltii* (Hornem) meadows was carried out after a genetic assessment of relict meadows and a marine donor site. The transplantation was followed over four years. The success was extremely low, while relict meadows made a slight recovery. We assessed genetic diversity of newly occurred patches and existing meadows and of the transplanted sites four years after the transplantation. Relict meadows have high genotypic richness values indicating high levels of sexual reproduction, while the assessed transplant site is extremely clonal. The transplant site is moreover genetically distinct from indigenous meadows. The lack of success of transplanted shoots could be due to an adaptation mismatch of the marine donor material to lagoon conditions.*

**Key-words:** *Zostera noltii*, microsatellites, ecosystem recovery, genetic diversity, transplantation

### **Introduction**

Seagrass meadows are recognized as one of the most important and valuable marine biomes, when considering biodiversity, economics and ecosystem services (Spalding *et al*, 2003). Genetic diversity has been shown to be an important contributing factor for ecosystem functioning (reviewed in Hughes & Stachowicz, 2009; 2011; Procaccini *et al*, 2007; Reusch & Hughes, 2006), and for transplantation success in particular, increasing seagrass population growth and fitness (Williams, 2001), associated invertebrate density, nitrogen retention, productivity (Reynolds *et al*, 2012) and growth performance (Procaccini & Piazzini, 2001) and maintaining high levels of genetic diversity and sexual reproduction (Sinclair *et al*, 2013; Olsen *et al*, 2014).

Here we assess the importance of genetic diversity of *Zostera noltii* (Hornem) for transplantation success in the French Berre lagoon, one of the largest saline coastal lagoons in the Mediterranean. In this lagoon, *Z. noltii* was not able to make a natural recovery after huge declines beginning at the end of the 19<sup>th</sup> century, despite legislation to reduce pollution and eutrophication adopted in 1994 and re-enforced in 2006 (Bernard *et al*, 2007). In 2009, restoration of the *Zostera* meadows was identified as a key action for the ecosystem recovery and after a genetic assessment of lagoon meadows and of the close-by fully marine meadow of Carteau (Procaccini *et al*, 2013), it was decided to re-enforce natural populations by transplanting *Z. noltii* from the marine meadow. However, the actual shoots used were not genotyped at the beginning of the transplantation trial. Survival of transplants was initially good (around 50% after 5 months at the here assessed

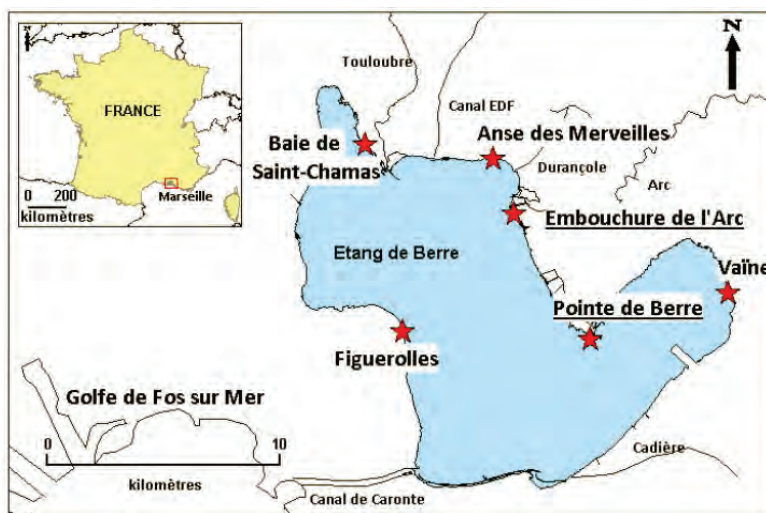


Berre and Arc sites), but was very low after a time frame of four years: 2% at Arc, 0.4% at Berre and 0% for four other sites (Bernard et al., 2013). In contrast, the remaining local meadows have seen a slight recovery and a slow progression towards the coast since 2009 and new patches of *Z. noltii* appeared in close proximity to the transplant sites. In this study we aim to assess whether new patches that were observed in close proximity to transplant sites originate from transplanted shoots or from the relict indigenous meadows which are at an approximate distance of one to two kilometres. Moreover, we investigate whether the low success of the transplantation-trial can be explained by the genetic make-up of the transplanted shoots.

## Material and Methods

### Sampling

Four years after the transplantation, samples were taken from the only two transplant sites where shoots had survived (Berre and Arc), as well as from newly appeared patches close to both sites and a newly appeared meadow in Vaïne (Fig. 1). Approximately 1-2 metres distance was kept between individual samples at each location. Samples were dried and stored in silica crystal before genetic analysis. Samples from Procaccini et al (2013) were also included for differentiation analyses.



**Fig. 1:** Map of the Berre lagoon with the six translocation sites and the two study sites of relict *Zostera noltii* meadows (underlined).

### DNA extraction and genetic analysis

DNA was extracted with the NucleoSpin® 96 Plant II kit (Macherey-Nagel) according to the manufacturer's instruction, following the protocol modified for the robotic station at the Molecular Service of the Stazione Zoologica (Tomasello et al., 2009).

Nine polymorphic microsatellites were used for the analysis of *Z. noltii* (Coyer et al, 2004). Microsatellites were combined in two different multiplexes in 96-well plates and all PCRs were run under the following conditions: 95 °C for 15min, 35 x (94 °C for 30 sec, 60 °C for 1 min 30 sec, 72 °C for 1 min), with a final extension step of 60 °C for 30 min. Analysis of the fragment data was done using the software GenClone (Arnaud-Haond & Belkhir, 2007) for clonal discrimination and identification of multilocus genotypes (MLGs). Genomic diversity measurements, assignment tests and F statistics were calculated using GenAlex (Peakall & Smouse, 2012). Allelic richness (A) was also estimated standardized to four genotypes (the minimum number in the Arc transplant

population) to account for differences in the number of MLGs per population, using the STANDARICH package (<http://www.ccmr.ualg.pt/maree/software.php?soft=sarich>) and R software (R Development Core Team, 2012). Principal coordinate analysis (PCoA), as implemented in GenAlex, was used to explore the genetic distance among populations.

### Results and discussion

Overall, 92 individuals were successfully genotyped at all loci and were used for further analysis. Heterozygosity and allelic richness are high in all groups ranging from 0.429 to 0.670 (observed heterozygosity) and from 2.70 to 2.84 (for allelic richness standardized for 4 genotypes) (

Table 1). Genotypic richness, i.e. the number of distinct clones in the sample, is considerably lower in the Arc transplant site compared to the other sampling sites, where *Z. noltii* re-appeared naturally (

Table 1). The sample from the Arc transplant site consists of only four clones, which may partly be due to the way the transplantation was carried out (using bunches of shoots). Only one sample for the Berre transplant site was collected, so no statements can be made for this site.

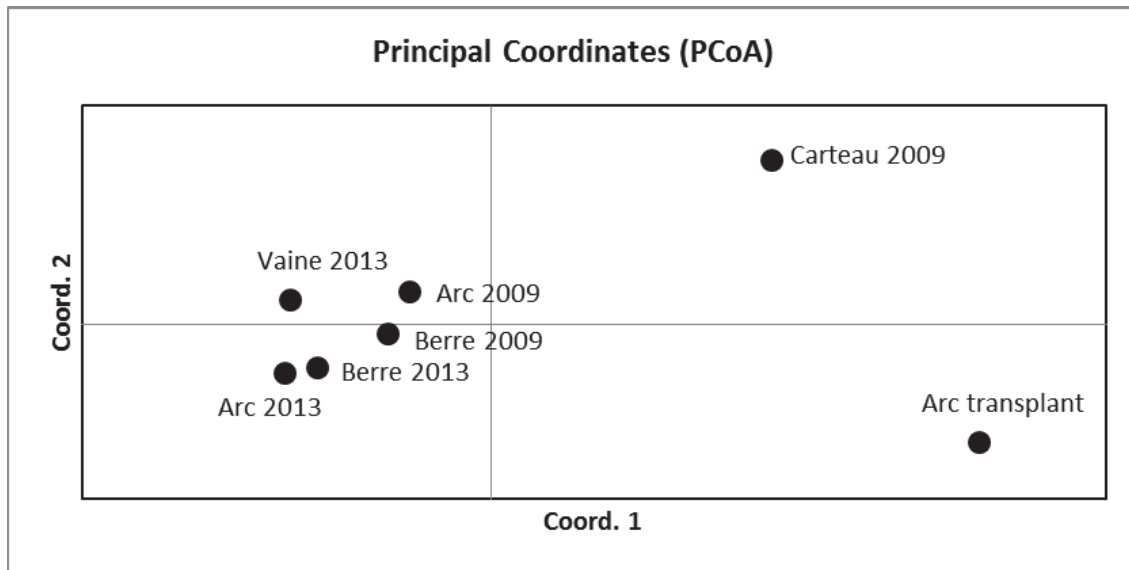
**Table 1 Genetic measurements of the samples collected in the Berre lagoon in 2013. N: total number of ramets analyzed; G: number of genotypes (genets); R: genotypic richness as calculated by  $[G-1]/[N-1]$ ; Na: Numbers of alleles per locus; A<sub>4</sub>: Allelic richness standardized for four genotypes; Ho: observed heterozygosity; He: expected heterozygosity; uHe: unbiased expected heterozygosity; F: fixation index as calculated by  $[1 - (Ho / He)]$ . All estimates regarding heterozygosity were performed after the removal of all but one of the identical genotypes.**

Population		N	G	R	Na	A <sub>4</sub>	Ho	He	Fis
Arc transplant	Mean	15	4	0.21	2.78	2.78	0.583	0.517	-0.143
	+ SE				0.40	0.00	0.102	0.079	0.095
Arc natural	Mean	21	19	0.9	3.89	2.71	0.532	0.535	-0.020
	+ SE				0.61	0.30	0.103	0.093	0.097
Berre transplant	Mean	1	1	NA	1.67	NA	0.670	0.33	-1.00
Berre natural	Mean	19	19	1	3.89	2.84	0.532	0.516	-0.035
	+ SE				0.59	0.32	0.093	0.087	0.040
Vaïne natural	Mean	36	36	1	4.44	2.7	0.429	0.488	0.160
	+ SE				0.71	0.19	0.094	0.100	0.075

A PCoA was performed to group samples according to their genetic similarity on two coordinates, which explain together 65% of the variation. The PCoA clearly shows that the transplanted shoots at Arc are genetically distinct from the new patches that were observed around the transplantation zone (Fig. 2). Samples from patches that newly occurred at Arc, Berre and Vaïne are in contrast quite similar to each other (Fig. 2). In this analysis, samples taken in 2009 at relict *Z. noltii* meadows in the Berre lagoon and the donor site Carteau (Procaccini *et al*, 2013) were included. Sites in the lagoon cluster together with newly occurred patches in the negative side of coordinate 1, indicating an overall similarity, while the transplanted site and the marine donor site are both in the positive side of the same coordinate (Fig. 2). This indicates an overall similarity between

new patches and lagoon sites, which suggests that *Z. noltii* is slightly expanding in the Berre Lagoon, probably due to improved salinity conditions. Newly occurring patches do not only mostly originate from existing meadows within the lagoon, but also have high genotypic richness, suggesting recruitment from sexual reproduction via seeds produced within the lagoon. The PCoA was also performed only on distinct genotypes for each population and population distances were comparable.

**Fig. 2: PCoA of samples collected in the Berre lagoon in 2013 (Arc transplant, Arc 2013, Berre 2013 and Vaïne 2013) and in 2009 (Berre, Arc and Carteau).**



Despite the close grouping of new patches from Vaïne, Berre and Arc in the PCoA in Figure 2, the genetic make-up at each location is distinct as seen in the assignment test shown in Tab. 2. Overall, 86% of samples are assigned to their own population. This genetic structure indicates that meadows are genetically distinct, although there is some gene flow within the lagoon. No samples collected within the lagoon, neither natural sites nor transplants, were assigned to Carteau, and all shoots sampled in Carteau in 2009 get assigned to their own population. This indicates the isolation of the lagoon from the open sea, despite the fact that Carteau represents the donor site for transplants. Sampling for the genetic study in 2009 and sampling for transplants took place in different parts of the meadow several hundred meters apart, but this is nevertheless a surprising result. Finally, two genotypes collected in the transplanted site of Arc, seem to derive from new Arc patches, suggesting that environmental conditions at the transplant site have sufficiently recovered for recruitment of indigenous shoots.

### Conclusions

This study is one of few examples where genetic data is available for a transplant-trial in seagrasses before and after restoration. We show that the naturally re-occurring and relict meadows in the Berre lagoon are genetically distinct from the donor material used for the transplants. The newly occurred patches at Berre and Arc and the meadow at Vaïne are genetically distinct from each other, although the assignment test shows that there is some gene flow between naturally re-appearing and relict meadows in the lagoon. High genotypic richness values in these meadows are proof of high levels of sexual reproduction.

**Tab. 2: Assignment test. Shown is the number of individuals assigned to their own population (Self Pop) or to another population (Other Pop), with information on which other population they get assigned to (To).**

Pop	Self Pop	Other Pop	To
<b>Berre 2009</b>	24	12	Arc 2013, Berre 2013, Berre transplant
<b>Arc 2009</b>	33	2	Arc transplant, Vaïne 2013
<b>Arc transplant</b>	13	2	Arc 2013
<b>Arc 2013</b>	19	2	Vaïne 2013, Berre 2013
<b>Berre transplant</b>	1	0	
<b>Berre 2013</b>	17	2	Arc 2013, Berre 2009
<b>Vaïne 2013</b>	29	7	Berre 2013, Arc 2013, Berre 2009, Arc 2009
<b>Carteau 2009</b>	31	0	
<b>Total</b>	167	27	
<b>Percent</b>	86%	14%	

It is very clear that the newly appeared patches in close proximity to the transplant sites derive from the lagoon gene-pool - not from the genotypes used for transplantation or from outside the lagoon. We even find indigenous genotypes at the transplant sites, demonstrating that environmental conditions and disturbance cannot be solely responsible for the failure of the transplants. Instead, the results indicate that there is an adaptation mismatch for the marine shoots in lagoon conditions. Moreover, genotypic richness is low at the transplant site at Arc, but very high at the other locations. We did not genotype the transplants at the beginning of the trial, so this could either be because of the use of few distinct genotypes in the transplantation or because few genotypes were able to persist. Mortality rates of the assessed restoration project were initially good, but very high on the time scale of years. This study therefore shows that it is crucial to follow up on transplantation studies for a time-frame of several years to capture delayed mortality that is not necessarily related to problems of the transplantation itself, but to lack of adaptive potential or plasticity of the used donor material. This study also shows that improving the natural conditions is crucial before considering re-enforcements. When transplantations are nevertheless deemed necessary to re-enforce local populations, the genetic make-up needs to be taken into account. Especially in highly dynamic and disturbed environments such as lagoons, the local adaptation of individuals seems to play an important role.

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## **MODELLING THE REFERENCE CONDITIONS OF THE UPPER LIMIT OF *POSIDONIA OCEANICA* MEADOW**

### **Abstract**

*The European directive 2008/60/EC (MSFD: Marine Strategy Framework Directive) requires Member States to define the ecological quality of marine environment. The purpose of the MSFD is to attain, or maintain, a good environmental condition of the marine environment by 2020. In order to achieve this goal, reference conditions of a given environment can be defined also by modelling. In this paper, a modelling approach based on 2D mapping with predictive equations formerly developed at regional scale (Liguria region) is applied at the Mediterranean basin-wide spatial scale. This predictive model identifies a portion of the seabed (the buffer zone) where the upper limit of meadows of *Posidonia oceanica* (L.) Delile, the dominant seagrass in Mediterranean coastal waters, is expected to be located under natural conditions, thus representing its reference conditions. This approach has been applied to four Mediterranean coastal areas with different coastal morphologies and hydrodynamic characteristics. For the theoretical determination of the buffer zone, in which the meadow upper limit is expected to lay, two predictive equations were applied to each area and data were processed by GIS to represent the actual position of the upper limit. Results show good applicability of the predictive 2D model also at the Mediterranean scale, but its applicability is restricted to those meadows settled on sandy or matte substrate. Application of this model at the Mediterranean scale allows to define the reference conditions of a priority coastal habitat and thereby to quantify the regression of *P. oceanica* meadows caused by human activities and natural impacts (e.g. alterations generated by rivers).*

**Key-words:** Seagrass, ecological modelling, reference conditions, *Posidonia oceanica*, Mediterranean Sea.

### **Introduction**

The sudden and continuous development of coastal cities in the last century incited the European Community to promulgate the Directive 2000/60/EC known as the Water Framework Directive (WFD): this document obligates the Member States to protect the water inland, surface waters, transitional waters, coastal waters and groundwater and wetlands through integrated management (EC, 2000). However the Directive presents different gaps, for example it does not include the analysis of ecological elements within spatial heterogeneity in three-dimensional space (Borja, 2005). The different theoretical and practical shortcomings of the WFD have been outdone with the enactment of Directive 2008/56/EC known as the Marine Strategy Framework Directive (MSFD) (EC, 2008). The MSFD establishes a framework for development of marine strategies designed to achieve GES (Good Environmental State) in the marine environment by the year 2020, using a holistic functional approach (Borja et al., 2010). Each Member State must identify type-specific reference conditions for each marine region or subregion and define the environmental targets and associated indicators for marine waters, necessary for GES

achievement. Reference conditions can be determined by: (I) comparison of the study area with similar pristine areas (Marine Protected Areas and Sites of Community Importance); (II) comparison with historical data; (III) modelling (EC, 2008). Due to binding European Union Directives, efforts have recently been made in most European countries to manage coastal marine ecosystems on the basis of a solid ecological background and recent scientific modelling (Vacchi et al., 2014).

*Posidonia oceanica* is the most important and abundant seagrass of the Mediterranean Sea and its meadows constitute a key coastal habitat (Boudouresque et al., 2012). The meadow upper limit usually occurs within the most dynamic region of the seafloor (Infantes et al., 2009; Vacchi et al., 2010, 2012, 2013) but this area is often subjected to the influence of anthropogenic impacts, which cause an offshore retreat of upper limit (Pergent et al., 1995; Montefalcone et al., 2010). In this paper, we applied a geospatial predictive model formerly developed at regional scale (Liguria region) and now implemented at Mediterranean scale. This model identifies the seafloor portion where the pristine meadow upper limit is expected to be located, thus determining the reference conditions.

### Materials and methods

For the theoretical determination of reference conditions, represented on maps as a buffer zone, we applied two equations to each study area. The model was tested in four Mediterranean coastal areas having different wave climate and geomorphological setting, but all meadows settled on sandy or mat substrate: Saleccia (northern Corsica), Acharavi (northern Corfu island), Marina di Pescia Romana - Montalto di Castro (Latium, central Italy) and Alassio (Liguria, northern Italy). The conceptual framework of this model is based on three essential components.

(I) Definition of the geometry breaking limit using breaking depth ( $d_b$ ). The breaking depth ( $d_b$ ) is the depth where waves break and was calculated using the formula  $d_b = H_b/\gamma_b$  (Smith, 2003), where  $H_b$  is the offshore wave high (return time 1 year) and  $\gamma_b$  is the breaking index (Weggel, 1972). For determination of breaking limit, the software Mike 21 sw ([www.dhi-italia.it](http://www.dhi-italia.it)) was used. Wave data were obtained from APAT (2006). For this study  $H_s$  (mean annual significant wave height; Sorensen, 2006) and  $T_s$  (significant wave period) are considered with one-year return time.

(II) Definition of the morphodynamic domain of the beach, i.e. the distinctive type of beach produced by topography and wave climate. The surf scaling index ( $\epsilon$ ) was applied:  $\epsilon = aw^2/g \tan^2\beta$  ( $a$ : breaker amplitude,  $w$ : incident wave radian energy,  $g$ : acceleration of gravity,  $\beta$ : slope of the beach in the surf-zone; Dean and Dalrymple, 2004). This index allows determining the values of two parameters:  $K_{min} = 5.94 + 0.29\epsilon$  and  $K_{max} = 17.83 + 0.41\epsilon$  (Vacchi et al., 2014), which represent, respectively, the minimal and the maximal theoretical distance of the meadow upper limit from the breaking depth. A GIS-based overlay analysis was performed to identify the portion of the seabed where the meadow upper limit should lie under natural conditions, represented as a buffer zone in the form of georeferenced polylines.

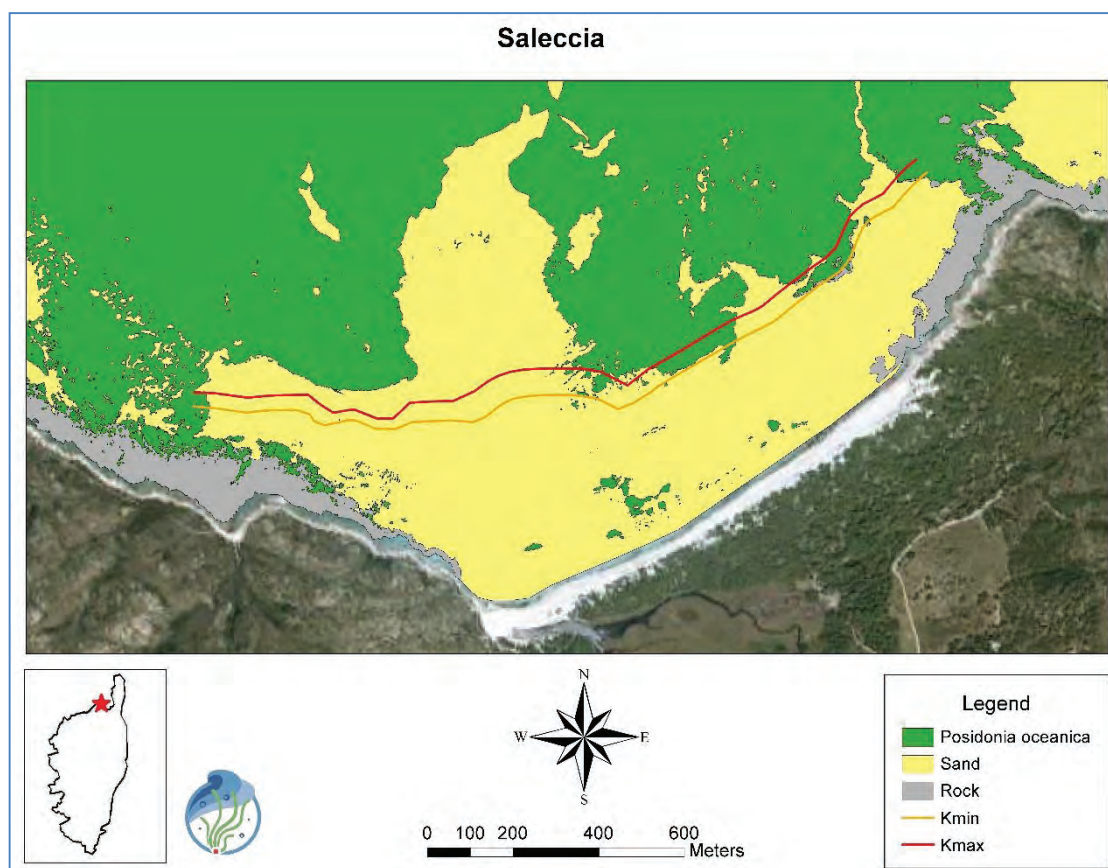
(III) Definition of the *P. oceanica* upper limit geometry using thematic bionomic 2D maps (1:10000) in digital format or detailed aerial photos.

### Results

The breaking depth is located at 4 m (Alassio), 6 m (Acharavi) or 7 m (Saleccia and Marina di Pescia Romana - Montalto di Castro), but its distance from the shoreline is

highly variable due to the different morphodynamic domains of the different study areas. In Saleccia the meadow is very close to the shoreline and its upper limit is located within the buffer zone defined by  $K_{min}$  and  $K_{max}$  parameters (Fig. 1), but in the central area of the meadow where the limit shows interrupted by sandy channels perpendicular to the coastline. In Acharavi the meadow has a very sharp upper limit and it places entirely within the buffer zone. In both Alassio and Marina di Pescia Romana - Montalto di Castro meadows the upper limit is placed only partially inside the buffer zone; in the southern sector it is retreated seaward and is not well defined showing an alternation of living *P. oceanica* and dead matte areas.

**Fig. 1:** Example of cartographic representation of  $K_{min}$  and  $K_{max}$  parameters (i.e., the buffer zone, corresponding to the reference conditions) and *Posidonia oceanica* meadow for Saleccia study area.



### Discussion

This model was previously validated along the Ligurian coast (NW Italy) and the present study suggests its applicability to the whole Mediterranean Sea. In Alassio and in Marina di Pescia Romana - Montalto di Castro, the two meadows showing signs of regression in correspondence of their upper limits, even where these limits fall inside the buffer zone they are very close to  $K_{max}$  value. The development of local human activities in these two study areas caused an alteration of the seagrass conservation state, highlighted by the presence of dead matte areas in correspondence of the upper limit. In Acharavi the upper limit is located within the reference conditions, although there are some minor variations caused by local changes in the seafloor morphology. In Saleccia the upper limit is placed inside the buffer zone and its shape in the central area of the meadow is likely due to the

influence by the Liscu River. Although the predictive model showed easily applicable to different Mediterranean meadows settled on sandy or matte bottoms, it cannot apply to rocky substrate, where the *P. oceanica* rhizomes are able to anchor strongly allowing the development of the meadow beyond the breaking limit. It is important to underline that the accurate position of  $K_{\min}$  and  $K_{\max}$  parameters depends strictly on the data quality, for example map scale. The presence of anthropogenic structures along the investigated coast might also modify wave characteristics. To improve the performance of the predictive model, it would be necessary to apply it in other Mediterranean beaches.

### Acknowledgments

The authors would like to thank for data concession: Dipartimento Ambiente (Regione Liguria), Gerard Pergent (Université de Corse Pascal Paoli), Giandomenico Ardizzone and Alessandro Criscoli (Università di Roma La Sapienza), and Eleana Karkani (University of Athens).

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## **COMBINING MODELLING AND HISTORICAL DATA TO DEFINE THE STATUS OF *POSIDONIA OCEANICA* MEADOWS**

### **Abstract**

Coastal marine ecosystems are particularly sensitive to global change due to natural and anthropogenic causes. Meadows of *Posidonia oceanica* (L.) Delile (Angiospermae, Posidoniaceae) are key Mediterranean coastal ecosystems that have been experiencing an alarming decline over the last decades. Regressed meadows have been shown to be prone to colonization by substitutes, either the seagrass *Cymodocea nodosa* or algae of the genus *Caulerpa*. All these substitutes exhibit a lower engineering capacity than *P. oceanica*, and thus lead to profound structural and functional changes in the ecosystem. As requested by the EU Marine Strategy Framework Directive, status of marine ecosystems has to be assessed through comparison with reference conditions, which can be defined by means of i) historical data, ii) pristine areas, and iii) modelling. In this study we combined a predictive model with historical data to define the reference conditions of the *P. oceanica* meadow of Bergeggi (Ligurian Sea, NW Mediterranean) and to evaluate changes in the meadow during the last 25 years. Starting from 1987, information collected through the years along a permanent underwater transect was available. The meadow experienced a dramatic decline through time and an ecosystem phase-shift due to the appearance of dead matte and the establishment of two substitutes, *Cymodocea nodosa* and the alien green alga *Caulerpa cylindracea*. In recent years the position of the upper limit of the meadow has been showing signs of recovery and has been slowly going back to the reference condition defined by the predictive model; from a compositional point of view, however, the arrival of the fast alien invader *C. cylindracea* has driven the ecosystem into an alternative state that seems hardly reversible.

**Key-words:** *Posidonia oceanica*, *Caulerpa cylindracea*, modelling, reference conditions, phase shift

### **Introduction**

The endemic seagrass *Posidonia oceanica* (L.) Delile is a key species (Pergent, 1992) forming extensive meadows which are the dominant and most productive coastal ecosystem in the Mediterranean Sea (Boudouresque *et al.*, 2006). Thanks to its capacity to build a “matte” (a persistent terrace formed by the interlace of rhizomes, roots and the entrapped sediment), the species can be viewed as an ecosystem engineer (Koch, 2001). In the last decades *Posidonia oceanica* meadows are experiencing a widespread decline (Peirano & Bianchi, 1997). There is growing evidence (Bianchi & Peirano, 1995) that, in degraded meadows, *P. oceanica* may be replaced by the other common Mediterranean seagrass *Cymodocea nodosa* (Ucria) Ascherson and/or by three species of the green algal genus *Caulerpa*, the native Mediterranean species *Caulerpa prolifera* (Forsskål) Lamouroux and the two alien species *Caulerpa taxifolia* (Vahl) C. Agardh and *Caulerpa cylindracea* (Forsskål) J. Agardh (Montefalcone *et al.*, 2007). The latter seems to be the fastest invader and has the higher colonisation potential among all the

substitutes. A community phase shift already begins with the appearance of dead matte areas within a meadow, which may remain uncolonised or be re-colonised by any of the potential substitutes, thus advancing the level of phase shift. *Cymodocea nodosa* and the three species of *Caulerpa* are not able to form structures comparable to the high and lignified matte built by *P. oceanica* (Molinier & Picard, 1952). The replacement of a “constructional” species like *P. oceanica* by a “non-constructional” species may be seen as a significant community change, which can cause dramatic modification in ecosystem structure and functioning (Hughes *et al.*, 2005). Regression and consequent seaward deepening of the upper limit of *Posidonia oceanica* meadows are generally considered to be due to anthropogenic pressures (Boudouresque *et al.*, 2006), although the influence of natural changes in environmental conditions has also been recognised (Giovannetti *et al.*, 2008). In absence of major human pressures, the structure of the upper limit of *P. oceanica* meadows and its position along the region of the underwater beach profile mostly reworked by wave action, are mainly controlled by local nearshore hydrodynamics. Vacchi *et al.* (2010, 2014) developed a predictive model to locate the upper limit position, providing evidence that the natural position of the meadow upper limit, in absence of major human pressures, can be predicted by physical parameters alone. The model delineates a region of the seafloor that can be considered as the baseline, i.e. the reference condition zone where the meadow upper limit should lie in natural conditions (governed only by hydrodynamics, in absence of significant anthropogenic impact). The problem of "sliding baseline", i.e. the change of baselines with time, is always been a difficult issue to resolve (Knowlton & Jackson, 2008; Hobday, 2011). Assessing the natural position of seagrass meadows limits is crucial to define their reference status and to disentangle the relative role of natural processes and anthropogenic impacts (Montefalcone *et al.*, 2010b). As asked by the recent EU Marine Strategy Framework Directive, the status of marine ecosystems has to be assessed through comparison with reference conditions, which can be defined by means of: i) historical data; ii) pristine areas; iii) predictive models. Historical data are often unreliable, pristine areas do not exist anymore, and the application of predictive models alone may not be sufficient. Thus, a multidisciplinary approach is necessary. In this study, we combined historical data with a predictive model to define the natural position of *P. oceanica* meadow upper limit and to assess the evolution over time of the meadow of Bergeggi (Ligurian Sea), where a Marine Protected Area (MPA) was established in 2007. For this area, historical data spanning a period of 25 years, collected through underwater transects, were available, thus providing information for a diachronic analysis.

### **Materials and Methods**

The meadow of *Posidonia oceanica* of Bergeggi extends uninterruptedly for about 83 ha. In summer 1987, 1992, 2004, 2009 and 2012, a 600 m long underwater transect perpendicular to the coast was dived to collect the following data: depth, type of substrate, morphology of the meadow, and the linear extent of five descriptors (living *P. oceanica*, dead matte, sand, *Cymodocea nodosa*, *Caulerpa cylindracea*). To evaluate the structural evolution of the meadow, a multivariate analysis was performed on cover data for the five periods investigated. Bray-Curtis similarity index was applied to  $\log(x+1)$  transformed data, and a MDS ordination was performed. To evaluate the spatial regression of the upper limit change of the meadow, the predictive model proposed by Vacchi *et al.* (2014) was applied. According to this model, three distinct

zones can be recognised within the *P. oceanica* meadows down the profile of the submerged beach: zone a, between the shoreline and the breaking depth, where only dead *matte* is occasionally found; zone b, between the breaking depth and the closure depth, where meadows are often in poor condition, with reduced cover of living *P. oceanica* and constant occurrence of dead *matte*; zone c, beyond the closure depth, where meadows flourish. Under natural conditions, the upper limit should generally lie in zone b, at a distance from the closure depth comprised between  $K_{\min}$  ( $= 5.94 + 0.29\varepsilon$ , where  $\varepsilon$  is the surf scaling index) and  $K_{\max}$  ( $= 17.83 + 0.41\varepsilon$ ).

## Results

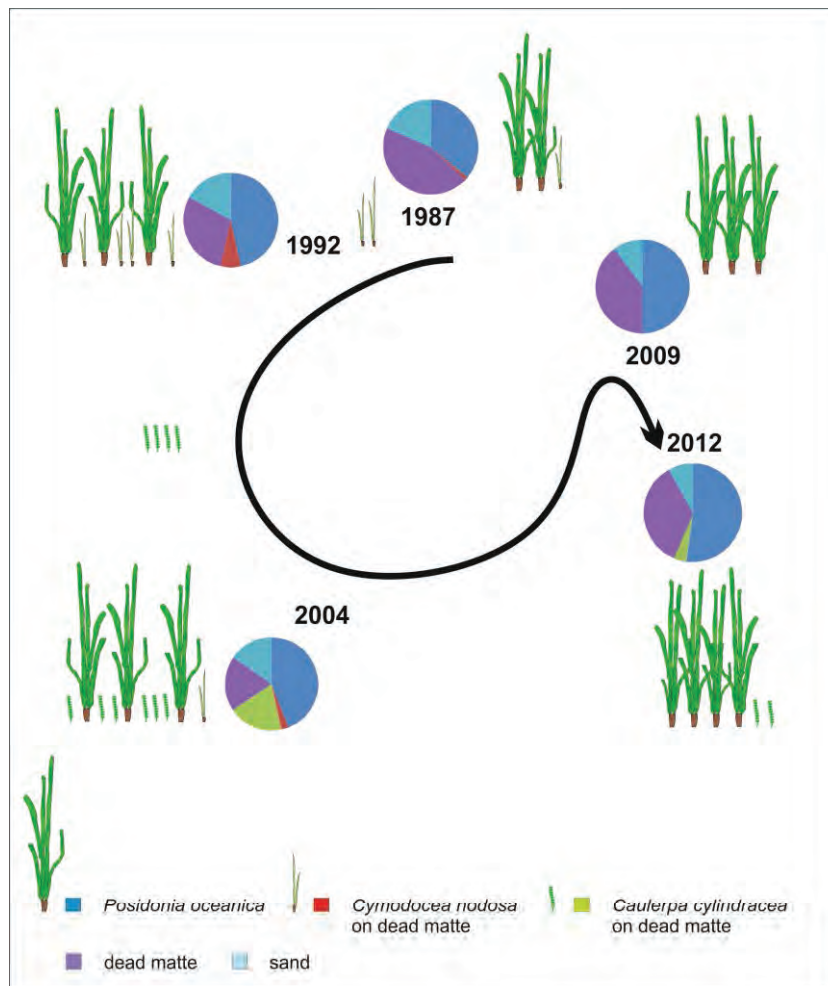
The diachronic analysis evidenced differences in the linear extent of the five descriptors. From 1987 to 2012, *Posidonia oceanica* along the transect increased at the expense of sand. *Cymodocea nodosa* increased from 1987 to 1992, when it reaches its maximum development before declining in 2004 and disappearing in subsequent years. In 2004, the alien species *Caulerpa cylindracea* appeared, colonizing areas of dead *matte* to occupy a quarter of the length of the transect. In 2009, *C. cylindracea* disappeared to reappear in 2012, although less abundant than in 2004. Multivariate analysis (Fig. 1) showed a shift between 1987 and 1992 due to *C. nodosa* colonizing dead *matte* areas, and major change in 2004 as a result of the appearance of *C. cylindracea*. In 2009, the system seemed to return to a state similar to the original one thanks to the lack of substitutes. However, the reappearance in 2012 of *C. cylindracea* determined a further change along a new trajectory. The upper limit of *P. oceanica* in the last 25 years has never been found within the area of seabed defined by  $K_{\min}$  and  $K_{\max}$  (Fig. 2). In 1987, the limit was at the closure depth (regressed about 26 m from the theoretical limit considered as the reference condition) and the surveys in the subsequent years confirmed an even stronger regression (34 m in 1992 and 37 m in 2004). In 2009, however, a recovery of the meadow was apparent, as the distance between the upper limit and the reference condition decreased to 7 m. In 2012, the upper limit was recorded 12 m offshore with respect to the reference condition.

## Discussions and Conclusions

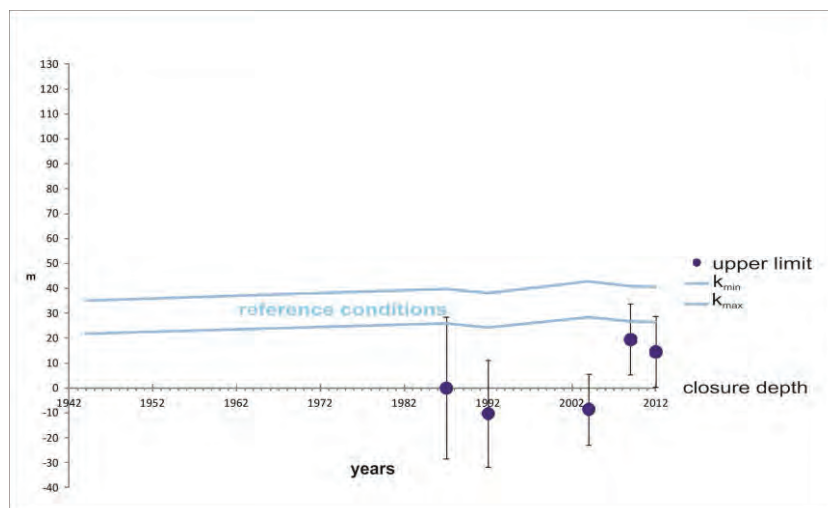
The meadow of *Posidonia oceanica* in the Bergeggi MPA showed, in 1987, a situation of advanced regression, witnessed by the dead *matte* that covered almost half the entire length of the transect. Colonization by *Cymodocea nodosa* in 1992 and by *Caulerpa cylindracea* in 2004, advanced the phase shift already started with the great extent of dead *matte* (Montefalcone *et al.*, 2007). In 1992, the replacement of *P. oceanica* by *C. nodosa*, a pioneer species in its primary ecological succession, caused a change in the ecosystem such that, in the absence of further disruptions, a return to the original state through a secondary succession could theoretically be possible. In contrast, the appearance in 2004 of *C. cylindracea*, whose arrival is a stochastic event (Montefalcone *et al.*, 2010a), has provoked the complete regression of *C. nodosa* and a strong phase shift that is difficult to reverse. *C. cylindracea* is a strong invader that is not part of the primary succession of *P. oceanica* meadows (Montefalcone *et al.*, 2007, and references therein). The absence of substitutes recorded in 2009 suggested a possible return to the initial state of the system, but the finding once again of *C. cylindracea* in 2012 indicated that the system remained in an alternative stable state, thus undergoing hysteresis (Montefalcone *et al.*, 2011). Periodic fluctuations in abundance within the area of

geographic distribution have already been observed in species of the genus *Caulerpa* (Meinesz, 1973).

Although in recent years the Bergeggi meadow has showed some recovery in term of advancing upper limit and increasing *P. oceanica* extent, the ecosystem is following a path different from the one that led to the change. The unpredictable behaviour of alien species confirms the need to monitor them over time in order to verify its future evolution and their effect on the habitat.



**Fig. 1: Structural change in the Bergeggi meadow in 25 years. The position of the five years represents their relative positioning in the MDS graph (2D Stress: 0)**



**Fig. 2: Spatial change relative to the upper limit of the meadow. The X axis (years) represents also the closure depth (0 value of the Y axis). The area between  $K_{min}$  e  $K_{max}$  is the reference condition defined by the model where the meadow upper limit should lie in natural conditions.**

The predictive model of Vacchi *et al.* (2010, 2014) has been useful to define the reference condition of the meadow of Bergeggi. In recent years, the upper limit has been re-approaching the reference condition: full improvement of environmental protection within the Bergeggi MPA might help the upper limit regaining its original position. The simple formulation and the easy application of the predictive model represents an innovative management tool for future studies because it support the discrimination of the influence by anthropogenic and natural causes on the development of *P. oceanica* meadows. The novel approach of combining the predictive model with historical data to evaluate changes seems an excellent approach to evaluate the evolution and the status of the meadows, improving the potential of monitoring for this priority habitat.

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## **PRELIMINARY STUDY ON THE DISTRIBUTION OF *POSIDONIA OCEANICA* ALONG THE DARDANELLE STRAIT**

### **Abstract**

*Posidonia meadows, being one of the most reliable bioindicators of a disturbed environment, were investigated in Dardanos Bay (Dardanelles Strait). This site deserves attention due to both excessive anthropogenic impacts and natural hydrodynamics of the strait. Providing information that can be extrapolated to the northern extension of *Posidonia oceanica* towards the Marmara Sea was also aimed. Lower limit was marked in order to be used for long-term monitoring. Several parameters such as meadow cover, meadow density, and type of limit were noted according to the standardized in situ measurements. The lower limit is situated at 17 m depth, meadow cover average value is 47.47 % and the percentage of plagiotropic rhizome is 51.82%. The phenological and lepidochronological property were also analyzed. Finally the Ecological Quality Ratio was calculated and the values between 0.42-0.52 correspond to a "poor" vitality for Dardanos meadow, according to the BiPo index.*

**Key-words:** *Posidonia oceanica*, Dardanelles Strait, phenology, lepidochronology

### **Introduction**

Magnoliophytes have a considerable ecological weight in coastal environments; many of them are ecosystem engineers, or at least key species. This is so for the Mediterranean. Among the other Magnoliophyte species, *Posidonia oceanica* (L.) Delile, is the most abundant seagrass in the Mediterranean Sea covering about 38.000 km<sup>2</sup> of coastal off shore sandy and rocky areas (Pergent-Martini & Pergent, 1994; Green & Short, 2003; Pergent, 2007). Its distribution is fairly well recorded (Green & Short, 2003). However, until Meinez *et al.* (2009) recorded the distribution of *P. oceanica* in the north-eastern side of Mediterranean basin and in the Marmara Sea, it was unclear particularly at the narrow Dardanelle Strait where Aegean Sea meets the Marmara Sea.

On the other hand an "Action Plan for the conservation of marine vegetation in the Mediterranean Sea" has been adopted by the Contracting Parties to Barcelona Convention in 1999 (UNEP-MAP-RAC/SPA, 1999). The Med*Posidonia* Programme that was initiated by RAC/SPA in partnership with the Total Corporate Foundation for Biodiversity and the Sea aims to obtain additional information on the presence and evolution of *P. oceanica* meadows in four Mediterranean countries (Algeria, Libya, Tunisia and Turkey) with a view to using these data to elaborate and/or adjust their conservation and sustainable use of biodiversity programs and to enhance capacity-building of national teams in order to ensure long-term monitoring (Pergent *et al.*, 2013). We aimed to determine the distribution and condition of *Posidonia* meadows located at Dardanos Bay along the Dardanelle Strait in order to contribute to the existing data and to set up a long-term monitoring infrastructure. The phenological and lepidochronological properties of the meadows were analyzed. Lower limit was marked. Several parameters such as meadow cover, meadow density, and type of limit were noted according to the standardized in situ measurements.

## Material and Methods

The study was performed in Dardanos Bay (Dardanelles Strait, Çanakkale, Northwestern part of Turkey) in February-May 2014. This bay deserves attention due to both excessive anthropogenic impacts and natural hydrodynamics of the strait.

Cirik *et al.* (2006) determined the lower limits of the meadow by means of SCUBA diving and GPS positioning. The monitoring system, standardized by Pergent *et al.* (2007), was set up along the lower limit of the meadow. Fifteen markers were placed with 10 m intervals, on the edge of the lower limit (Tab. 1 and Fig. 1), in order to follow any changes or evolution (Pergent, 2007; Boudouresque *et al.*, 2012). The precise position of each marker was recorded and density, bottom cover, percentage of plagiotropic rhizomes and exposure or burial of rhizomes was measured according to Pergent (2007). The types of limits (progressive, sharp C+, sharp C-, sparse or regressive) were also recorded. Three orthotropic rhizomes were collected at each station. Reference station was chosen at 15m depth, to assess the vitality of the meadow. The upper limit of the meadow was determined. The samples, which were collected from upper, lower and reference limits, were preserved for laboratory analysis. Phenologic and lepidochronologic measurements of the samples were done in the laboratory. All these parameters were integrated in standardized grid to determine the vitality of the meadow according to Pergent (2007) (Tab. 1 to 3).

All data were analyzed by one-way analysis of variance (ANOVA) with Tukey's multiple comparison tests. The differences were considered significant when  $p < 0.05$ . Before the ANOVA, all data were checked for homogeneity of variance (Levene's test for equal variances) and normal distribution (Anderson–Darling test) using the PASW STATISTICS 19 software for Windows (IBM SPSS Inc., Chicago, IL).



**Fig. 1: The study area and the locations of the markers.**

**Tab. 1: Evaluation of the type of lower limit (Pergent, 2007).**

Type of limit	Description	
Progressive	Plagiotrophic rhizome in front of the limit	High
Sharp High Cover	Sharp limit with meadow cover >25 %	Good
Sharp Low Cover	Sharp limit with meadow cover < 25 %	Moderate
Sparse	Shoot density < 100 shoots/m <sup>2</sup> and cover 15 %	Poor
Regressive	Dead mate in front of the limit	Bad

**Tab. 2: Standardized values of deep of lower limit (m), bottom cover (%) and Plagiotrophic rhizomes (%) (Pergent, 2007).**

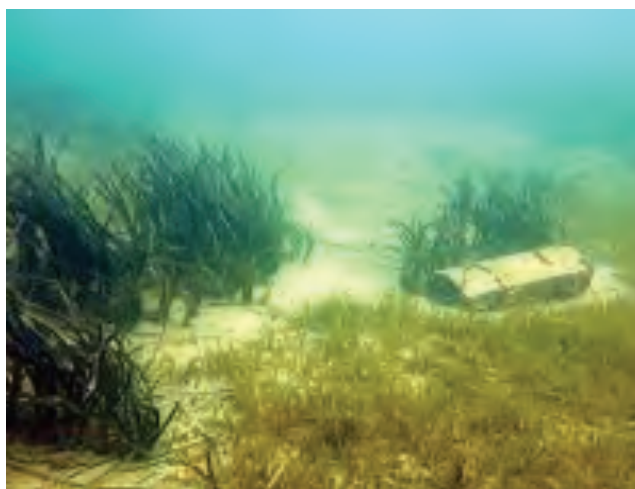
	High	Good	Moderate	Poor	Bad
<b>Depth of Lower Limit (m)</b>	>34.2	34.2 – 30.4	30.4 – 26.6	26.6 – 22.8	<22.8
<b>Leaf cover (%)</b>	>35	35 – 25	25 – 15	15 – 5	< 5
<b>Plagiotrophic rhizomes (%)</b>	>70	70-30	< 30		

**Tab. 3: Standardized values of foliar surface (in cm<sup>2</sup> per shoot), number of leaves produced per year and rhizome elongation (in mm per year) at reference depth (-15m) (Pergent, 2007).**

	High	Good	Moderate	Poor	Bad
<b>Foliar surface (cm<sup>2</sup> shoot<sup>-1</sup>)</b>	>362	362 – 292	292 – 221	221 – 150	<150
<b>Number of leaves produced (y<sup>-1</sup>)</b>	>8.0	8.0 – 7.5	7.5 – 7.0	7.0 – 6.5	<6.5
<b>Rhizome elongation (mm y<sup>-1</sup>)</b>	>11	11 – 8	8 – 5	5 – 2	<2

## Results

Firstly, we searched for markers which had been set up eight years ago. It was discovered that, only four markers out of fifteen were at their original positions and 11 markers were forced out of their positions. Also, *Caulerpa racemosa*, invasive alien green algae, was observed between 17 – 25 m depth along the Dardanos Bay (Fig. 2).

**Fig. 2: *Caulerpa racemosa* which distributed along the lower limit of the meadow.**

The representative portion of the lower limits of the meadow equipped with fifteen markers and photographs of each marker were taken in order to provide the most precise reference base. It was found out that the lower limit was at 17 m depth and the meadow showed sparse distribution. The depth of lower limit of *Posidonia* meadow is classified as poor (See Tab. 3) according to Pergent (2007). The markers' coordinates and main characteristics of the meadow around the markers were shown in Table 4.

**Tab. 4: GPS locations and main characteristics of the meadow where marked out in Dardanos Bay**

No	Latitude	Longitude	Cover (%)	Density (fascicles m <sup>-2</sup> )	Plagiotropic rhizome (%)	Exposure (cm)
1	40°04'42.80''N	26°21'12.30''E	28	45	44	-1,5
2	40°04'42.34''N	26°21'11.45''E	20	35	46	-1
3	40°04'41.50''N	26°21'11.90''E	52	70	34	-2
4	40°04'40.43''N	26°21'11.48''E	72	132	67	-2
5	40°04'39.30''N	26°21'10.60''E	60	100	54	-1,5
6	40°04'38.12''N	26°21'10.44''E	32	45	66	-1,5
7	40°04'37.40''N	26°21'09.67''E	48	52	42	-1
8	40°04'36.44''N	26°21'09.65''E	64	85	57	-1,5
9	40°04'35.63''N	26°21'09.40''E	60	60	58	-1,5
10	40°04'34.70''N	26°21'09.00''E	48	77	39	-1,5
11	40°04'33.60''N	26°21'08.70''E	40	96	40	-2
12	40°04'32.70''N	26°21'08.20''E	48	45	44	-2
13	40°04'32.07''N	26°21'07.76''E	36	50	58	-2
14	40°04'31.40''N	26°21'07.91''E	32	26	57	-2
15	40°04'30.60''N	26°21'08.08''E	72	102	47	-1,5

The meadow density of lower limit varied from 26 - 102 shoots m<sup>-2</sup>; with an average of 64.93 ±28.72 shoots m<sup>-2</sup>. The cover ratio of meadow ranged from 20 to 72 % with an average value of 47.47 ±16.06 %. The average plagiotropic rhizome ratio was found as 51.82 ±12.13 %. The exposures of the rhizomes were measured between -2 to -1 cm.

The shoots were sampled along the lower limit, the reference depth and the upper limit for phenological and lepidochronological analysis. The average number of leaves produced annually was 6.46 ±0.50 and the average growth rate of the rhizomes was 6.56 ±0.44 mm yr<sup>-1</sup>. The average leaf number was found 5.7 ±0.6 fascicles<sup>-1</sup>. Average length and width measured as 34.48±12.37 cm and 1.00±0.15 cm. The leaf area index (LAI) changed from 26.84±12.46 to 45.89±16.6 cm<sup>2</sup> m<sup>-2</sup>. At the reference depth, the average foliar index was found 153.30±20.76 cm<sup>2</sup>.fascicles<sup>-1</sup>. The LAI was also calculated as 39.38±14.49 cm<sup>2</sup> m<sup>-2</sup>. The samples which were collected from upper limit showed us that, the average leaf number was changed from 4.25±0.71 to 4.63±0.52 fascicles<sup>-1</sup>. The maximum LAI was calculated as 23.84±11.19 cm<sup>2</sup> m<sup>-2</sup> (Tab. 5).

**Tab. 5: Phenological parameters measured at Dardanos Bay at the lower limit, 15 m depth (reference site) and upper limit level.**

	Lower Limit			Reference depth (15m)			Upper Limit		
	Adults	Intermed.	Global	Adults	Intermed.	Global	Adults	Intermed.	Global
Nb leaves (shoot <sup>-1</sup> )	3.5±0.3	3.0±0,5	5.7±0.6	3.7±0.6	3.0±0.0	3.3±0.5	4.6±0.5	4.3±0.7	4.4±0.6
Length (cm)	43.7±10.1	29.2±10.1	36.5±12.4	38.8±8.5	33.1±14.8	35.9±12.1	24.7±8.8	25.4±11.6	25.1±10.2
Width (cm)	1.0±0.2	1.0±0.1	1.0±0.2	1.0±0.2	1.0±0.1	1.1±0.1	1.0±0.1	1.0±0.1	1.0±0.1
Coefficient A (%)	49.9±17.4	7.4±14.7	28.7±16.2	8.8±13.6	3.6±6.7	6.2±10.8	58.9±14.7	24.7±9.1	41.8±21.2
F.surf.(cm <sup>2</sup> shoot <sup>-1</sup> )	128.4±37.9	81.4±35.3	209.7±36.6	141.8±22.1	164.7±34.8	153.2±30.8	102.4±32.5	117.2±52.1	109.8±43.4
LAI (cm <sup>2</sup> m <sup>-2</sup> )	45.9±16.6	26.8±12.5	36.4±17.4	43.1±12.6	35.7±15.1	39.4±14.5	23.6±9.5	23.8±11.2	23.7±10.2

The lepidochronological analysis of the meadow is summarized in Table 6. The maximum growth of the rhizome was found 7.89 ±0.60 mm for lower limit, 6.82±0.72 mm for reference limit and 6.42 ±0.89 mm for upper limit. According to Pergent (2007) the health of the meadow was found weak.



**Tab. 6: The number of scales per cycle and speed growth of the rhizomes at lower, reference and upper limits in the Dardanos Bay.**

Lepidochronological year	The lower limit		Reference limit (15m)		The upper limit	
	Number of Scales	Growth of rhizomes (mm yr <sup>-1</sup> )	Number of Scales	Growth of rhizomes (mm yr <sup>-1</sup> )	Number of Scales	Growth of rhizomes (mm yr <sup>-1</sup> )
2013	6.81±0.60	3.89±0.93	2.25±0.45	6.10±0.90	3.63±0.50	3.72±0.50
2012	6.06±1.12	5.89±1.69	4.06±0.77	5.71±1.30	3.13±0.34	5.43±0.54
2011	6.25±1.48	7.44±1.81	3.31±0.48	5.34±1.02	3.00±0.00	6.42±0.89
2010	6.31±1.35	7.89±0.60	3.38±0.50	6.82±0.72		
2009	6.50±1.83	7.56±0.88	4.06±0.85	7.63±0.74		
2008	6.75±1.84	7.24±1.34	3.13±0.34	6.04±1.03		

### Discussion

According to our results, *P. oceanica* meadows along the Dardanos Bay, exhibit a weak vitality. The standardized BiPo index was used to evaluate and compare the vitality of the meadows (Bund and Somolini, 2007; Lopez y Rojo *et al.*, 2010). The Ecological Quality Ratio (EQR) was calculated and compared to the standardized scale (Tab.7). The Ecologic Quality Ratios were found between 0.42 to 0.52.

**Tab. 7: Evaluation of Ecological Quality Ratio (EQR) using BiPo index. H=High, G=Good, M=Moderate, P=Poor and B=Bad**

	Lower Limit				Reference Depth (15 m)	
	Depth (m)	Type	Cover (%)	Plagio (%)	Foliar surface (cm <sup>2</sup> shoot <sup>-1</sup> )	Number of Scales
Values	17	M	47.47	51.82	153	3.35
EQR	0.43 (P)	0.53(P)	0.48 (P)	0.52 (P)	0.42 (P)	0.42 (P)

According to standardized values, the vitality of the meadows was determined as “poor”. There are local purse-seiners fishing heavily along Karaliman, Güzelyalı and Dardanos line, during the season. That’s why we couldn't find markers which were located in 2005 and the meadow didn’t get a chance to expand the lower limit due to the stress. We conclude that, the coast guard controls should be increased for this region (Karaliman, Güzelyalı and Dardanos Bay) and the area where the meadow is dense has to be preserved legally. In addition, the citizens of Çanakkale should be informed by scientist about *Posidonia* meadows, a public awareness should be raised.

### Acknowledgements

The study is supported by The Scientific and Technological Research Council of Turkey (TUBITAK) for Graduate Scholarship Program for National Students (2209/A).

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## **MONITORING AND EXPANSION OF POSIDONIA MONITORING NETWORKS ALONG CORSICAN COASTLINE**

### **Abstract**

*The distribution and the vitality of the *Posidonia oceanica* meadow were monitored in the western Mediterranean at 15 sites along the coasts of Corsica (1 000 km of coastline) using two monitoring systems, the *Posidonia* Monitoring Network and SeagrassNet, between 2004 and 2013. While the vitality of the meadow is satisfactory overall, due to the low impact of human pressure along these coasts, patterns of change over time show a slight degradation of the main descriptors of the meadow. The meadow's vitality index has declined on average by 8.1%, the BiPo index by 9.8% and the position of the lower limit has regressed at 6 sites and only progressed at one site; however, the majority remains stable (8 sites). While this pattern of change may be the reflection of local alterations in the environment (increase or decline in human pressure), the regressive dynamic of the meadow observed at the lower limit at several reference sites (e.g. marine protected areas, sites distant from sources of human impact) is more worrying; both monitoring systems showed a significant decline for the common site of Stareso.*

*Two hypotheses might explain the regression observed: (i) the rise in mean sea level during the study period (more than 2 cm), which may result in a significant regression in sectors where the slope is relatively slight, and (ii) the North Atlantic Oscillation (NAO), which declined from 2002 to reach very low values in 2010, resulting in an increase in precipitation (inflow from coastal rivers) and a decline in the amount of light available (increase in cloud cover and inputs of particulate matter).*

**Key-words:** *Posidonia oceanica*, Monitoring Network, Vitality, Mediterranean Sea, Climate Change

### **Introduction**

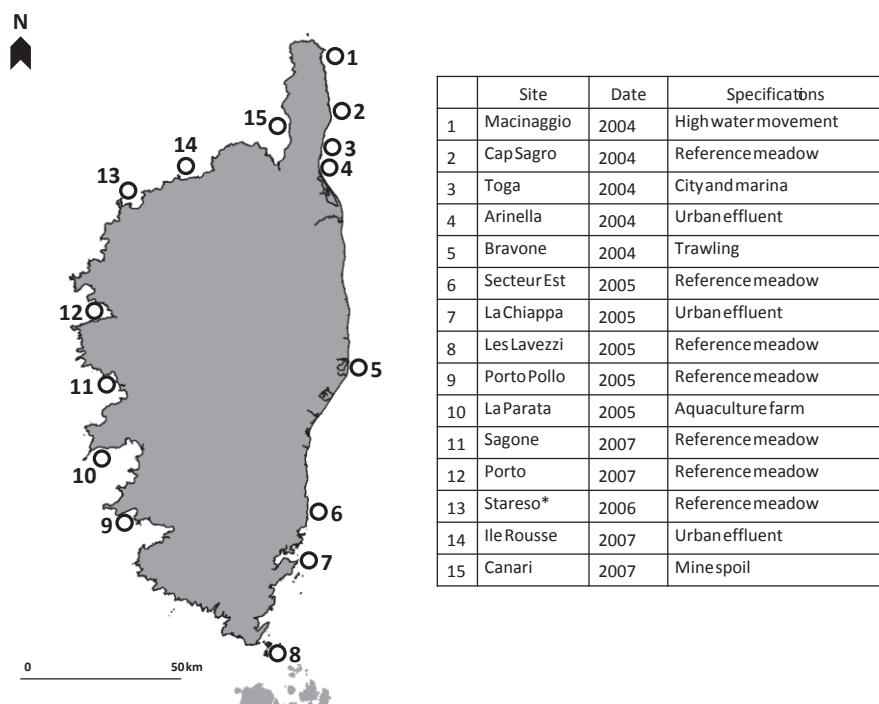
The development of powerful bioindicators has provided a basis for setting up various monitoring systems throughout the world. Because of their ecological roles, their sensitivity to disturbances and their very extensive geographical range, the seagrasses often constitute excellent biological indicators (Pergent-Martini *et al.*, 2005). Several networks based on these species have thus been developed over the past decades in order to investigate the general state of the ecosystem (Short *et al.*, 2006; Lopez y Royo, 2010a). The first network was the *Posidonia* Monitoring Network (PMN) for the Provence - Alpes - Côte d'Azur region, founded in 1984 (Boudouresque *et al.*, 2000). This initiative was extended in 1988 in the Euro-Mediterranean region with Cost 647 (Spain, France, Italy and Greece). In 2001, the first worldwide network, SeagrassNet, was set up in the United States (Short *et al.*, 2002); it now includes 122 sites in 33 countries (two in the Mediterranean, one of them at Calvi in Corsica; <http://www.seagrassnet.org/>).

The development of monitoring networks in the Mediterranean has been strengthened by the adoption, by the contracting parties of the Barcelona Convention, of the Action Plan for the Conservation of Marine Vegetation in the Mediterranean Sea (UNEP-MAP-RAC/SPA, 1999) and of the guidelines for the Standardization of Mapping and

Monitoring Methods for Mediterranean Seagrasses (UNEP-MAP-RAC/SPA, 2011). These networks have generally a two-fold purpose: (i) to monitor the state of conservation of *Posidonia oceanica* meadows, and (ii) to use *Posidonia oceanica* meadows as an indicator of the quality of the environment. The aim of the present work is to present the results of the monitoring activities on the distribution and the vitality of the *Posidonia oceanica* meadow at 15 sites situated at the lower limit along 1 000 km of coastline (Corsica Island, North Western Mediterranean Basin, France) in order to assess the dynamic of the meadows and to determine the possible causes of the changes observed. Developed between 2004 and 2007, this monitoring network is based on the PMN method but one site is also monitored using the SeagrassNet protocol in order to inter-calibrate the two monitoring methods.

### Material and Methods

The 15 sites taken into account correspond to reference sites or sites exposed to significant human and/or natural pressure; the Stareso site is also monitored on the basis of the SeagrassNet network (Fig. 1).



**Fig. 1: Location, specifications and dates of setting up of the sites of Posidonia Monitoring Network (PMN) along the coast of Corsica. \*: SeagrassNet and PMN site.**

For each site, specific measurements were assessed at two depths for the PMN (an intermediate depth around -15 m and the lower limit) and at 3 depths for the SeagrassNet (the upper limit at -8 m, an intermediate depth at -25 m and the lower limit at -37 m). The PMN was set up between 2004 and 2007 and follow-up visits to all sites were carried out between May and July 2013; the SeagrassNet protocol was initiated in October 2006 with follow-ups in October 2009 and October 2012.

The PMN procedure consists in setting up eleven fixed markers every five meters, along the edge of the lower limit of the meadow; the precise position of each marker is then recorded with a compass (Boudouresque *et al.*, 2000). A detailed map of the position of the meadow along the limit is established on the basis of these compass bearings and of

assembled vertical photographs; horizontal photographs of the meadow above its limit were also taken at the level of each marker (Pergent *et al.*, 2005). Several descriptors were also measured, at the intermediate depth and at the lower limit, in order to assess the vitality of the meadow and the quality of the water body (BiPo index in Lopez y Royo *et al.*, 2010a). The data interpretation is based on the standardized scales validated by the Barcelona Convention (UNEP-MAP-RAC/SPA, 2011) and on the Ecological Quality Ratio, calculated in the BiPo index (Lopez y Royo *et al.*, 2010b).

The SeagrassNet protocol requires that two markers be placed at an interval of 50 meters, parallel to the coastline, at the three depths (Short *et al.*, 2006). The *in situ* measurements (cover, density) and sampling (*Posidonia oceanica* shoots) are carried out at 12 precise distances along the transect. Density is measured in 25x25cm quadrats and cover is estimated within 40 cmx40 cm quadrats by means of video images. The 12 shoots collected are divided into three parts: leaf, rhizome (maximum length of 10 cm) and sheaths. The length of the longest leaf gives the canopy height. Each shoot part is then dried and weighed in order to measure the biomass.

## Results

Only three of the 165 PMN markers placed along the lower limit were not recovered in 2013 (less than 2%); these were replaced on the basis of the compass bearings taken when they were initially installed.

The meadow had regressed for 73 markers, remained stable for 63 and it had progressed for 29; while the regressive dynamic is high (44%), the meadow had progressed or remained stable in more than half of the markers. With regard to the sites, the meadow had regressed at six sites, eight had remained stable and one had progressed. The highest regressions observed concerned both the sites with the highest human pressure impact (Arinella and La Parata) and the reference sites (Cap Sagro and Porto Pollo). The only progression was recorded at the Canari site.

The typology of the lower limit remained the same for 8 sites out of 15; in contrast, for the 7 other sites, the differences observed always reflect a degradation of the limits, in particular for the sites of Cap Sagro, Arinella and Porto Pollo, where the lower limit declined from 'sharp high cover limit' to 'regressive limit'.

A similarly regressive dynamic was found for the density recorded above the markers. During the study period, 11 sites out of 15 showed a significant change in this descriptor: nine sites presented a decline in the mean density whereas two presented an increase. The most significant decline concerns the site of Arinella, which dropped from  $130 \pm 21$  to  $41 \pm 7$  shoots per  $m^2$ . Over the whole set of sites, the mean density declined from  $121 \pm 27$  to  $90 \pm 28$  shoots per  $m^2$ , with a decline of 25%.

The mean vitality of the meadow, calculated on the basis of all the eight descriptors measured at the lower limit and at intermediate depth, remained 'good', even if a decline in the mean vitality index is recorded over time ( $3.8 \pm 0.2$  to  $3.5 \pm 0.2$ ) and the number of sites ranked as 'good' declines over course of the study period (Tab. 1). This decline in vitality was observed both at the reference sites (57%) and at the high human pressure impact sites (50%).

The quality of the water body (BiPo Index) declined over the course of the study period ( $0.74 \pm 0.06$  to  $0.67 \pm 0.07$ ) but also remained 'good' at the scale of the Corsican coasts as a whole (Tab. 2). This decline is observed both at the reference sites (86%) and at the high human pressure impact sites (75%).



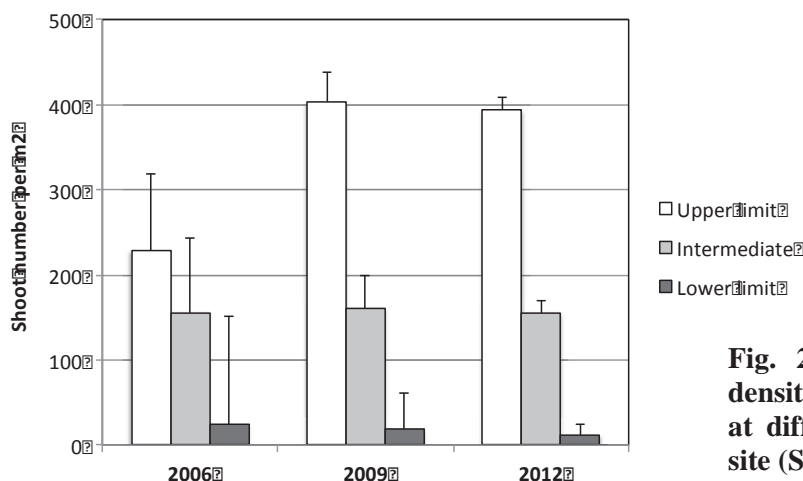
**Tab. 1: Mean vitality of *Posidonia oceanica* meadow at the PMN sites during the setting up (2004 - 2007) and the monitoring phase (2013). (ref): Reference site.**

	2004-2007	2013		2004-2007	2013
Macinaggio	4.1 (Good)	4.1 (Good)	Porto Pollo (ref)	4.0 (Good)	3.1 (Normal)
Cap Sagro (ref)	3.6 (Good)	3.1 (Normal)	La Parata	3.8 (Good)	3.0 (Normal)
Toga	2.9 (Normal)	3.3 (Normal)	Sagone (ref)	4.4 (Good)	3.6 (Good)
Arinella	4.0 (Good)	3.1 (Normal)	Porto (ref)	3.8 (Good)	3.8 (Good)
Bravone	3.9 (Good)	3.4 (Normal)	Stareso (ref)	4.3 (Good)	3.6 (Good)
Secteur Est (ref)	3.5 (Good)	3.5 (Good)	Ile Rousse	3.1 (Normal)	3.1 (Normal)
La Chiappa	3.9 (Good)	3.1 (Normal)	Canari	3.9 (Good)	4.3 (Good)
Lavezzi (ref)	3.9 (Good)	4.1 (Good)			

**Tab. 2: Mean water body quality (BiPo index) at the PMN sites during the setting up (2004 - 2007) and the monitoring phase (2013). (ref): Reference site.**

	2004-2007	2013		2004-2007	2013
Macinaggio	0.89 (High)	0.95 (High)	Porto Pollo (ref)	0.74 (Good)	0.60 (Good)
Cap Sagro (ref)	0.84 (High)	0.69 (Good)	La Parata	0.59 (Good)	0.55 (Good)
Toga	0.52 (Normal)	0.49 (Normal)	Sagone (ref)	0.80 (High)	0.65 (Good)
Arinella	0.75 (Good)	0.61 (Good)	Porto (ref)	0.87 (High)	0.80 (High)
Bravone	0.67 (Good)	0.59 (Good)	Stareso (ref)	0.87 (High)	0.69 (Good)
Secteur Est (ref)	0.77 (Good)	0.66 (Good)	Ile Rousse	0.59 (Good)	0.49 (Normal)
La Chiappa	0.77 (Good)	0.61 (Good)	Canari	0.69 (Good)	0.83 (High)
Lavezzi (ref)	0.76 (Good)	0.82 (High)			

The three transects of the SeagrassNet system (Stareso site) were carried out at each follow-up visit to the site (2009 and 2012). The meadow density declined when the depth increased for the three years studied (Fig. 2).



**Fig. 2: Changes in meadow density between 2006 and 2012 at different depths at Stareso site (SeagrassNet system).**

Between 2006 and 2012, the density increased at the upper limit (-9 m), remained stable at the intermediate depth (-25 m) and declined at the lower limit (-38 m). At the lower limit, it declined from  $24 \pm 16$  to  $12 \pm 13$  shoots per  $m^2$ . The meadow cover also declined at the lower limit, where it dropped respectively from  $23 \pm 10\%$  in 2006, to  $20 \pm 9\%$  in 2009 and to  $19 \pm 13\%$  in 2012.

## Discussion

The protocol used for the PMN along the Corsican coasts provided a basis for (i) validating the sustainability of the structures used (99% still found after 5 to 9 years without maintenance), (ii) comparing with precision the data acquired during the setting up phase (2004-2007) and at the time of the first follow-up visit (2013), (iii) detecting and quantifying the alterations recorded with regard to the lower limit, the vitality of the meadow and the quality of the water body, and (iv) comparing the data collected with those resulting from the SeagrassNet monitoring system, for the Stareso site.

Even if the mean vitality of the *Posidonia oceanica* meadows and the mean quality of the water body along the Corsican coasts are considered as 'good', the patterns of change over time show a general degradation, with a vitality index that has declined on average by 9.8%. However, taking into account changes in (i) the position of the lower limit, (ii) the vitality index and (iii) the BiPo index, the median shows a stability for 9 sites (including 4 reference sites), regression for 5 sites (including 3 reference sites), and progression for 1 site.

The regression of the lower limit of the meadows, especially in the reference sites, cannot be attributed to local degradation of the environmental conditions. In addition, a similar phenomenon was observed for the Provence – Alpes – Côte d'Azur Region PMN (Boudouresque *et al.*, 2000) including Marine Protected Areas (Mayot *et al.*, 2006; Bonhomme *et al.*, 2010).

The lower limit corresponds to the depth of compensation of the species, and any reduction of this light will inevitably entail a decline in the vitality of the meadow and a regression of the position of the lower limit. Two hypotheses might explain the regression observed:

- The mean sea level: Measurements taken in Corsica, at the Ajaccio tidal gauge station, show a mean increase of 2 cm over the past 10 years (SONEL network; <http://www.sonel.org/-Maregraphes-.html>). For the deep lower limits (below -35 m), this increase in the height of the water column over the past decade has resulted in a decline in the amount of light at the seabed. Thus, for a slope of 2%, an increase of 2 cm in the depth of the water column corresponds to a linear regression of 1 m of the position of the lower limit of the seagrass meadow.

- The North Atlantic Oscillation (NAO): The NAO index declined after 2002 to reach very low values in 2010 (Hurrell and National Center for Atmospheric Research Staff, 2013) and these slight values may play a role in the penetration of the light within the water column (increase in inflow from coastal rivers, inputs of particulate matter and cloud cover) at Mediterranean scale (Visbeck *et al.*, 2001).

For the sites exposed to the persistence, or even the amplification, of human pressures, the state of conservation of the meadows continues to decline. This is the case of the sites of Arinella (urban sewage outfall) and La Parata (discharges from an aquaculture facility). In contrast, at sites where these pressures are in decline, the state of conservation of the meadow has stabilized (Toga, Ile Rousse) or progressed (Canari).

The extension of the PMN in Corsica, with the installation of six new reference sites in the "Réserve Naturelle des Bouches de Bonifacio" (south coast), in 2013, should provide the means to monitor more precisely the patterns of change in the seagrass meadow in the sites where human impact is low and confirm the possible impact of global climate change.

## Acknowledgment

This work was supported by a grant from the “Office de l’Environnement de Corse” and SeagrassNet network support.

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## **ARSENIC CONCENTRATIONS IN SEAGRASS AROUND THE MEDITERRANEAN COAST AND SEASONAL VARIATIONS**

### **Abstract**

*Arsenic's occurrence in the environment could be due to human activities as well as to natural sources. In this study, *Posidonia oceanica* and *Cymodocea nodosa* are collected in 84 sites around the Mediterranean basin. In addition, both seagrass are collected monthly, in two sites (Calvi in Corsica and Salammbô in Tunisia). Arsenic concentrations in *C. nodosa* present seasonal variations in relation with spring phytoplankton blooms. For both species arsenic concentration is higher in the vicinity of geological sources (mining), lagoon outlets and industrial activities. Moreover, Mediterranean islands (Balearic, Sardinia, Corsica, Malta, Crete and Cyprus) and the Southern basin coastline exhibit lower concentrations in Arsenic than the rest of the Mediterranean basin. The wide spread distribution of these two species would encourage their use in a global monitoring network devoted to Arsenic contamination.*

**Key-words:** Coastal contamination, *Posidonia oceanica*, *Cymodocea nodosa*, Arsenic, Human-induced pressure.

### **Introduction**

Seagrasses are often used to monitor trace metals, and in the Mediterranean Sea most of these studies focused on *Posidonia oceanica* (L.) Delile meadows, due to their widespread distribution around the Mediterranean Sea and of their ability to accumulate the trace metals (see for instance: Augier *et al.*, 1978; Costantini *et al.*, 1991; Pergent-Martini & Pergent, 2000; Lafabrie *et al.*, 2008; Luy *et al.*, 2012). Metals and metalloids are regarded as serious pollutants of the aquatic environment because of their toxicity, persistence, difficulties in biodegrading and their tendency to concentrate in aquatic organisms (Ikem & Egiebor, 2005 in Luy *et al.*, 2012). Among these metalloids, Arsenic (As) is widely distributed in the biosphere. Its occurrence in the environment could be due to human activities (mining, pesticides, burning of fossil fuel etc.) as well as to natural (geogenic) sources (Kim *et al.*, 2002). As contamination of the environment from both anthropogenic and natural sources has occurred in many parts of the world and is recognized as a global problem (Storelli & Marcotrigiano, 2000).

Only few studies deal with the potential use of the main Mediterranean seagrasses as bio-integrator for As (Grauby *et al.*, 1991; Luy *et al.*, 2012). However these two seagrasses constitute a continuous belt all around the Mediterranean coastlines. Thus, in order to increase the interest in using seagrass in Mediterranean monitoring programs, it would be interesting to confirm their capacities of bio-integrator for this element.

The aim of this study is (i) to assess the potential of *P. oceanica* and of *Cymodocea nodosa* (U.) Aschers. as As bio-integrators, (ii) to determine seasonal variations of As concentrations and (iii) to evaluate the level of As concentration along the Mediterranean coasts.

### Material and methods

*P. oceanica* and *C. nodosa* leaves were collected monthly in two sites (Fig. 1) during the vegetative period; i.e. between March and October in Calvi and between January and December 2007 in Salammbô). Other 82 sites were sampled (between May 2007 and July 2008), 15 sites concerned *C. nodosa* and 68 *P. oceanica* (Tab. 1).

**Tab. 1: Coordinates of sampling sites in decimal degrees (WGS 84). Site number is indicated for *Posidonia oceanica* and followed by c and in bold for *Cymodocea nodosa*.**

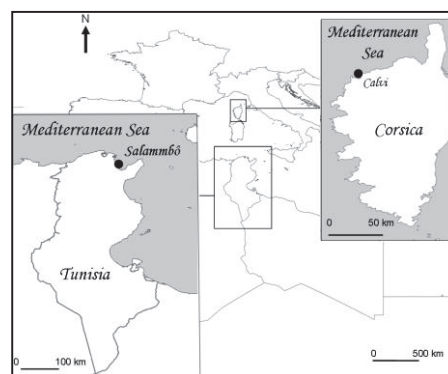
N°	Station	North	East	N°	Station	North	East
ALGERIA							
1	Annaba	36.893456	7.779814				
CROATIA							
2	Brbinjsica	44.055117	14.991983	5	Seget Donji	43.511983	16.213789
3	Island Vlasnik	42.753353	16.796739	6	Zadar	44.103583	15.235861
4	Lavdara	43.948000	15.198533				
CYPRUS							
7	Cape Greco	35.152778	34.173889	8	Larnaca	34.870117	33.655133
EGYPT							
9	Alexandria	31.094064	29.692653				
FRANCE							
10	Arinella	42.666667	9.450000	16	La Parata	41.883333	8.616667
11	Canari	42.783333	9.333333	17	Macinaggio	42.983333	9.483333
12	Cavalaire	43.178500	6.540380	18	Porto	42.250000	8.650000
13	Diane	42.116667	9.566667	19	Porto-Pollo	41.733333	8.766667
14	Ile Rousse	42.633333	8.933333	20	Sagone	42.083333	8.666667
15	La Chiappa	41.583333	9.366667	21	Toga	42.700000	9.450000
GREECE							
22	Crete	35.399658	25.027119	24	Ligaria	35.401069	25.028414
23	Kalogria	40.129519	23.760458				
ITALY							
25	Agropoli	40.375783	15.001250	34	Porto Ercole	42.436717	11.206133
26	Alghero	40.580044	8.309533	35	Porto Torres	40.838728	8.417222
<b>1c</b>	Bari	41.096733	16.964767	36	Santa Marinella	42.040267	11.894617
27	Bosa	40.284808	8.474883	37	Scarrupata	40.697056	13.916522
28	Cagliari 1	39.222444	9.239306	38	Talamone	42.559624	11.157310
29	Cagliari 2	39.207083	9.302083	39	Tavolara	40.849633	9.692300
<b>2c</b>	Civitavecchia	42.141716	11.743600	<b>4c</b>	Torre Canne	40.840417	17.469800
30	Gallipoli	40.057417	17.976183	<b>5c</b>	Torre Lapillo	40.282667	17.842183
31	Lacco Ameno	40.753817	13.891842	40	Torre Mozza	42.946567	10.694533
32	Olbia	41.000444	9.621306	<b>6c</b>	Torre Salinas	39.351722	9.593583
33	Oristano	39.833042	8.553292	41	Torre San Giovanni	39.897350	18.097500
<b>3c</b>	Porto Cesaro	40.257967	17.890850				
LIBYA							
42	Garabouilli	32.814530	13.705220	43	Taiura	32.897528	13.355822
MALTA							
44	Bahar ic-Caghaq	35.953650	14.448467	47	Oalet Marku Bav	35.949267	14.453017
45	Dahlet ix-Xmajjar	35.998983	14.366783	48	Rdum il-Kbir	36.065017	14.296233
46	Marsaxlokk Bav	35.828633	14.547200	49	St Paul's Bav	35.954783	14.392733
SLOVENIA							
50	Gulf of Trieste	45.548683	13.696233				
SPAIN							
51	Alicante	38.251947	0.512400	54	Medes	42.048889	3.219167
52	Chafarinas	35.180006	0.569414	<b>11c</b>	Salines	40.599906	0.711792
<b>7c</b>	Depuradora	40.621092	0.636822	<b>12c</b>	Tancada	40.635772	0.737244
<b>8c</b>	Galatxo	40.587508	0.646989	<b>13c</b>	Torre Sant Joan	40.630797	0.740547
<b>9c</b>	Irta	40.625719	0.665625	<b>55</b>	Torredembarra	41.146983	1.426717
53	Mallorca	39.760083	3.406533	<b>14c</b>	Trabucador	40.618517	0.729028
<b>10c</b>	Mari	40.583769	0.608322				



**Tab. 1 (continued): Coordinates of sampling sites in decimal degrees (WGS 84). Site number is indicated for *Posidonia oceanica* and followed by c and in bold for *Cymodocea nodosa*.**

N°	Station	North	East	N°	Station	North	East
TUNISIA							
56	Kerkenah	34.681514	11.113286	61	Sidi Ali el Mekki	37.172792	10.262000
57	La Galite	37.536801	8.938876	62	Sidi Mechreg	37.164717	9.122424
58	La Goulette	36.813900	10.307603	63	Tabarka	36.954744	8.767150
59	Monastir	35.775128	10.843294	64	Zembra	37.136125	10.808897
60	Rades	36.767925	10.301869				
TURKEY							
65	Gokceada	40.224400	25.867894	67	Turgutlar bay	36.154100	33.445267
66	Mersin	36.790522	34.637447	68- <b>15c</b>	Urla	38.360847	26.794650

The *P. oceanica* and *C. nodosa* leaf blades were cleaned (epiphytes scraped off), rinsed (ultrapure water) and either lyophilised (Heto® FD4-85 freeze dryer, HetoHolten A/S) or dried at 30°C to constant weight, before they were manually reduced to powder. Analyses were run with quality assurance procedures at the Laboratory of Rouen / ETSA (Rouen, France). They were performed using the hydride generation and atomic absorption spectrometry method.

**Fig. 1: Sampling sites of Calvi (Corsica) and Salammbô (Tunisia).**

## Results

Arsenic concentrations in *C. nodosa* were higher than in *P. oceanica* all year round (respectively a mean of  $10.0 \pm 1.3$  and  $0.9 \pm 0.1$   $\mu\text{g.g}^{-1}$  DW in Calvi, and a mean of  $0.6 \pm 0.1$  and  $0.4 \pm 0.0$   $\mu\text{g.g}^{-1}$  DW in Salammbô), with an important difference between the both species, observed in Calvi.

Seasonal variations were observed for *C. nodosa* in the two sites, with a maximum concentration in spring ( $27.8 \pm 3.44$   $\mu\text{g.g}^{-1}$  DW) in Calvi, and at the end of winter ( $1.3 \pm 0.1$   $\mu\text{g.g}^{-1}$  DW) in Salammbô. Two concentration peaks emerge during the sampling period: the first in April and the second in July in Calvi and the first in February and the second in September in Salammbô. A significant correlation (Pearson's correlation test,  $p < 0.05$ ) was observed between Calvi and Salammbô, with a lag of two months (probably due to seasonal gaps between the northern and southern Mediterranean coast).

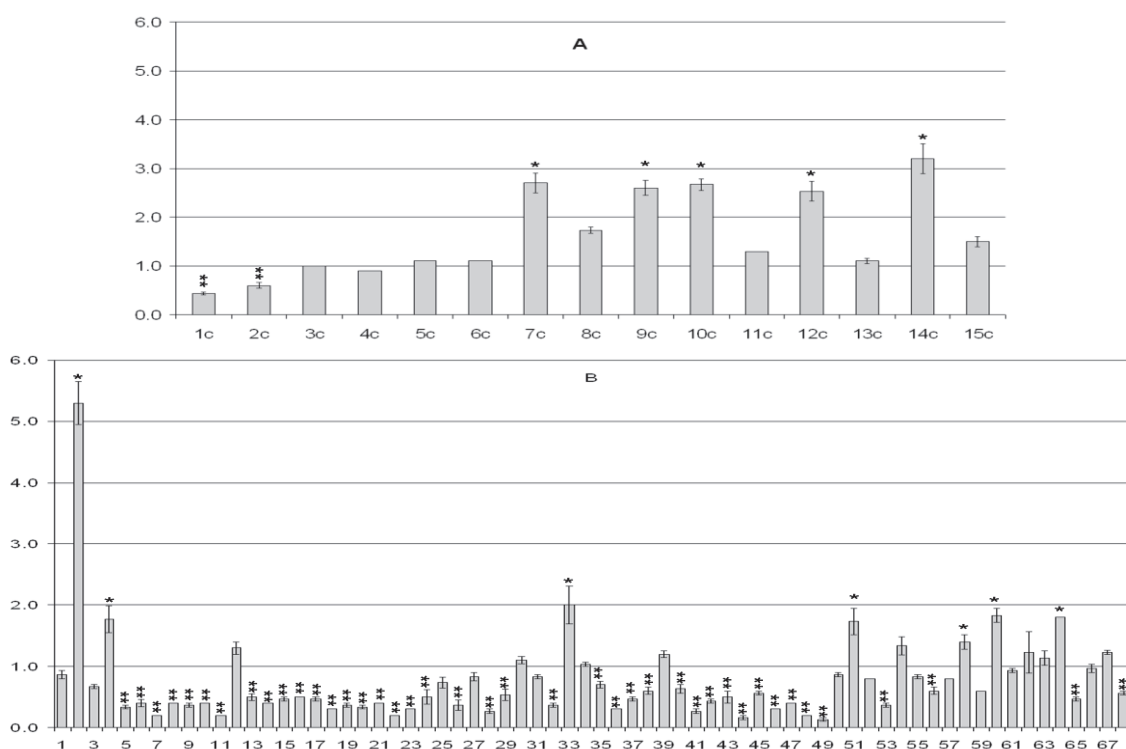
For *P. oceanica*, As concentrations were similar all year round (repeated measures ANOVA,  $p < 0.05$  and Tukey HSD Test) even a maximum is observed in summer in Calvi ( $1.9 \pm 0.1$   $\mu\text{g.g}^{-1}$  DW).

Highest As concentrations were reported in Trabuccador (Spain) for *C. nodosa* (Fig. 2A) and in Brbjinscica (Croatia) for *P. oceanica* (Fig. 2B). For both seagrass, the highest values are significantly different from the lowest values of arsenic concentrations (Nested design ANOVA,  $p < 0.05$ ).

## Discussion

Very few studies concern As concentrations in seagrasses and their seasonal variations (Fourqurean *et al.*, 2007, Malea & Krevekidis, 2013). Nutrients such as phosphorus (P)

are required for growth of phytoplankton and seagrasses (Romero *et al.*, 2006). The chemical similarities between As and P can explain substitutions of these elements. Thus, it has been suggested that indiscriminate uptake of As by marine biota can occur when P concentrations in the water column are depleted (Apte *et al.*, 1986; Michel *et al.*, 1998). In this study, peaks in As occur just after phytoplankton blooms, identified at the end of March and at the beginning of June by Goffart *et al.*, (2002) in Corsica while arsenic concentration peaks occurred in April and July. The uptake of P by phytoplankton, which results in a depletion of this nutrient in the water column, could explain high As concentrations in seagrass during the following months (Howard *et al.*, 1995). This phenomenon appears more visible in *C. nodosa*, perhaps due to the first's higher leaf growth rate (Cancemi *et al.*, 2002) which induces a higher uptake of nutrients.



**Fig. 2: Arsenic concentrations ( $\mu\text{g.g}^{-1}$  DW), in *C. nodosa* (A) and in *P. oceanica* (B) in Mediterranean sites. (Mean  $\pm$  SE). Sites with \* are significantly different from those with \*\*.**

The average concentrations in Calvi (more than 15 times higher than in Salammbô) could be explained by the presence, in proximity of Calvi, of a geological restricted zone rich in As and of a mine containing arsenopyrite (BRGM, 1994).

Concentrations of As, in *P. oceanica* around the western Mediterranean basin, exhibit a significant difference (Kruskal-wallis,  $p < 0.05$ ) between Northern and Southern coasts. The significant higher concentrations observed on northern coast are probably linked to urbanization (UNEP, 2005). Indeed, metal contamination in coastal environments is often associated with impacts induced by human activities. Significant difference was also identified between continental sites and islands (Kruskal-wallis,  $p < 0.05$ ), due to reduced human activities in the Mediterranean islands studied (Biggi, 2001). The highest value, recorded in Brbinjsica (Croatia), confirms high As contamination yet observed in Adriatic Sea (Fattorini *et al.*, 2008).

The highest concentrations of As, in *C. nodosa* ( $> 2.0 \mu\text{g.g}^{-1}$  DW; Fig. 2A), were observed at the outlets of rivers and lagoons. The presence of As in estuaries has already been pointed out due to the accumulation of pollutants, released by upstream industries, and to specific mechanisms of desorption in estuaries (Michel *et al.*, 1998). Conversely, the two sites with lowest values (less than  $0.6 \mu\text{g.g}^{-1}$  DW) were located in open sea (Bari and Civitavecchia).

### Conclusion

Concentrations of As, found in *P. oceanica* do not show important seasonal variations, conversely to *C. nodosa* where these variations seem linked to the availability of nutrients (P in water column); indeed similarities between As and P could induce an increased uptake of this metalloid after phytoplankton blooms.

For both seagrass species concentrations are higher in the vicinity of geological sources, lagoon outlets and in the vicinity of industrial activities. Moreover, Mediterranean islands (Balearic, Sardinia, Corsica, Malta, Crete and Cyprus), as well as the Southern basin coastline, exhibit lower concentrations in As.

The potential of seagrasses as contaminant bio-indicator is confirmed and the widely spread distribution of these two species in the Mediterranean basin would encourage their use in a global monitoring network devoted to As contamination, especially for *P. oceanica* where concentrations are few affected by seasonal variations.

### Aknowledgements

This work was supported by a grant from the “Collectivité Territoriale de Corse” and the collaboration of several Mediterranean laboratories.

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## **ASSESSMENT OF THE CONSERVATION STATUS OF *POSIDONIA OCEANICA* MEADOWS IN THE SAMARIA NATIONAL PARK, AN MPA IN CRETE, GREECE**

### **Abstract**

*Posidonia oceanica* is seagrass endemic to the Mediterranean, and a key species in the coastal zone. It is widely employed as the ideal biological indicator for assessing the quality of water under the Water Framework Directive (WFD) as well as for assessing the health status of coastal ecosystems. To achieve both, it is crucial to develop a network of monitoring stations and cartographic work within the area of interest, in our case Samaria National Park. This is one of the biggest National Parks in Greece, including the well known gorge of Samaria, part of the NATURA 2000 network (SCI: GR4340008) and has an extended coastline of 32 km. The information about the spatial extent of the seagrass meadows from the area is poor, whereas there is no available information about their health status based on specific descriptors. Within a 3-years project, a cost-effective monitoring system has been established aiming to assess the conservation status, identify pressures and threats, and to draw and propose appropriate management measures. Taking into account the limited resources (personnel and funds) we established a network of monitoring stations at two depths (10 and 20 m). There, we monitor a series of descriptors (min - max depth, shoot density, CI index) that can be easily accessed by the staff of the Samaria National Park Managing Authority after minimal training. Moreover, we use low cost acoustic instruments (Lowrance echosounder + Structure Scan) for a complete habitat mapping of the meadows in the previously mapped areas as well as in new ones, after information from local fishermen.

**Key-words:** Seagrass meadow, habitat mapping, Crete, Greece, MPA.

### **Introduction**

For the proper management of the marine resources, essential is the knowledge of its spatial distribution, especially when these are within the limits of a marine protected area. Marine resources are both the species and habitats that occur in the area, reproduce and feed in. The Environment Agency of the European Union has set an encoding for the marine habitats of the European countries starting from the areas of the Natura 2000 network. In total, nine marine habitats have been identified (E. C., 2007), one of which is a priority habitat for the European Union, the seagrass meadows (*Posidonia oceanica*, 1120\*). It is an endemic plant in the Mediterranean basin which forms meadows in the coastal zone, mainly on soft bottoms but also on biogenic patches on hard bottoms. The bathymetric limits of the seagrass meadows starts from 10 cm below the water surface and can be extend deep to 40-45 meters, depending on the water column clarity. It is a plant with tolerance in the variability of the water temperature, the sea currents but very sensitive in the changes of salinity.

*Posidonia oceanica* (L.) Delile, is one of the most important habitats in the Mediterranean Sea, from the biological view, the natural balance as well as the economic point as it offers very high primary production in the marine ecosystem. A big portion of the



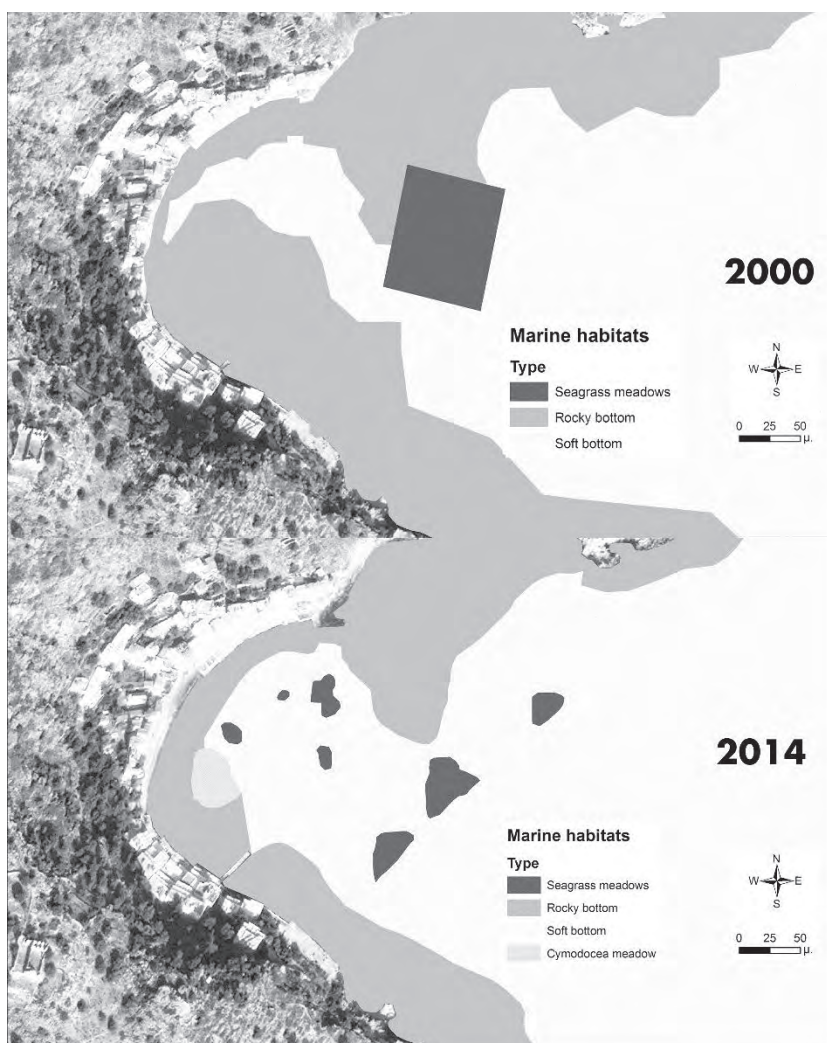
meadows, in the form of dead leaves, provide nutrients in the ecosystem. Meadows act as nursery ground for a lot of fish and crustacean species with commercial value, control the flow of the surface sediments by trapping them between the rhizomes and protect the sand beached from the erosion (Boudouresque *et al.*, 2012). Also, it is a habitat with high diversity as more than 25% of the species that occur in the Mediterranean, can be found in these meadows (Boudouresque *et al.*, 2007). But this habitat is sensitive and from 1960 to day, in several locations in the Mediterranean, the meadows have been downsized spatially or totally disappear mainly due to anthropogenic activities but also due to the changes in the climate (Koutsoubas *et al.*, 2005). Due to the abovementioned characteristics and the pressures of the meadows, a monitoring system has to be established in several areas in order to monitor the status of it, by applying common descriptors (Buia *et al.*, 2004, Pergent *et al.*, 2005) and tools for intercomparison of the results between the sites but also in between the different times of data collection (Pasqualini *et al.*, 2000). Up to now, the only available data for the spatial distribution of the seagrass meadows in the project area, the National Park of Samaria - Leuka Ori (SAC - GR4340008), where from 2001 by the former Ministry of Environment, Physical Planning and Public Works (now Ministry of Environment & Climate change). In the framework of the integrated management of the coastal zone, the spatial distribution of the seagrass meadows will be imprinted by using state-of-the-art tools and methods. Also, the ecological status of the meadows will be identified by implementing descriptors that are available for a pan-mediterranean use and comparison.

### **Material and Methods**

The project area has a total surface of 1470 hectares with a maximum depth of 55 meters and is characterized by an extended rocky shore, with isolated small sandy beaches. The rocky bottom (habitat code 1170) is extended up to 15 meters depth, where the soft bottom ecosystems occur. Deeper than 50 meters, coralligenous formations occur. Based on the previous dataset, oceanographic cruises have been conducted and by using low cost side scan sonar (SSS) (Kaeser *et al.*, 2012) and single beam echosounder in order to collect data for the mapping of the meadows and the associated depths. The cruises have been narrowed only in the areas that the existence of the meadows is confirmed neither from the previous habitat mapping nor from local fishermen. For the rest of the area, very high resolution (VHR) satellite images (World View II from Google Earth) and colored aerial orthophotos have been analyzed by image interpretation as well as by using Remote Sensing tools. When new areas have been identified, new cruises have been organized in order to collect data. SSS data have been used to delineate the meadows below 20 meters as for the rest depths (shallower than 20m.), the VHR image provide us excellent data for an accurate mapping of the meadows in the scale of 1:1000. Same scale has been set also in the analysis of the SSS data. The sonar data have been analyzed by using the Reefmaster software (Reefmaster, 2014), a low cost (200 € with two licenses) software with great capabilities. In the areas where the spatial extend of the meadows has been delineated, several dives have been conducted in order to collect ecological data like the min-max depth, shoot density and Conservation Index data (CI). Two depths have been selected for the collection of the data, 10 & 20 meters. These depths have been selected as are "safe" depths for non scientific divers as the rangers of the management body are certified for SCUBA diving up to 25 meters and are the depths that almost all small meadows have their extend.

## Results

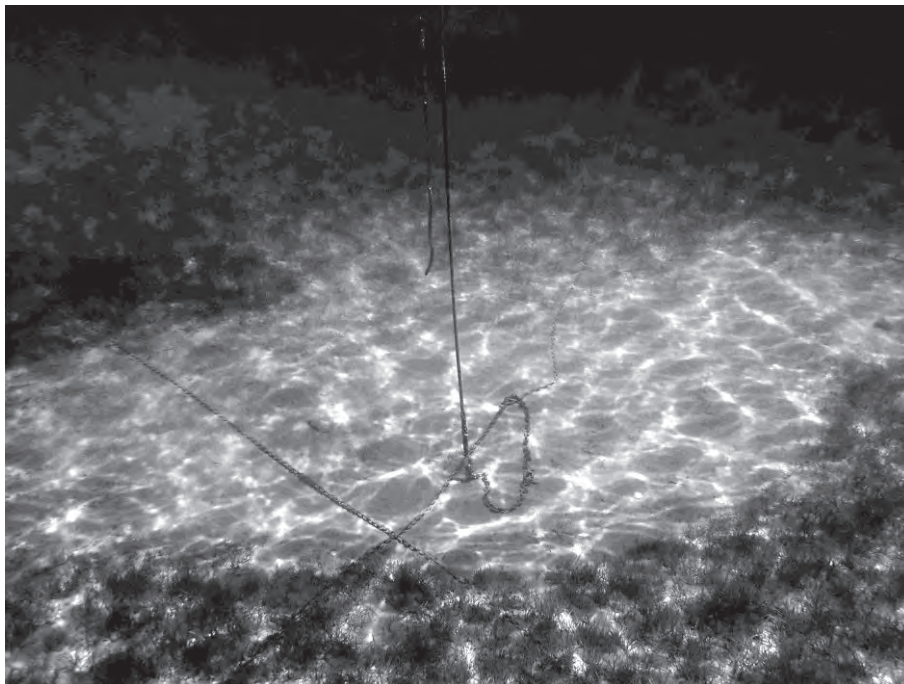
In 2001, the seagrass meadows have a total cover of 16.7 hectares. Up to now and having the 95% of the meadows mapped, we calculate that the total cover is 10.4 hectares. The loss is more than 30% of the initial cover. Meanwhile, a lot of misidentifications of meadows that have been included as meadows but do not exist, have been spotted (Case 1 - Loutro cove, Figure 1). Several small meadows with less than 0.01 hectares in remote areas have been identified, mainly on rocky substrates after the analysis of the VHR satellite images. Some of the already mapped meadows have wrong position or strange shape (exact square shape) and thus these have been remapped.



**Fig. 1. Comparison of the mapping results between 2000 and 2014.**

The occurrence of dead matte in all of the meadows provides us an insight about the past extend of the meadows. The limit of the identified meadows has a sharp end with no small isolated plants deeper than the limits. All meadows are constituted by small patches with small discontinuous areas of sand/gravel. Only two have a continuous extend for more than 50 meters. The shoot density measurements in the predefined depths in all identified meadows reveal values that classify them in the category on normal density (ND) or beds in equilibrium (Buia *et al.*, 2004, Pergent *et al.*, 1995). Conservation Index was not measured in all meadows due to the discontinuous shapes that have. In some of them,

measures have been taken in scale of 10m and the values were between 0.83 and 0.87, classified as medium to normal ecological status according to Montefalcone *et al.* (2006). Pressures from anchoring have been recorded in the locations with high boat traffic (Loutro cove) having as a result the total local destruction of the meadow (Cymodocea meadow) in a radius of five meters, the length of the chain that is used for anchoring in the recorded cases (Fig. 2).



**Fig. 2. Seagrass destruction due to anchorage.**

### **Discussion and Conclusions**

2001 was the year of the marine habitat mapping in all Natura 2000 site of Greece. Due to the widespread and large extend of the sites in the 18.000 km of coastline, several errors occur in this database. Thus, this dataset should not be considered as accurate but as a surrogate of the areas that seagrass meadows exist. The update of this database is imperative by implementing new tools and methods. In our case, the new one should be considered as the baseline data for this protected area. Even if the used descriptors provide us values that state the meadows as good in the terms of the ecological status, the reduce in the spatial extend, the occurrence of *dead matte* in several locations and the limited anthropogenic activities in the coastal zone (apart from the port construction which finish in 2001), show that the reduction is probably due to natural causes. No other evident activities could be accused for the reduction of the meadows. A timeframe comparison of the two datasets should be done by taking into account the different scale of the data (2000 > 1:50.000, 2014 > 1: 1000). The extracted differences will not be as accurate as should be for future use but are an indicator of the spatial extend and its reduction. The recorded dead matte seems that has been formed decades ago but further investigation should be done in order to provide useful informations about the dead frontals and its age. Last decade, more frequent, strong south winds affect several areas in the project area and the combination of them with the sedimentation that cause (Boudouresque *et al.*, 2009), could be a reason for the reduction of the meadows due to sedimentation and destruction of the meadows shallower than 12 meters. In some areas where meadows have dead

matte, the seagrass *Cymodocea nodosa* (Ucria) Ascherson, 1870 form dense meadows, replacing the former inhabitant of the meadow. Also in some areas, the alien seagrass *Halophila stipulacea* (Forsskal, 1775) Ascherson has invaded into the *Cymodocea* meadows, forming dense patches. The use of low cost acoustic tools is an innovation in this project as this approach reduces dramatically the cost of the habitat mapping. The total cost of the equipment and software is less than 1800 €, 8 times less than the cheapest classic side scan sonar and the expensive software for the analysis. We use Lowrance equipment that give us the opportunity to record the acoustic data in XTF format, an open access format for side scan data and is readable from a variety of software. The single beam data can be analyzed in order to extract the bathymetry of the project area by implementing spatial geostatistical analysis and thus the management body can have a high accurate bathymetry dataset, valuable for several projects and management activities. This project has been designed in order to support Management bodies with limited personnel and funding. Also, the personnel is from a variety of sciences, not solely marine biologists, but can be trained and thus to become valuable for the monitoring of the seagrass meadows. The acoustic tools and software as well as the selected descriptors have been used and applied by them on site with success. Training is on hands, on the field and at the office by the expert and the knowledge is transferred smoothly. Next steps for the monitoring include the installation of permanent markers in selected sites, the detailed bathymetry extraction in the selected areas and further and the identification of the reasons for the regression of the meadows and the implementation of actions for the protection before is too late and these disappear.

### **Acknowledgments**

The Project is funded in the framework of Sub-Project 1 "Actions for the Protection and Preservation of Biodiversity" of the Project "PROTECTION AND PRESERVATION OF BIODIVERSITY OF THE NATIONAL PARK OF SAMARIA (WHITE MOUNTAINS)" which is elaborated in the framework of Priority Axis 9 - "Protection of Natural Environment and Biodiversity" of the Operational Programme "Environment and Sustainable Development 2007-2013" and co-financed by Greece and the European Union through the European Regional Development Fund (E.R.D.F.). The ERDF aims to strengthen economic and social cohesion in the European Union by correcting major regional imbalances and participating in the development and restructuring of regions, ensuring in the same time synergy with interventions from other structural funds.

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## **THE CONCEPTUALIZATION OF TRACE ELEMENT FLOWS WITHIN *POSIDONIA OCEANICA* MEADOWS: A COLLABORATIVE PROPOSAL TO FILL KNOWLEDGE GAPS**

### **Abstract**

*The Mediterranean seagrass *Posidonia oceanica* (L.) Delile is regarded as a relevant indicator of bioavailable trace elements (TEs) since it efficiently and proportionally bioaccumulates these chemicals from its surrounding environment. Recent studies have further brought supplementary insights in our understanding of the processes driving TE kinetics within that species. But at a higher degree of organization, the distribution and flows of TEs between *P. oceanica* meadow components, i.e. seagrass shoots, epiphytes, associated algae and animals and detritus, have not been investigated yet. The present study therefore aims to propose a conceptualization model of *P. oceanica* meadows, regarded as the juxtaposition of the 5 separate components listed above exchanging flows of TEs between themselves and with their environment, i.e. water and sediment. This model can be drawn in energy circuit language. By writing such a model, one in essence is writing equations describing a system. To be relevant, its elaboration requires the compilation of a large amount of quantitative data. This conceptualization can thus be regarded as a collaborative innovation requiring scientists to fill the model through the continuous supplement of new data. In fine, this model will lead to the exact quantification of the role played by *P. oceanica* meadows in the coastal biogeochemistry of TEs in the Mediterranean.*

**Key-words:** Mediterranean, seagrass meadow, trace element cycling model, energy circuit language

### **Introduction**

Seagrass meadows are susceptible to the adverse effects of anthropogenic chemicals due to their restricted habitat in shallow, subtidal areas where exposure is greatest to chemicals originating from point and nonpoint sources (Lewis and Devereux, 2009). Among the diversity of chemical toxicants are trace elements (TEs). Their major anthropogenic emission sources result from mining and smelting activities (Callender, 2003); other important sources are linked to industrial, agricultural and urban activities (UNEP/MAP/Blue\_Plan, 2009). As TEs are toxic for aquatic organisms from threshold levels and as they are therefore likely to cause multiple damage at the population, the community and the ecosystem levels (Amiard, 2011), their environmental occurrence has to be accurately monitored.

In the Mediterranean, *P. oceanica* is regarded as a powerful indicator of bioavailable TEs since it highly bioconcentrates these chemicals. Bioaccumulation factor values of *P. oceanica* leaves from seawater ranged between 2 000 and 36 000 for Cd, Cr, Cu, Pb and Zn in 2 Italian uncontaminated areas (Campanella *et al.*, 2001; Conti *et al.*, 2007). *P. oceanica* also accumulates TEs at levels reflecting the status of contamination of its environment (water and sediment), as shown from experiments and field studies (Warnau *et al.*, 1996; Lafabrie *et al.*, 2007). This seagrass is further resistant to pollution and persists in the vicinity of important contamination sources (Boudouresque *et al.*, 2012). In addition, *P. oceanica* ability to record the past levels of TEs, coupled to the dating

possibilities offered by the lepidochronology, provide relevant biological archives about the temporal evolution of the contamination of the Mediterranean littoral (Pergent-Martini, 1998).

If *P. oceanica* usefulness as relevant indicator of the chemical contamination of Mediterranean coastal waters has been widely approved (Pergent-Martini and Pergent, 2000; Richir and Gobert, in press), its ecological and ecotoxicological roles in the global cycling of TEs have, in contrast, been much less documented (but see Sanz-Lázaro *et al.*, 2012, and Richir *et al.*, 2013); and at a higher degree of organization, no global study has so far conceptualized and quantified the distribution and flows of TEs between *P. oceanica* meadow components. In the framework of the STARECAPMED project, the present work therefore aims to propose a model drawn in energy circuit language to better understand the quantitative role played by *P. oceanica* meadows in the coastal biogeochemistry of TEs in the Mediterranean.

### **Model conceptualization**

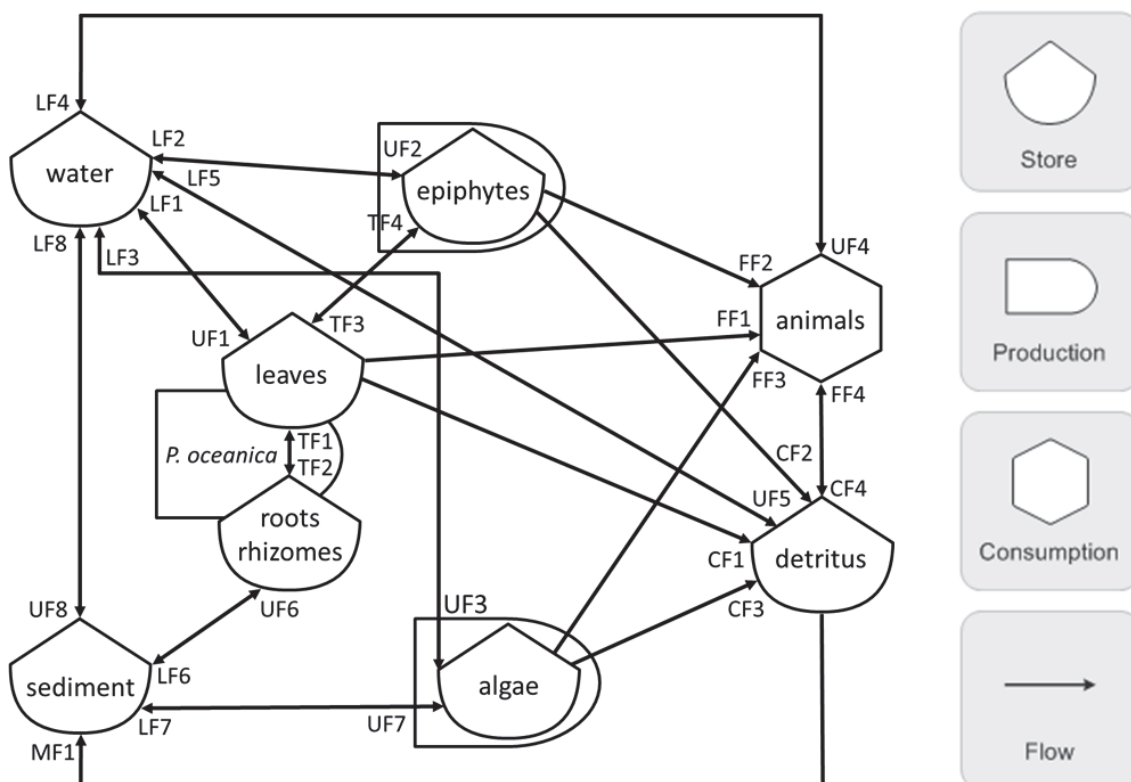
Seagrass meadows can be conceptualized as the juxtaposition of 5 separate components, *i.e.* seagrass shoots, epiphytes, associated algae and animals and detritus, exchanging flows of TEs between themselves and with their environment, *i.e.* water and sediment. This conceptualization can be drawn in energy circuit language (Fig. 1). Each symbol (production, consumption, storage and flow) of the energy circuit language is rigorously and mathematically defined. By writing such a diagram, one, in essence, is writing equations describing a system. Varying the size of symbols further indicates their physical size and their importance (Odum and Odum, 2000; Brown, 2004).

No detailed study has so far modelled the cycling of TEs within *P. oceanica* meadows. Given that the exact quantification of flows and distribution of TEs between *P. oceanica* bed components remains partly poorly known, a similar size was given to symbols modelling their cycling. An ecological relevant size of the symbols could be gained by combining elemental analyses with TE uptake and loss experiments, as did Schroeder and Thorhaug (1980) for a *Thalassia testudinum* community. The model given here for a *P. oceanica* meadow is derived from their work, the only detailed study to our knowledge of the global cycling of TEs within a seagrass ecosystem. Since providing details on all the components of a *P. oceanica* meadow and their interconnecting flows would exceed the requested length for this proceeding, particular attention will therefore be delivered to the processes (no data given here) directly related to the sole seagrass and its associated epiphytes. TE flows discussed in the next section will be referred to their corresponding lettered-numbered label given in Fig. 1 model for clarity purpose.

### **Model implementation to *P. oceanica***

Sanz-Lázaro *et al.* (2012) recently demonstrated the key role played by *P. oceanica* (as a species, not as an ecosystem) in the cycling of TEs in Mediterranean coastal waters. These authors calculated the TE incorporation rates in *P. oceanica* rhizomes, roots and new leaves from mean tissue concentrations and tissue production rates, subtracted TE loss rates through leaf shedding, mechanical breakage (CF1) and grazing (FF1), and extrapolated the balances obtained for their reference meadow to the whole Mediterranean (Tab. 1, according to the estimate of the total cover of 50 000 km<sup>2</sup> of *P. oceanica* meadows; Pasqualini *et al.*, 1998). Depending on the plant compartment where TEs were mainly accumulated and on their incorporation and loss dynamics, Sanz-Lázaro *et al.* (2012) calculated that *P. oceanica* could act either as a sink (positive

balance) or as a source (negative balance; TEs given back accessible to others components of the system or exported) for these chemicals.



**Fig. 1:** Trace element (TE) cycling between the different components of a *P. oceanica* meadow. Model is drawn in energy circuit language (left) and symbol meaning is given (right; Odum and Odum, 2000). In such schematic representations, varying the size of symbols indicates their physical size and their importance (Brown, 2004). Given that the quantification of flows and distribution of TEs between *P. oceanica* bed components remains partly poorly known, a similar size was given to each symbol. The 29 lettered-numbered TE flows between *P. oceanica* bed components are the following: Uptake Flow (UF) leaves (UF1), epiphytes (UF2), algae (UF3) animals (UF4) and detritus (UF5) from water, roots/rhizomes (UF6) and algae (UF7) from sediment, and sediment from water (UF8); Loss Flow (LF) leaves (LF1), epiphytes (LF2), algae (LF3) animals (LF4) and detritus (LF5) to water, roots/rhizomes (LF6) and algae (LF7) to sediment, and sediment to water (LF8). Translocation Flow (TF) roots/rhizomes to leaves (TF1), leaves to roots/rhizomes (TF2), epiphytes to leaves (TF3) and leaves to epiphytes (TF4); Feeding Flow (FF) animals on leaves (FF1), epiphytes (FF2), algae (FF3) and detritus (FF4); Conversion Flow (CF) leaves (CF1), epiphytes (CF2), algae (CF3) and animals (CF4) to detritus; Mineralization Flow (MF) detritus to sediment (MF1). Flow exchanges with adjacent ecosystems are not shown (modified after Schroeder and Thorhaug, 1980).

Seagrass leaves provide an expanded area to sorb and sequester chemicals (UF1, UF2), and their roots-rhizomes system facilitate the absorption and accumulation of sediment contaminants (UF6; Ralph et al., 2006; Lewis and Devereux, 2009). The extent to which these uptake processes are passive or subject to active physiological regulation will determine the final accumulation behaviour of seagrass compartments relative to the TE levels they are exposed to (Schlacher-Hoenlinger and Schlacher, 1998). TEs accumulated by the leaf canopy and the roots-rhizomes system may afterward be translocated to below-

(basipetal, TF2) or above-ground (acropetal, TF1) tissues, respectively (Ralph *et al.*, 2006; Richir *et al.*, 2013). TE concentrations in both above- and below-ground compartments further follow an annual cycle (Malea *et al.*, 1994; Schlacher-Hoenlinger and Schlacher, 1998; Richir and Gobert, in press). This seasonality had been initially only attributed to variations in the plant growth dynamics that induced a dilution of the accumulated TEs (Lyngby and Brix, 1982); but climatic patterns (seasonal rainfalls and storm frequency), leading to changes in chemical loads in the water and sediment, show an equal or greater influence on this seasonality (Schlacher-Hoenlinger and Schlacher, 1998; Prange and Dennison, 2000).

**Tab. 1: Annual balances of the amounts of some trace elements (TEs) in *P. oceanica* (tons y<sup>-1</sup>) for the whole Mediterranean (Sanz-Lázaro *et al.*, 2012). Positive (left) or negative (right) balance values indicate either the incorporation (sink) or the release (source) of TEs by *P. oceanica*, respectively. These balances, expressed in equivalent % to the 2010 TE's mean world production (tons y<sup>-1</sup>; data compiled from the Mineral Yearbooks published by the US Geological Survey on [www.usgs.gov](http://www.usgs.gov)), reflect the quantitative importance of the role played by this species in the cycling of TEs.**

	TE	balance	world prod.	equivalence		TE	balance	world prod.	equivalence
sink	Fe	1 891	2 590 000	0.073 %	source	Pb	-7.9	4 140	0.19 %
	Ni	175	1590	11 %		Cd	-11	21	53 %
	Cr	30	7 290	0.41 %		Co	-21	90	24 %
	As	4.6	52.8	8.7 %		Cu	-45	16 000	0.28 %
	Ag	3.6	23.1	16 %		Zn	-1 459	12 000	12 %

The only few studies that have so far monitored the levels of TEs in *P. oceanica* epiphytes reported a more efficient bioaccumulation of many TEs (*e.g.* V, Cr, Fe, As, Pb etc.) in these latest compared to the other seagrass compartments (Schlacher-Hoenlinger and Schlacher, 1998; Sanz-Lázaro *et al.*, 2012; Richir *et al.*, 2013). In addition to the direct uptake of TEs from the water column (UF2), it has been suggested that their leaching from the seagrass leaves could be an important route of transfert from the plant to its epiphytes (TF4; McRoy and Goering, 1974). Seagrass leaves and more certainly epiphytes (as preferential food resource for numerous grazers; Lepoint *et al.*, 2000) further represent sources of potentially toxic chemicals to the grazer community (FF1, FF2). Since epiphytes are a key component of *P. oceanica* meadows and since they are ubiquitous on seagrass species, they should be taken more into consideration when studying TE cycling in seagrass meadows (Sanz-Lázaro *et al.*, 2012).

Seagrass-accumulated chemicals and those associated with the epiphytic layer can be lost to the surrounding water in a dissolved form (LF1, LF2) or be exported bound to blade fragments (CF1, CF2) at senescence (Lewis and Devereux, 2009). Some TEs like Zn, Cd, Sr or Rb show high release rate through decomposition of *P. oceanica* detritus and are expected to be released in the meadow (LF5); *a contrario*, others like Cs, Tl or Bi show low release rate through decomposition and are more likely to be buried (MF1) or exported to adjacent ecosystems (Sanz-Lázaro *et al.*, 2012). Finally, the well-developed belowground system of *P. oceanica* roots and rhizomes forming mattes can persist for thousands of years (Mateo *et al.*, 1997), thenceforth sequestering a fraction of the accumulated and potentially toxic TEs and reducing the total amount bioavailable to other organisms (Pergent and Pergent-Martini, 1999; Sanz-Lázaro *et al.*, 2012). Seagrasses

therefore act not only as biological filters, but also as storage compartments, thereby favouring the decrease of environmental toxic substances (Kaldy, 2006).

### Conclusion

This work aimed to propose a model conceptualized in energy circuit language to quantify the cycling of TEs within *P. oceanica* meadows. The present synthetic discussion of the current knowledge related to the sole seagrass and its epiphytes demonstrates that the compilation of previously published researches can give a first estimation of TE distribution between *P. oceanica* compartments. A similar approach applied to the entire model will lead to the consecutive creation of a summary database and will highlight which components request further investigations to properly fill the model. Much less studies have however experimentally quantified TE uptake and loss flows from and to the environment and their subsequent redistribution processes between *P. oceanica* compartments and the seagrass meadow components. Since experimental designs may strongly differ between studies, consensual protocols must be defined to efficiently fill the model with accurate flows. *In fine*, this model will lead to the exact quantification of the role played by *P. oceanica* meadows in the coastal biogeochemistry of TEs in the Mediterranean.

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## **DISTRIBUTION AND GENETIC VARIATION OF TWO BIOCONSTRUCTOR CORALLINE ALGAE (*LITHOPHYLLUM BYSSOIDES* (LAMARCK) FOSLIE AND *L. STICTAEFORME* (ARESCHOUG) HAUCK) ALONG THE ITALIAN COASTS**

### **Abstract**

*Coralline red algae represent a worldwide component of hard-bottom coastal communities, where they play a key role in many ecological processes. Some species are ecosystem engineers that produce biogenic platforms, reefs and other calcified structures providing a wealth of habitats for many other species, especially in temperate seas. In the Mediterranean Sea these bioconstructions are widespread and occur both in the littoral zone (trottoirs of *Lithophyllum byssoides*) and in the sublittoral zone (coralligenous bottoms, where coralline algae are one of the main constituents). A detailed knowledge of their distribution and composition is essential for their conservation, but molecular data assessing taxonomic identity and population structure in corallines are extremely limited. The distribution and genetic variation in two important bioconstructor corallines, *Lithophyllum byssoides* and *L. stictaeforme* (Corallinales, Corallinaceae) are investigated using sequences of the plastid *psbA* gene and mitochondrial *cox2,3* spacer. Populations of *L. byssoides* occur on all parts of the Italian coastline where rocky shores exist and the molecular data show that Mediterranean populations of this species represent a sister lineage to Atlantic populations. *L. stictaeforme* is widespread along the Italian shores, particularly along the Ligurian and Tyrrhenian sides. In molecular phylogenies Mediterranean populations of *L. stictaeforme* form a well-supported clade, to which North Atlantic *Lithophyllum* spp. are the closest relatives. Overall the results indicate a high genetic variability in these species, with the possible existence of cryptic species.*

**Key-words:** Bioconstructions, coralline algae, Italy, *Lithophyllum*, molecular phylogeny.

### **Introduction**

The Mediterranean Sea is the largest (2,969,000 km<sup>2</sup>) and deepest (average 1,460 m, maximum 5,267 m) enclosed sea on Earth (Coll *et al.*, 2010). Its geological and hydrological diversity, determined by the complex geological history of this basin from the Mesozoic era (250 mya), has favoured the establishment of a large biological diversity, with approximately 17,000 species recorded (Coll *et al.*, 2010).

Marine bioconstructors are algal or animal species producing rigid structures (usually calcified bodies, tubes or shells) that, accumulating over time, form new substrata. The accumulation of these structures modifies the features of the bottom and produces new surface available for the settlement of other benthic organisms. The surface of the substratum is formed by living organisms and resides above multiple layers of dead biomass, which is usually colonized by boring animals. Bioconstructors are therefore habitat-building organisms and contribute substantially to the high biodiversity of the environments where they occur. Although bioconstructor species exist in every sea, in the

Mediterranean they play a crucial role, because substrata produced by biological activity reach here a spatial and bathymetric extent unknown in other seas.

Although numerous species of seaweeds and animals are able to act as habitat builders, coralline red algae (orders Corallinales and Sporolithales) are the uncontested leaders of temperate marine bioconstruction. These organisms occur in every coastal ecosystem where stable surfaces are present and contribute significantly to the formation and consolidation of hard substrata. In the Mediterranean, encrusting corallines are the builders of two key habitats. Coralligenous concretions, which occupy large portions of the deep infralittoral and circalittoral zone, are produced by growth of encrusting corallines and other red algae (*Lithophyllum stictaeforme*, *Mesophyllum alternans*, *Neogoniolithon mamillosum*, *Peyssonnelia rosa-marina*) (Ballesteros, 2006). *Lithophyllum byssoides* a species typical of the mid littoral zone, forms hemispherical bulky thalli that in optimal conditions coalesce and grow producing rims up to 3 m thick called “trottoirs” (Verlaque 2010). These habitats are now threatened by multiple anthropic stressors (Piazzi *et al.*, 2012; Ponti *et al.*, 2014). A detailed knowledge of the taxonomic circumscription, genetic variation and population connectivity of the bioconstructor species that produce them is essential for their management, but unfortunately this information is currently lacking. In general, the body of molecular data available for coralline algae is still relatively limited and the most important studies on this topic have been published in recent years (e.g., Broom *et al.*, 2008; Bittner *et al.*, 2011). As part of a project focused on the bioconstructions of Italian shores, we are currently investigating the distribution and genetic structure of *Lithophyllum byssoides* and *Lithophyllum stictaeforme* along the coasts of Italy. We present here some results based on analysis of sequences of the plastid *psbA* gene and mitochondrial *cox2,3* spacer. These results will be integrated by additional data to be presented in future studies, for which the work is currently in progress.

### Materials and methods

Samples of *Lithophyllum byssoides* and *Lithophyllum stictaeforme* were collected by snorkelling or SCUBA diving as follows: *L. byssoides*: Gallinara Island, Italy, 13/06/2013 (34 thalli); *L. stictaeforme*: Gallinara Island, Italy, 14 June 2013 (4 thalli); Portofino, Italy, 28 June 2013 (28 thalli); Trezze of Gulf of Trieste, Italy, 5 October 2013; Voula Beach, Athens, Greece, 11 September 2011 (2 thalli). The samples were air-dried and subsequently placed in bags with silica gel for permanent storage. DNA was extracted following the modified protocol of the QIAGEN DNeasy Blood & Tissue Kit ® (QIAGEN, Crawley, U.K.) by Broom *et al.* (2008). PCR amplification of the *psbA* gene was performed following the protocol of Bittner *et al.* (2011). PCR amplification of the *cox2,3* spacer was performed following the protocol of Hernandez-Kantun *et al.* (in press, a). PCR products were visualized and quantified in 1.5% agarose gels stained with GelRed™ (Biotium, Hayward, CA, USA) using Low DNA Mass Ladder (Invitrogen, Carlsbad, CA, USA) as reference under UV. The products of successful reactions with expected length and yield were purified and sequenced commercially (Macrogen, Amsterdam, The Netherlands).

Phylogenetic analyses were performed separately on a *psbA* dataset (844 bp) and on a *cox2,3* dataset (326 bp) formed by sequences of Corallinaceae belonging to the subfamily Lithophylloideae. Besides new sequences generated in this study, the *psbA* dataset consisted of sequences retrieved from GenBank and sequences from Hernandez-Kantun *et al.* (in press, b); *Spongites yendoi* DQ167869 and *Hydrolithon onkodes* AB576036 were the outgroup taxa. The *psbA* alignment did not contain any gaps and its correctness was

checked verifying that no stop codons occurred in the protein translation. The *cox2,3* dataset was obtained by adding the new sequences to the dataset of Hernandez-Kantun *et al.* (in press, a), in which *Lithophorella* sp. KJ801362 was the outgroup taxon. The two datasets were aligned using Clustalw2 in Mega version 5. Neighbour Joining (NJ) distance analyses were performed also in Mega version 5 using uncorrected p-distances. Maximum Likelihood (ML) analyses were performed using RAxML 1.3 (Mac version) with the GTR (General Time Reversible) model with gamma distribution and invariant sites. Statistical support was estimated by Bootstrap (BP) analyses with 1000 resamplings.

## Results

Thalli of *Lithophyllum byssoides* were collected in the mid littoral zone on the northeastern side of Gallinara Island, where this species occurred as numerous hemispherical thalli, 5-10 cm wide, and did not produce well-developed trottoirs. *Lithophyllum stictaeforme* was common in the infralittoral and circalittoral zones at the sites surveyed, ranging in depth from 20 to 50 m.

In the *psbA* phylogeny, *Lithophyllum byssoides* formed a clade with robust BP support, in which two well-supported sister lineages reflected a biogeographical separation between Atlantic and Mediterranean (Fig. 1). The Atlantic lineage was formed by two samples from Cadiz and La Coruña (Spain), whereas the Mediterranean included two samples from Gallinara and the GenBank sequence GQ917720 (obtained from a sample from Banyuls-sur-Mer, Mediterranean France). The highest uncorrected pairwise divergence was 4.38% (Gallinara 15 vs. La Coruña JQ896251).

In the *psbA* phylogeny, *Lithophyllum stictaeforme* formed a well-supported monophyletic group (Fig. 1), which belonged to a larger clade including also samples of *Lithophyllum* from Atlantic Europe and the Mediterranean. A biogeographical separation was evident within the *L. stictaeforme* group: a well-supported lineage consisted of samples from the western Mediterranean (Gallinara and Portofino); sister to it was a lineage of eastern Mediterranean samples (Gulf of Trieste and a sample from Voula Beach, Athens, Greece). The highest pairwise divergence within *L. stictaeforme* was 4.2% (Portofino/Gallinara vs. Greece). GenBank sequences of *L. stictaeforme* from other geographical regions (FJ361442, New Zealand) were not closely related to Mediterranean *L. stictaeforme* and should be referred to a different species.

The *cox2,3* analyses yielded results in agreement with the *psbA* results (Fig. 2). Again, both *L. byssoides* and *L. stictaeforme* formed well-supported lineages. For *L. byssoides*, samples from Gallinara formed a sister lineage to a sample from Atlantic Spain (pairwise divergence: 22.49%). *L. stictaeforme* was sister to a group of *Lithophyllum* samples from Atlantic Europe.

## Discussion

*Lithophyllum byssoides* is a well-defined species, characterized by its superficial anastomosed lamellae, slightly protruding conceptacles and mid littoral habitat. Its morphology is homogeneous throughout its geographical range and does not reflect the clear genetic differentiation between Atlantic and Mediterranean specimens unravelled by our analyses. The unexpectedly high divergence in the *psbA* sequences suggests that the recognition of the Mediterranean and Atlantic lineages as different species may be an appropriate solution, but this requires corroboration based on additional samples from other regions. If this solution is adopted, the correct identity of *L. byssoides* would become a major taxonomic problem because the type locality is dubious (supposedly the English Channel, where this species, however, does not occur). *Lithophyllum stictaeforme* is



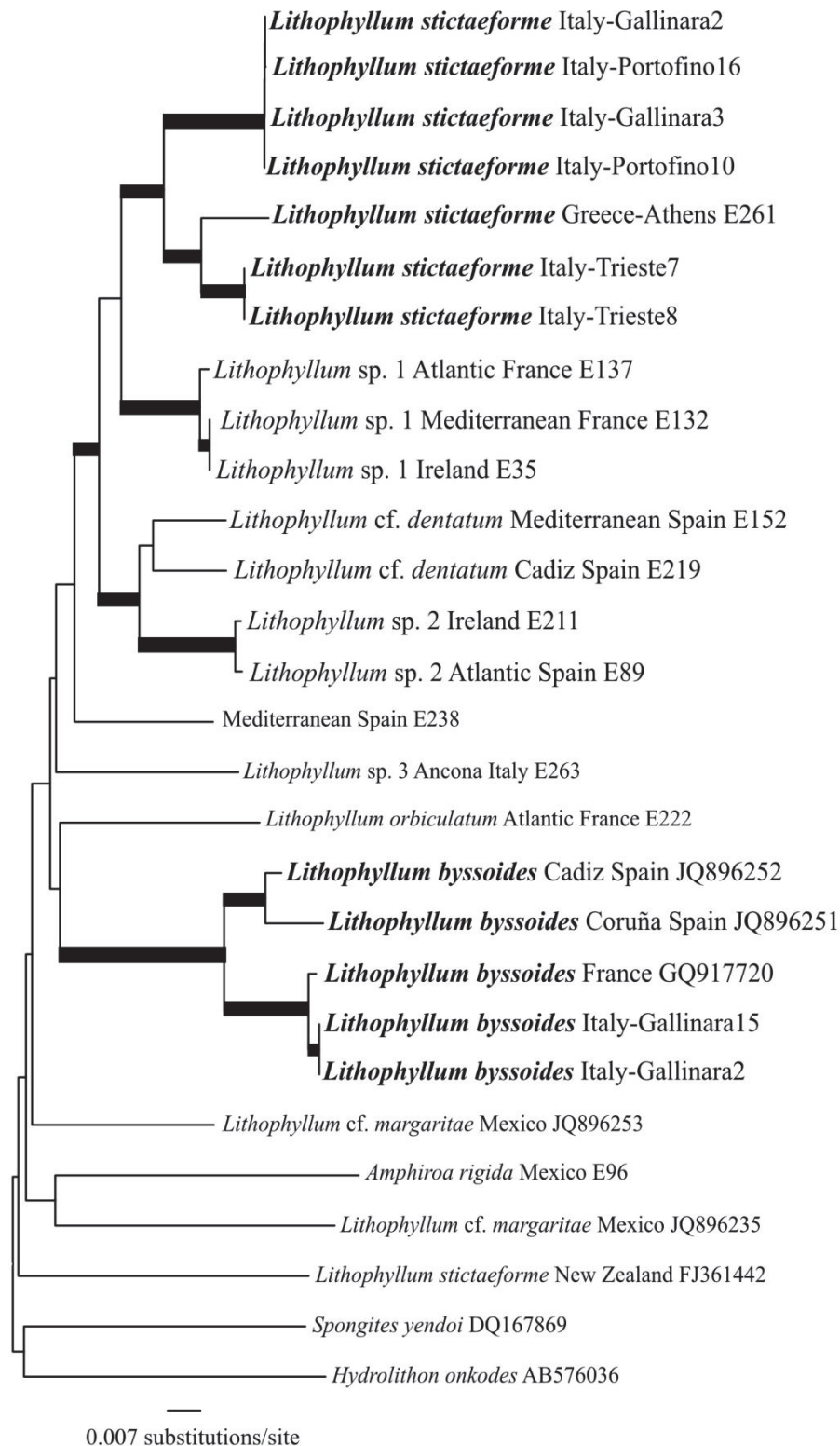
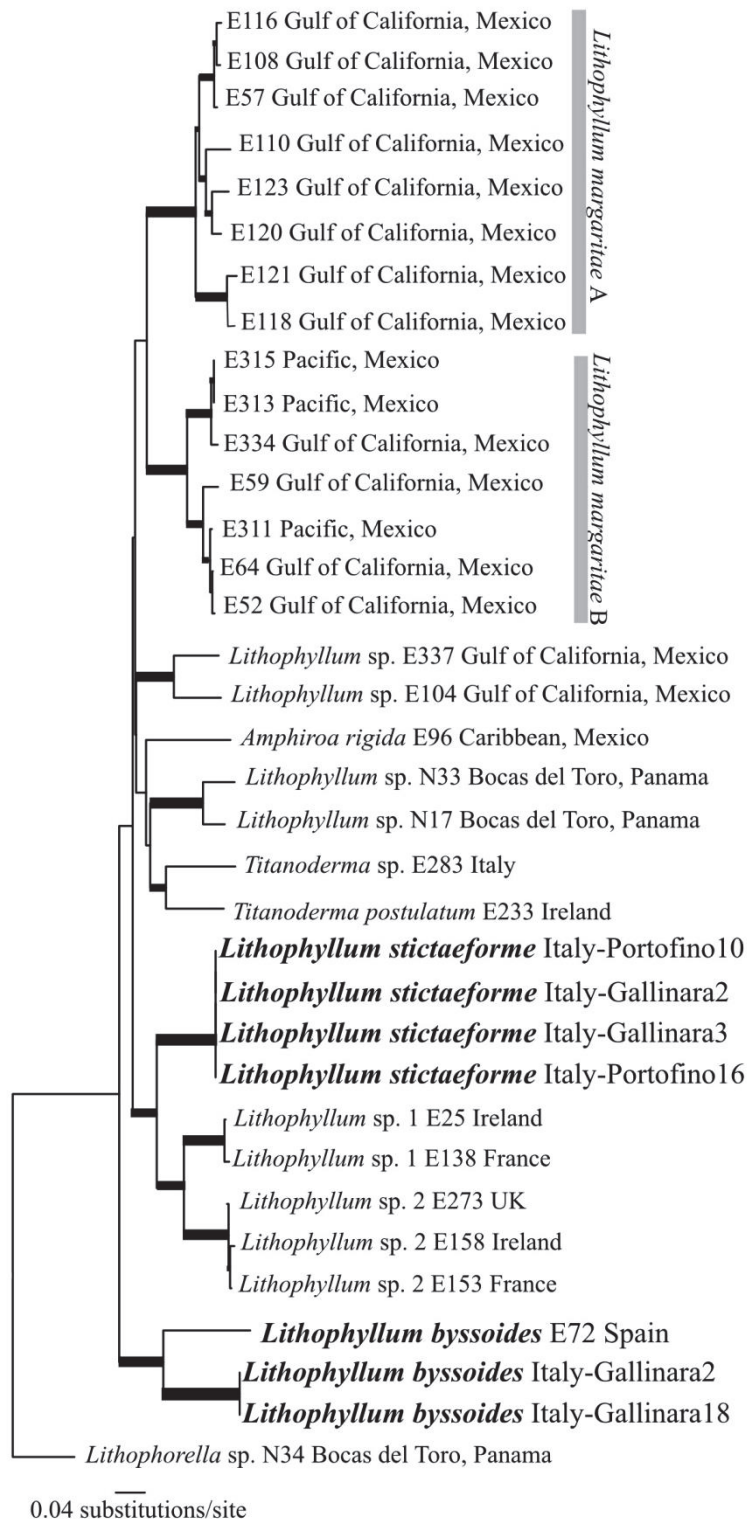


Fig. 1: Phylogeny of the *psbA* gene in the subclass Lithophylloideae inferred by NJ analysis. Sequences of *Lithophyllum byssoides* and *L. stictaeforme* are indicated in bold. Thick lines indicate branches receiving high support in bootstrap analyses (>80% BP for both ML and NJ).





**Fig. 2:** Phylogeny of the *cox2,3* in the subclass Lithophylloideae inferred by NJ analysis. Sequences of *Lithophyllum byssoides* and *L. stictaeforme* are indicated in bold. Thick lines indicate branches receiving high support in bootstrap analyses (>80% BP for both ML and NJ).

widespread in the Mediterranean Sea, but its taxonomic status is not settled (there is no general agreement whether *Lithophyllum cabiochae* should be considered a synonym or a separate species from it). Even for this species our analyses indicate a possible biogeographic separation, in this case between western and eastern Mediterranean populations. Overall, our results show a high genetic variation in *L. byssoides* and *L. stictaeforme*, and suggest that geographic distributions reflect molecular phylogenetic patterns better than morphological data. Recent studies for other coralline algae are delineating a similar scenario (Hernandez-Kantun *et al.*, in press a, in press b).

Again, the addition of samples from other geographical regions will be essential to obtain an accurate taxonomic circumscription and a detailed assessment of the phylogeography of these two important bioconstructor species. We are currently processing collections obtained from other sites of the Italian coasts, which will allow to draw stronger conclusions in forthcoming studies.

### Acknowledgements

The study was supported by a grant of M.I.U.R. (Italian Ministry for Education, Universities and Research) through a P.R.I.N. 2011 Project (Coastal bioconstructions: structure, function and management).

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## MONITORING OF *POSIDONIA* MEADOWS UNDER THE EC HABITATS DIRECTIVE: VEHICULAR VIDEOGRAPHY CAN ESTIMATE TRENDS IN COVERAGE AT LOW COST AND HIGH PRECISION

### Abstract

Regular monitoring of *Posidonia oceanica* (L.) (Posidoniaceae) is required under EC directives, and its purpose is to inform management agencies when and where actions are necessary to protect and restore ecological health and water quality. The Habitats Directive requires each EU member nation to report specifically on temporal trend in areal extent of *Posidonia* habitat. The success of the HD hinges on the power of the monitoring protocol to reveal a significant negative trend, wherever that trend occurs in nature. Statistical power is determined by the size of the sampling error of the protocol, which includes measurement uncertainty and natural spatial variation in the descriptor. We review several descriptors of *Posidonia* status, and consider published estimates of their typical sampling errors, to compare their ability to demonstrate a 10% temporal trend. We find that the protocols with potentially high statistical power are fixed-plot methods such as SeagrassNet and balisage, high resolution aerial photography, and remote underwater videography. The lowest statistical power is found for random plot protocols using descriptors from the interior meadow such as shoot density measured by SCUBA in 40 x 40 cm quadrats. The main advantage of videography over direct SCUBA methods is the ability to sample 20-30 times the area sampled by the equivalent SCUBA labour, but with none of the fatigue or discomfort. The main advantage of videography over aerial photography or acoustic imaging is its high resolution and near-perfect habitat classification accuracy for live seagrass, dead matte, and non-seagrass benthic habitats. We recommend remote underwater videography as a low-cost method of uncovering trends in *Posidonia* cover with high statistical power.

**Key-words:** *Posidonia oceanica*, monitoring, Habitats Directive

### Introduction

The Habitats Directive (HD) of the European Commission (EC) requires all Member States to develop a program for the regular monitoring of *Posidonia oceanica* habitat. The purpose of this requirement is to trigger timely recovery actions in the case of loss or degradation of the habitat during the previous observation period. The success of the HD therefore requires each Member State to develop a monitoring protocol for *Posidonia* that is statistically capable of detecting small declines in conservation status. If the monitoring protocol used by a Member State is not capable of detecting small declines, then the habitat and its quality can decrease undetected, and therefore without the recognition that recovery actions are necessary. A statistically powerful monitoring protocol therefore requires a low sampling error per unit effort. A high sampling error can overwhelm and hide any decline in the descriptor, rendering the protocol powerless and its cost and effort wasted. The HD contains two classes of descriptors, 1) surface area of habitat/range, which is explicit and required by the HD for all priority habitats, and 2) some descriptor(s) of structure/function within the habitat, which are left unspecified. Here we review the literature to obtain sampling error of descriptors of *Posidonia*

conservation status, and compare the resulting statistical power obtained from their use in monitoring *Posidonia*.

## Methods

The Web of Science database was used to find published papers that use acoustic, balisage, aerial or satellite imagery, shoot density, or videography in monitoring of *Posidonia oceanica*, and the sampling errors of these descriptors were extracted from the presented data, and also calculated from theory. Sampling errors originated either as 1) natural spatial variability across the sampling region, and 2) errors inherent in the sampling equipment or methodology. From this precision for each of the five monitoring protocols, the resulting minimum sample size necessary per monitoring event to detect a loss of 10% was calculated. This minimum sample size assumes a 2-tailed t-test with conventional Type I error of 0.05 and Type II error of 0.2. For fixed-plot methods (balisage and SeagrassNet), the t-test is assumed to be one-sample (using differences from paired measurements), and for all other methods a two-sample t-test is assumed.

**Tab. 1: References describing versions of indicated monitoring methods used in this work.**

Method	Reference
Balisage	Boudouresque <i>et al.</i> 2007
SeagrassNet	Short <i>et al.</i> 2006
Remote videography	Puhr <i>et al.</i> 2014
Acoustic single beam	Puhr <i>et al.</i> 2014
Acoustic sidescan	Montefalcone <i>et al.</i> 2013
Acoustic multibeam	De Falco <i>et al.</i> 2010
Remote sensing	Borfecchia <i>et al.</i> 2013; Bakran-Petricioli <i>et al.</i> 2006
Aerial imagery	Puhr <i>et al.</i> 2014

For each method we assume the monitoring site is a segment of shoreline 1 km in length, and the seagrass bed is a fringing bed of moderate patchiness, with approximately 50% cover overall across the entire site within the typical depth limits of *Posidonia oceanica*.

## Results

Acoustic modeling (sidescan, multibeam, single-beam) mapping. Several studies have shown that the classification error for live *Posidonia* during modeling of the above acoustic data can vary tremendously depending on bottom conditions, from approximately 10% to 100% (e.g. Montefalcone *et al.* 2013). We have chosen 20% as an optimistic intermediate value, as found recently for the distinction between live *Posidonia* and macroalgae on sediment (e.g. Puhr *et al.* 2013). We assume an additional 0.2 coefficient of variation (CV, the ratio of standard deviation in seagrass proportion cover to mean proportion cover) due to natural spatial variation among acoustic transects within the sampled site, for a total of 0.3 as the total CV in seagrass cover across spatial transects.

Aerial and remote-sensing imagery. We assume the classification error to be approximately 6% for *Posidonia* to a depth of 4 m using high-resolution aerial imagery, and 25% for aerial or satellite imagery to a depth of 12.5 m (Puhr *et al.* 2014). We assume no natural variability for this method within the local site, because a single image encompasses an entire sampled field site.

Random shoot density quadrats. Shoot density has high sampling error due to both method error (depth estimation by SCUBA wrist gauge) and extremely high natural variability at the scale of sampling, a 40 x 40 cm quadrat (Pergent *et al.* 1995; Balestri *et*

*al.*, 2003). In a patchy meadow with 50% cover, as commonly occurs at depths below the optimum, the minimum CV mathematically is 1. In reality the CV will often be much higher due to variance in shoot density within patches.

Video transects. This method uses video or still footage along a sampled transect, in which the camera is deployed by a vehicle: AUV, ROV, or boat-towed using a towfish or sled, and the camera is precisely geopositioned using surface DGPS and/or underwater telemetry. We do not consider diver-deployed videography, because vehicular deployment allows coverage of 20 - 30 times the sea bottom per unit effort that is possible using SCUBA (Puhr *et al.* 2014), and precise geopositioning is impossible. In this method, used in a random-plot design, we assume negligible classification error (over 98% inter-rater agreement on cover, Puhr *et al.* 2014), and all sampling error is natural spatial variation. Spatial variation from this method is substantially lower than that for random shoot density quadrats, because a transect can be considered the sum of a large number of independent point samples from the same distribution; hence by the central limit theorem, the sampling error can be made arbitrarily low by increasing the length of the transect. For transects perpendicular to shore covering both upper and lower margins of the meadow, we assume a coefficient of variation from spatial variability among transects equal to 0.2. If the design is fixed plot, and transects are revisited (by DGPS navigation), the spatial error in the difference in cover between monitoring events is likely to be much smaller, but revisitation error will then introduce a small method error.

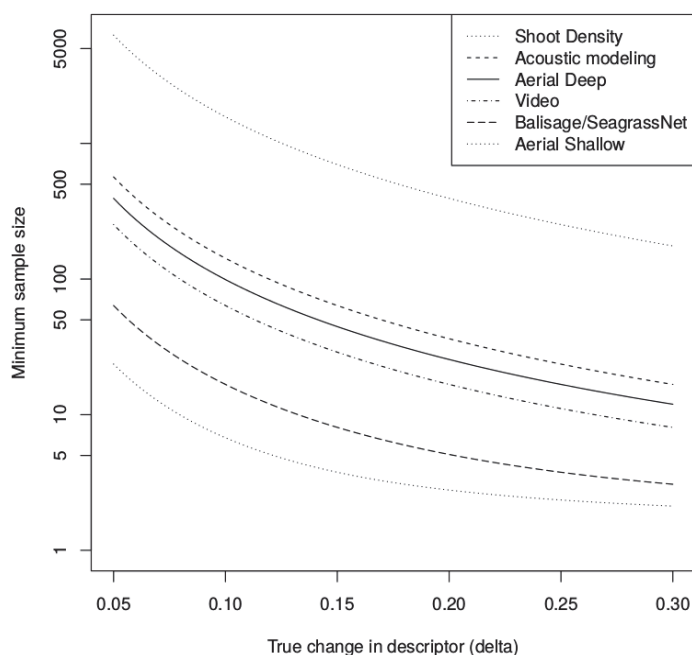
Fixed-plot methods. The major advantage of balisage, SeagrassNet, and acoustic telemetry is that they are fixed-plot: sampling locations are fixed in space by reference to permanent markers (concrete blocks or screw anchors) placed at intervals along the margin of the seagrass bed or in the interior of the bed. Thus, very small changes in the location of the bed margin and other descriptors can be documented at each marker (Sandulli *et al.* 1998; Borg *et al.* 2006; Boudouresque *et al.* 2007 and 2012). For a mean change in margin position of about 10%, expressed as a loss of 20% at half the permanent markers, the resulting coefficient of variation in the change of margin position is exactly 0.1.

Minimum sample sizes. Figure 1 shows the minimum number of independent samples necessary to test the statistical two-tailed null hypothesis of no change in the descriptor between two monitoring events, assuming a Type I error of 0.05, a Type II error of 0.2, sampling coefficients of variation as assumed in the text above, and a true difference ( $\delta$ ) in the descriptor relative to the mean as shown on the horizontal axis. Note that in the balisage and SeagrassNet methods the sampling design is assumed to be fixed-plot, and all others are random-plot designs. These other methods can be converted to an approximately fixed plot design by precise DGPS navigational revisitation of previous sampling transects. This will introduce an error in revisitation; however, it can greatly reduce the overall sampling error if the revisitation error is small relative to the region sampled, which is true for all methods except for the shoot density method.

## Discussion

To satisfy the HD, the descriptor must 1) provide an estimate of the total surface area occupied by *Posidonia* habitat within an EU Member State at the time of reporting; 2) provide an estimate of the temporal trend of that surface area during the previous monitoring period; and 3) provide an estimate of the overall conservation status of *Posidonia* habitat at the time of reporting.





**Fig. 1. Minimum sample sizes to demonstrate the indicated decline (delta) at a power of 80%, for each indicated monitoring method. Sample units are 40 x 40 cm quadrats for shoot density, a single field transect for acoustic or videographic methods, a single image for aerial deep or shallow photography, and a single marked plot or marked transect for balisage/SeagrassNet methods.**

Range and surface area of *Posidonia* habitat. The only descriptors mandated under the HD are the potential and actual surface area occupied by the habitat. Precision per unit effort, however, is a compromise between classification error and natural spatial variation among replicates. Larger-scale methods with higher coverage (remote acoustic and visual mapping systems) have high classification error but low spatial variation, while microscale methods with low coverage (direct SCUBA plots) have negligible classification error but prohibitively high spatial variance. Between these two extremes is videography, which has negligible classification error (< 2%) but moderate spatial variance among transects due to the relatively large surface area potentially sampled per transect (Puhr *et al.* 2014). This can amount to over a hectare of observed sea bottom per field day under calm conditions, corresponding to over 1 km transect length per hour. As a result, the total sampling error of videography per unit effort is the smallest, and precision is highest, among the three considered methods providing seagrass spatial cover.

Note, however, that acoustic and visual imagery methods are problematic for additional reasons. First, all aerial or satellite sensing techniques currently cannot reliably identify *Posidonia* habitat at depths greater than about 10 - 15 m, which prevents them from providing direct estimates of the total habitat occupancy, or its temporal trend. Second, classification error of visual imagery methods can be strongly biased depending on small variation in atmospheric and oceanographic conditions under which the images are collected. Thus for the classification error to be considered random, replicate image samples must be made across the full range of ambient conditions that influence that error. This means that these replicate samples must be dispersed throughout the monitoring period, and not just concentrated at the end points. Third, acoustic methods currently are unable to consistently distinguish among live *Posidonia*, bare gravel, and macroalgae on sediment at any depth (Puhr *et al.* 2013; Montefalcone *et al.* 2013).

Conservation status of *Posidonia*. The basic choice here is whether conservation status is conceived as the standing value of a descriptor or its temporal change during the monitoring interval. Our results show that a common descriptor of conservation status, standing shoot density obtained by a diver from random 40 by 40 cm quadrats, is too spatially variable to be used meaningfully as a descriptor of the mean state of a meadow.

Because shoot density can be measured only within the time and labor limitations of SCUBA, a minimum of several hundred, depth-controlled, independent samples for each study meadow simply is not feasible for routine, regular monitoring of multiple sites as required under the HD. The low statistical power of such measures was pointed out over 12 years ago out by Duarte (2002): “The low power of monitoring techniques implies that most monitoring programmes can only detect a reliable tendency towards seagrass loss when the seagrass meadows monitored have already experienced substantial damage. There is, therefore, an urgent need to design more effective monitoring approaches, capable of detecting losses of 10% or less, as well as to develop early warning indicators of decline.” As an indicator of conservation status, standing shoot density and similar measures are problematic also because they can be low or zero entirely due to normal, natural variation within and among meadows. For all these reasons, the current scientific consensus among *Posidonia* experts is that standing shoot density should be avoided as a management descriptor for *Posidonia oceanica*: “Furthermore, the variability of shoot density is considerable, at short or mid distance, within the meadow, thus interpreting this parameter is very tricky and requires the greatest prudence .... To avoid simplistic mistakes, we suggest that it should not be routinely used by administrations responsible for the coastal environment” (Boudouresque *et al.* 2012). Here, the authors are referring to the standing shoot density as a measure of conservation status.

Estimation of temporal trend in interior meadow descriptors. The above problems can be solved easily by defining conservation status not as the standing value of the descriptor, but by its trend over time, e.g. simply as its mean change within the meadow during the monitoring period as measured in the balisage method (Sandulli *et al.* 1998; Borg *et al.*, 2006; Boudouresque *et al.* 2012).

However, this course of action is not without risk, as underwater markers are known to move, and can cause changes in sedimentation that influence seagrass cover near the markers (Boudouresque *et al.* 2012); they thus are considered by some management agencies to constitute an anthropogenic disturbance.

Estimation of temporal trend in habitat occupancy. While the sampling error of microscale, SCUBA-based interior descriptors is large and prohibitive, the sampling error of surface cover is far lower due to the fact that this descriptor can be measured by non-SCUBA methods and hence can be assessed over a larger area (20 to 30 times) per replicate.

## **Conclusions**

Because spatial variation within a meadow of the seagrass *Posidonia oceanica* is high, monitoring the conservation status of a local meadow along a 1-km shoreline is difficult, but can be accomplished in one of two ways: 1) using a remote method capable of sampling a large area in a short period of time, which reduces random sampling error among replicates; or 2) ignoring spatial standing variation and instead directly monitoring the change or trend in status at permanently marked sampling locations (fixed-plot methods), whose variation within a site is likely to be small if loss is caused by diffuse changes in water quality. A third possibility, using direct SCUBA to assess a microscale descriptor such as shoot density, and attempting to distinguish mean change in this descriptor across a meadow from its very large spatial variation, is not a viable method because it requires a prohibitively large field sampling effort to demonstrate a moderate loss. Shoot density and similar microscale descriptors should be avoided unless they are used within a fixed-plot design with permanent underwater markers. Management

agencies using standing shoot density as a key descriptor should seriously and honestly evaluate the cost-effectiveness and goals of the monitoring protocol. If permanent underwater markers are to be avoided, then vehicular videography is the method with currently the lowest sampling error for monitoring surface cover, and has negligible classification error. Other remote methods involving modeling acoustic or visual data have large classification errors that require substantial field effort to overcome. We recommend vehicular videography as a simple and high-accuracy method for satisfying the *Posidonia* habitat coverage requirement of the EC Habitats Directive.

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## **EFFECTS OF THE INVASIVE SEAGRASS *HALOPHILA STIPULACEA* ON THE NATIVE SEAGRASS *CYMODOCEA NODOSA***

### **Abstract**

*Halophila stipulacea* is a dioecious seagrass that colonized the Mediterranean basin following the opening of the Suez Canal (1869). It was one of the first lessepsian migrants reported from at least 11 Mediterranean countries. Since it first recorded in 2003 in the Gabes gulf, *H. stipulacea* showed a rapid expansion along the Tunisian coasts. We investigated the ecological survey of *H. stipulacea* in Marina Cap Monastir during the period May 2011 and May 2014. A total of four transect lines along 100 m and 50 m intervals at depths ranging from 0.5 to 4 m were performed. The mean percentage cover of all benthic plants was estimated showing a quick colonization inside the Marina by *H. stipulacea*. In the first field campaign (May 2011), this plant covered a surface area of 2 000 m<sup>2</sup>. It increased rapidly reaching a value of 22 000 m<sup>2</sup> in May 2014. Besides *Cymodocea nodosa* which was abundant in early 2010 on the outside of the Marina harbor is currently observed in only very restricted areas.

**Key-words:** *Halophila stipulacea*; Lessepsian migrant; *Cymodocea nodosa*; Competition; Tunisia.

### **Introduction**

*Halophila stipulacea* (Forsskål) Ascherson is a marine euryhaline seagrass with a wide ecological range of tropical to subtropical affinity (Lipkin, 1975). This species native to the western Indian Ocean (den Hartog, 1970) was one of first lessepsian migrants. In the Mediterranean Sea, its first report was in 1895 off the Cyprus coast (Fritsch, 1895). Thereafter, it spread along the eastern Mediterranean coasts (Lipkin, 1975; Kashta & Pizzuto, 1995). *H. stipulacea* colonized progressively westward through Malta (Lanfranco, 1970) and the Ionian coast of Sicily (Biliotti & Abdelahad, 1990; Alongi, 1993). Currently, Vlora Bay (Albania) is the northern limit of distribution for *H. stipulacea* in the Mediterranean (Kashta & Pizzuto, 1995).

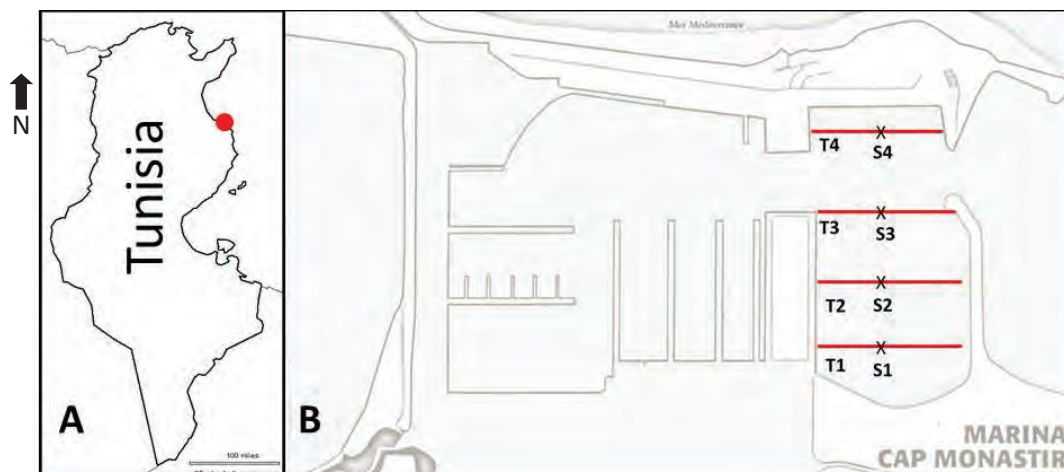
The first observation of *H. stipulacea* on the Tunisian coast was done at Sfax (south east Tunisia) by Missaoui et al (2003). This species was then observed at Monastir (Sghaier et al., 2011) which is considered its north distribution limit off the Mediterranean southern coasts (Fig. 1).

According to Boudouresque and Verlaque (2002), *H. stipulacea* in the Eastern Mediterranean, can be tentatively added to this list of invasive species. No direct damage to local seagrass meadows in the Mediterranean by *H. stipulacea* has been reported yet; however, it seems that this invasive species prefers settling on dead mattes of endemic Mediterranean seagrass *P. oceanica* (Gambi et al., 2009), and therefore might prevent the future recovery of its mattes.

In this point of view we assess the cover evolution of cover and density of *H. stipulacea* meadows in Marina Cap Monastir during the period May 2011 and May 2014.

### Materials and methods

The study site is located in the outer harbor of Marina Cap Monastir (35°46'44"N10°50'07"E Tunisia) (Fig. 1), with sandy substratum at 1.5 m depth colonizing the seagrass meadow of *Cymodocea nodosa* (Ucria) Ascherson, 1870 and some algal species, of which *Caulerpa prolifera* (Fosskti) Lamouroux is the most abundant. Maximum depth in this part of the Marina is approximately 4 m, with an average of 0.8 m; the bottom is relatively flat and muddy.



**Fig. 1: A. location of the Marina Cap Monastir - B. Map of the Marina with the position of the four transects (T1-T4) and the four sampling stations (S1-S4).**

We have investigated the distribution of the two seagrasses (*H. stipulacea* and *C. nodosa*) in Marina Cap Monastir through prospecting campaigns covering the whole Marina during May 2011, 2012, 2013 and 2014. Observations was carried out along 4 transects (T1 to T4) and according to a prospecting itinerary along the Marina docks (Fig. 1). The distribution of the both seagrasses was mapped using a GPS (Global Positioning System). Quantitative samples were taken over a surface of 20 x 20 cm (10 replicates) in four sampling stations (Fig. 1 and Table 1). In each quantitative sample and to estimate the shoot density, the number of shoots of *C. nodosa* and *H. stipulacea* was counted.

A standardized one-hour transects of various lengths were surveyed by snorkeling or/and diving, at depth range between 0 to 30 m in neighbouring areas of the Cap Marina Monastir aiming to asses new *H. stipulacea* patch.

**Tab. 1: Depth and substratum type in the four sampling stations**

	S1	S2	S3	S4
<b>Depth (m)</b>	0.5	0.8	2	1
<b>Substratum</b>	sandy	muddy	muddy	sandy

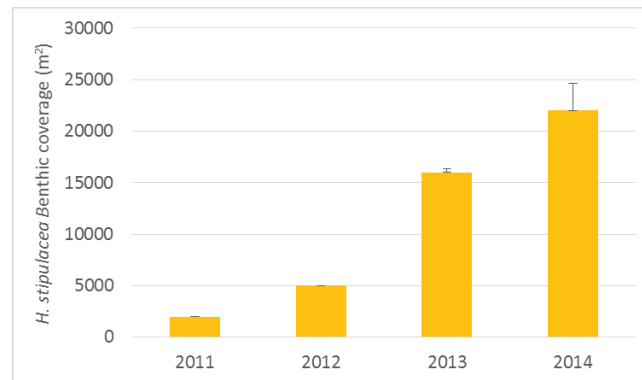
### Results

#### *Halophila stipulacea* coverage

The mean percentage cover of all benthic plants was estimated showing a quick *H. stipulacea* colonization inside the Marina. The covered surface area in the first field



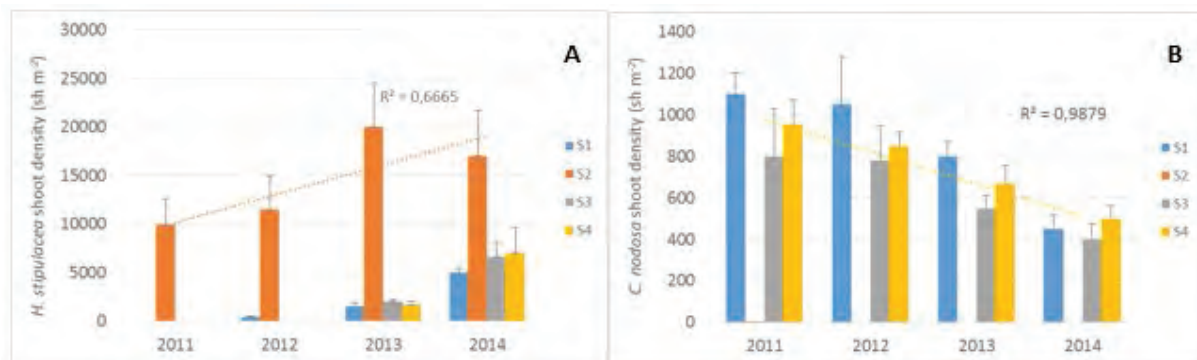
campaign (May 2011) was 2 000 m<sup>2</sup>. It increased rapidly reaching a value of 22 000 m<sup>2</sup> in May 2014 (Fig. 2 and 4). Besides *C. nodosa*, abundant in early 2010 in the outer harbor of the Marina, was currently observed in only very restricted areas.



**Fig. 2:** *Halophila stipulacea* benthic coverage during the four campaigns.

### Shoot density

The analysis of the influence of time on the four sampling stations reflects a fast increase of *H. stipulacea* shoot density (Fig. 3A, from 2 475 in May 2011 to 8 895 shoots m<sup>-2</sup> in May 2014). The evolution over time of *C. nodosa* shoot density showed a clear decrease from 714 shoots m<sup>-2</sup> in May 2011 to 313 in May 2014 (Fig. 3B).



**Fig. 3:** Evolution of shoot density of the two species *Halophila stipulacea* (A) and *Cymodocea nodosa* (B).

Liberated fragments of the invasive seagrass *H. stipulacea* were found 1km south of the Marina in popular anchorages area frequented by fishing and recreational boats, however, the distribution of the seagrass is confined to these locations.

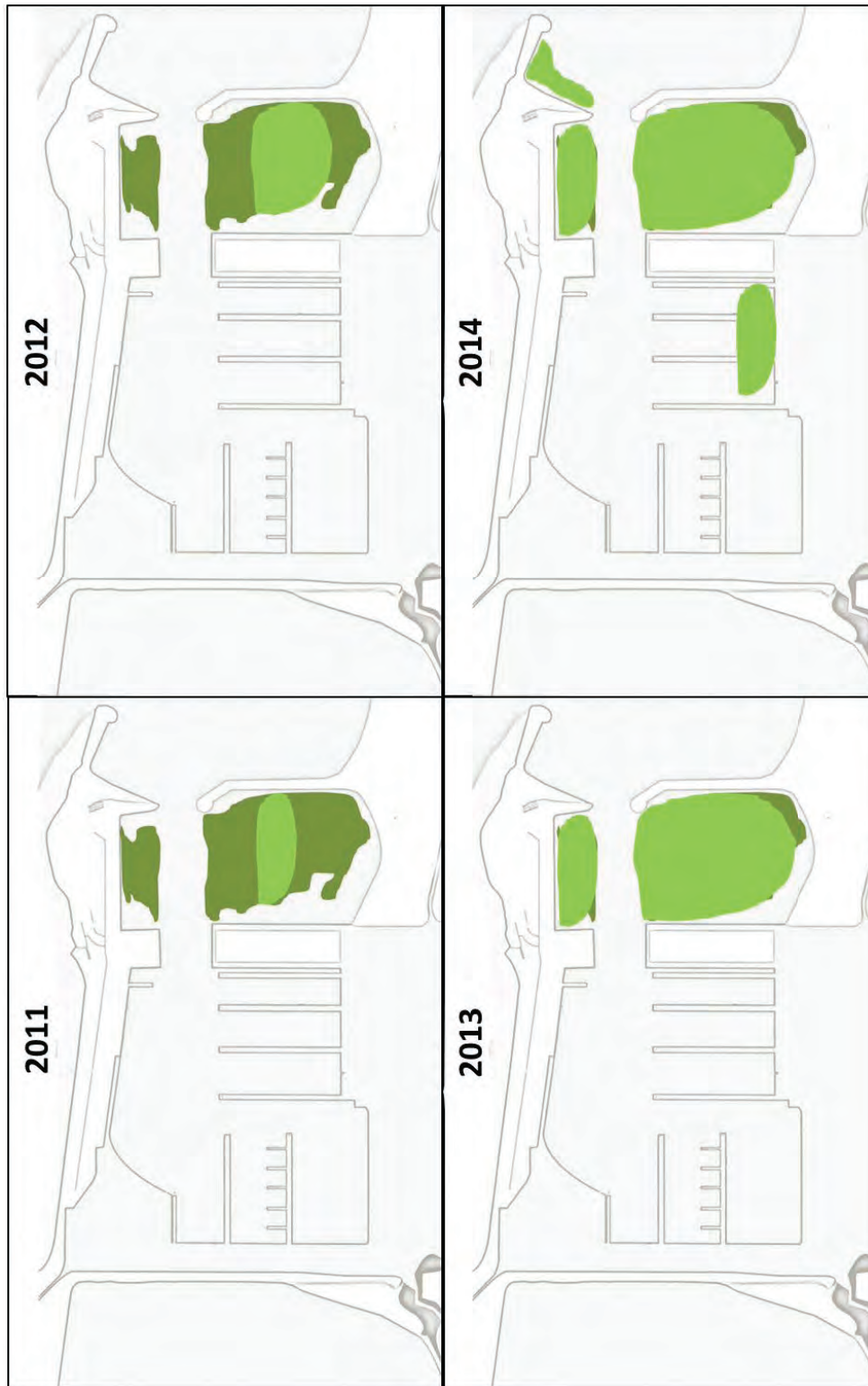


Fig. 4: Temporal changes in the spatial distribution of *Halophila stipulacea* (light green) and *Cymodocea nodosa* (dark green) in Marina Cap Monastir from May 2011 to May 2014.

### Discussion and conclusion

Three years following our first record in 2011 where a single 0.2 ha monospecific *H. stipulacea* patch in Marina Cap Monastir was reported, the current distribution of this species covering of 2.2 ha, *H. stipulacea* is unquestionably now well established in Cap Monastir.

Within the four years of the field campaigns, *C. nodosa* has disappeared within the permanent transect. A drastic change in the structure of the benthic macrophyte community in the study area invaded by *H. stipulacea*, with a significant decrease in *C. nodosa* shoot density was observed when *H. stipulacea* reached its maximum invasion. Similar to the Eastern Caribbean report (Willette *et al.*, 2013), *H. stipulacea* does compete with the native Caribbean seagrass *S. filiforme* for space and has empirically been shown to alter the abundance and composition of seagrass-associated organisms and the local seagrass community (Willette & Ambrose, 2012). This substitution of *C. nodosa* by opportunistic "introduced" species (*H. stipulacea*) with weaker structuring capacities may trigger profound changes within the communities (Montefalcone *et al.*, 2010).

Further, more investigations on the interaction of *H. stipulacea* with native seagrass will be encouraged, including those with other *Halophila* species inhabiting ecologically similar habitats.

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## **DYNAMICS OF BENTHIC MACROPHYTES IN THE SOUTHERN TUNIS LAGOON (TUNISIA, MEDITERRANEAN SEA)**

### **Abstract**

*The Southern Tunis Lagoon (north-eastern Tunisia) has long been severely impacted by man-induced eutrophication. Like the Northern Tunis Lagoon, the Southern Tunis Lagoon has been the subject of an ambitious plan of restoration, implemented between 1998 and 2001. The program consisted mainly in deepening the lagoon, reducing its surface area and establishing one-way water circulation, ensuring a continuous and relatively rapid water renewal. Since then, the water quality has conspicuously improved and the macrophyte community dramatically changed. The authors show the decline of pollution-tolerant species and of free-living macrophytes. In contrast, the Ulvophyceae *Caulerpa prolifera* and the seagrass *Cymodocea nodosa* have become the dominant species, while species diversity has conspicuously increased. Although the restoration program is undoubtedly a success story, it is worth emphasizing that a return to a hypothetical natural condition is less clear, as 'natural' conditions continuously changed over the Holocene, then human, history of the lagoon.*

**Key-words:** *Caulerpa prolifera*, *Cymodocea nodosa*, brackish lagoon, eutrophication, water quality improvement.

### **Introduction**

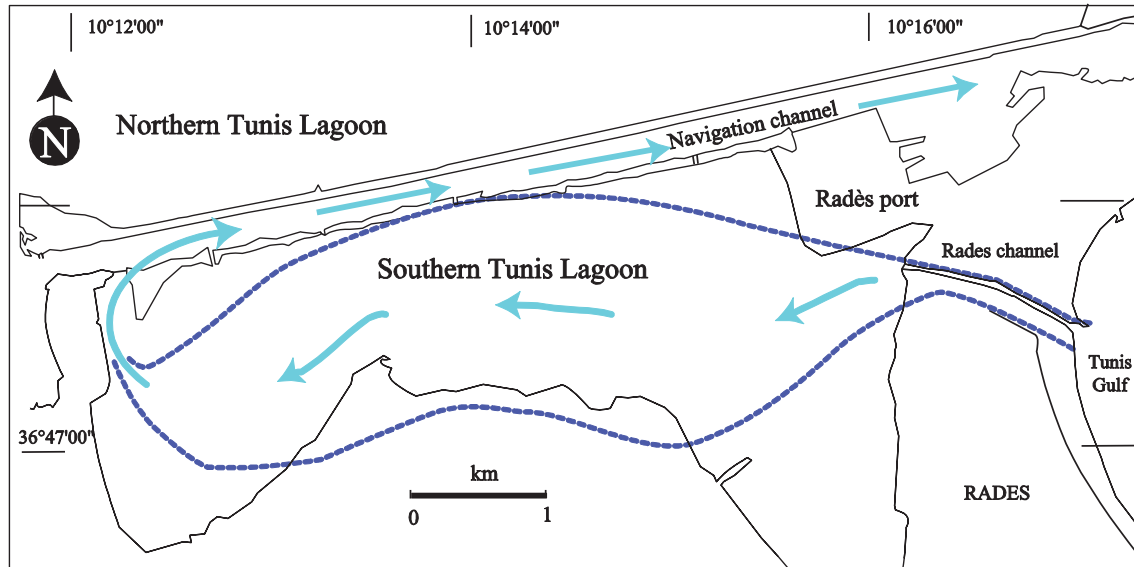
The Tunis Lagoon, often incorrectly called 'Lac de Tunis', is located in the Gulf of Tunis (Tunisia), between the city of Tunis and the open sea. It was artificially divided into two lagoons, the Northern Tunis Lagoon and the Southern Tunis Lagoon, by a dam used as a highway and railway and by a navigation channel. It therefore constitutes today a complex of two shallow lagoons. This complex has a high heritage value, included in the Ramsar Convention list of wetlands of international importance (Schneider, 1977). It is also of importance for the Tunisian artisanal fishery.

During decades, the untreated sewage of the city of Tunis (~150 000 and more than 1 million inhabitants in early and late 20<sup>th</sup> century, respectively) was discharged into the Tunis Lagoon. The primary producers gradually changed from *Ulva* sp., *Zostera noltei*, *Ruppia cirrhosa* and *Acetabularia acetabulum* (early 20<sup>th</sup> century; Heldt, 1929), to a *Caulerpa prolifera* dominated stand (Molinier & Picard, 1954), then an *Ulva* sp., *Chaetomorpha* sp. and *Gracilaria* sp. stand (Zaouali, 1971; Baier et al., 1977) and it finally became one of the most polluted brackish lagoons worldwide, with blooms of *Ulva* spp. (Ulvophyceae), red tides, fish mass mortality, due to anoxia, and a strong smell of hydrogen sulphide, due to the summer bacterial decomposition of the *Ulva* biomass (Schneider, 1977; Zaouali, 1977; Belkhir & Hadj Ali Salem, 1981).

After the successful restoration of the Northern Tunis Lagoon (Ben Maïz, 1993), a 3-year plan for the restoration of the Southern Tunis Lagoon was initiated in 1998 (VandenBroeck & Ben Charrada, 2001). This plan encompassed the dredging of highly contaminated sediments (heavy metals, oil), the deepening of the lagoon (to a mean depth of 2 m, up to 4.5 m), reduction of its surface area (from 1 500 to 710 ha) and the induction of water cyclonic circulation and exchange with the open sea, reducing the residence time of the water to 4-7 d (Fig. 1).



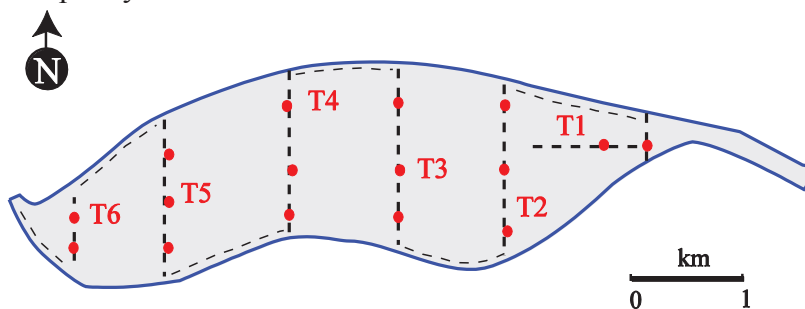
Here, we describe the changes in the benthic macrophyte stands which occurred after the implementation of the restoration plan of the Southern Tunis Lagoon.



**Fig. 1: The southern Tunis Lagoon, within the framework of the restoration plan. Arrows: new water circulation. Dashed line: the after-restoration limits of the lagoon.**

### Material and methods

Eight surveys of the benthic macrophytes of the Southern Tunis Lagoon were performed in March (spring), June (summer), September (autumn), and December (winter) 2010 and 2011. For mapping, a GPS (Global Positioning System) was used. The macrophyte biomass (dry mass DM,  $\text{g m}^{-2}$ ) was measured at 16 sites (3 samples of  $0.25 \text{ m}^2$  per site), along 6 transects (T1 through T6) (Fig. 2), by scuba diving. At the scale of the lagoon, the total biomass of dominant species was assessed by multiplying their mass within a sample by the surface area of the considered habitat.



**Fig. 2: Study transects (T1 through T6) and samples (red dots) for the assessment of the macrophyte biomass in the Southern Tunis Lagoon**

### Results

The following macrophytes species were observed in the Southern Tunis Lagoon, in 2010-2011: *Cymodocea nodosa* (Ucria) Ascherson, *Zostera noltei* Hornemann, *Ruppia cirrhosa* (Petagna) Grande (Magnoliophyta, Archaeplastida), *Caulerpa prolifera* (Forsskål) J.V. Lamouroux, *Caulerpa cylindracea* Sonder, *Cladophora* sp., *Chaetomorpha linum* (O.F. Müller) Kützing, *Codium fragile* (Suringar) Hariot subsp. *tomentosoides* (van Goor) P.C. Silva, *Ulva intestinalis* Linnaeus, *Ulva linza* (Linnaeus) J. Agardh, *Ulva prolifera* O.F. Müller, *Ulva rigida* C. Agardh, *Ulva* sp. (Chlorobionta, Archaeplastida), *Gracilaria* cf. *gracilis* (Stackhouse) Steentoft, Irvine & Farnham, *Gracilaria bursa-pastoris* (Gmelin) Silva, *Hypnea musciformis* (Wulfen) Lamouroux,



*Jania rubens* (Linnaeus) J.V. Lamouroux, *Corallina elongata* J. Ellis & Solander (Rhodobionta, Archaeplastida) and *Cystoseira barbata* (Stackhouse) C. Agardh, *Ectocarpus* sp. and *Petalonia fascia* (O.F. Müller) Kuntze (Phaeophyceae, Stramenopiles).

Three species were abundant everywhere: the sessile *Caulerpa prolifera* and the free-living *Chaetomorpha linum* and *Ectocarpus* sp. *Cymodocea nodosa*, *Caulerpa cylindracea*, *Codium fragile* and *Cystoseira barbata* were mainly abundant in the eastern part of the lagoon, where the marine influence is higher. *Ruppia cirrhosa* was localized within small bodies of stagnant water, near the navigation channel. *Zostera noltei* was observed sporadically in the eastern part of the lagoon. Species abundance was also season-dependent (Tab. 1) and year-dependent: in 2011, free-living species (*C. linum* and *Ectocarpus* sp.) were less abundant, while fixed species (*C. prolifera* and *C. nodosa*) were more abundant, than in 2010.

Year	Season	West	Centre	East
2010	Spring	Cl	E	Cn Cp (E)
	Summer	Cp Cl (E)	E Cp	Cn Cp (Cb)
	Autumn	Cp (Cn)	Cp (Cl Cn)	Cn Cp
	Winter	Cp (Cn)	Cp E	Cn Cp (Cb)
2011	Spring	Cl Cp	Cp E	Cn Cp (Cc Cb)
	Summer	Cp (Cn)	Cp (Cl)	Cn Cp (Cc)
	Autumn	Cp	Cp	Cn (Cb)
	Winter	Cp (Cl Cn)	Cp Cl	Cc Cn Cp (Cl)

**Tab. 1: Seasonal changes in the abundance of macrophytes in the Southern Tunis Lagoon (years 2010-2011).**

**Cb = *Cystoseira barbata***

**Cc = *Caulerpa cylindracea***

**Cl = *Chaetomorpha linum***

**Cn = *Cymodocea nodosa***

**Cp = *Caulerpa prolifera***

**E = *Ectocarpus* sp.**

**Brackets mean lesser abundance.**

The percent cover of macrophytes, irrespective of the species, was also site-, season- and year-dependent (Fig. 3 and 4). In 2010, the cover was higher in the west and the centre in winter and spring, in the centre and the east in summer, and in the west in autumn (Fig. 3). In 2011, the seasonal pattern was similar in winter and spring, but different in summer (higher in the west and east) and in autumn (higher in the west and centre) (Fig. 4). The macrophyte biomass ranged from 0 to ~500 g DM m<sup>-2</sup> (Tab. 2). There were no conspicuous trends between sites, seasons and years. In 2010, a decreasing eastward gradient was however observed in summer. In 2011, this gradient was present to a greater or lesser extent year round.

Year	Season	T6	T5	T4	T3	T2	T1
2010	Spring	310	266	237	97	71	157
	Summer	98	83	125	270	391	500
	Autumn	218	218	266	148	62	146
	Winter	196	284	237	299	235	81
2011	Spring	298	249	117	132	163	101
	Summer	312	305	119	95	243	194
	Autumn	317	303	302	223	189	76
	Winter	373	294	305	481	112	41

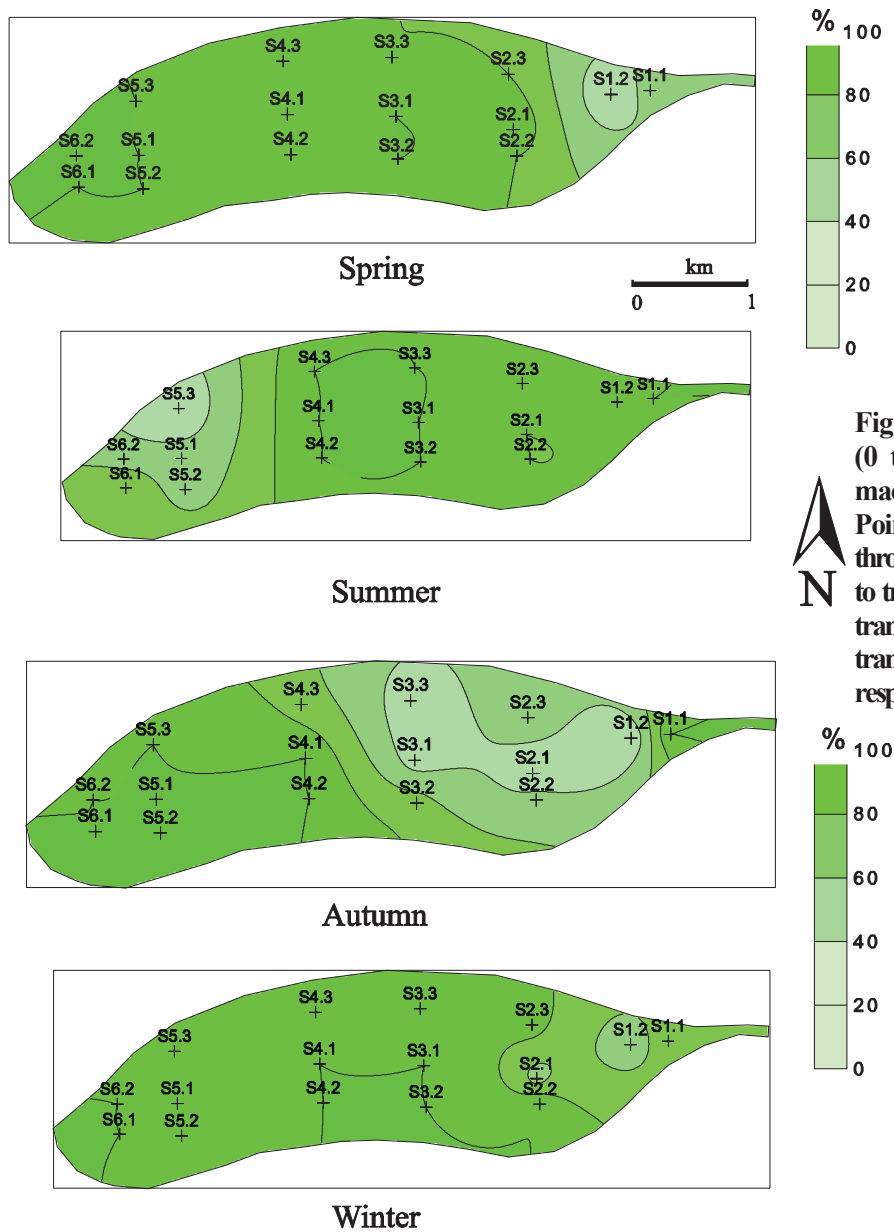
**Tab. 2: Biomass of benthic macrophytes (including free-living) (dry mass DM, g m<sup>-2</sup>), along transects T1 (East) through T6 (West) (see Fig. 2 for localisation).**

## Discussion and conclusions

Before the implementation of the restoration plan of the Southern Tunis Lagoon (1998-2001), the macrophyte species diversity was very low, with mainly species of the genus

*Ulva*. Ten years later, the recovery of the macrophyte species diversity is impressive, with ~20 common species, in addition to rare and inconspicuous small species, probably present but not searched for in the framework of the present study. Taking into consideration that species diversity is naturally low in brackish lagoons, the number of observed species is usual for a healthy lagoon habitat.

Today, the dominant macrophyte species are the seagrass *Cymodocea nodosa* (mainly in the eastern zone) and the Chlorobionta *Caulerpa prolifera* (mainly in the central and western zones). These species, which thrive both in Mediterranean lagoons and in the open sea, can be considered as indicators of relatively healthy conditions (Boudouresque *et al.*, 2009). In addition, these species stabilize the sediment, by means of their roots, rhizomes and rhizoids, and contribute to hindering sediment re-suspension and therefore enhance water transparency. In contrast, free-living macrophytes, such as *Ectocarpus* sp. and *Chaetomorpha linum*, often considered as indicators of only slight eutrophication (Trabelsi, 1995), are on the decline.



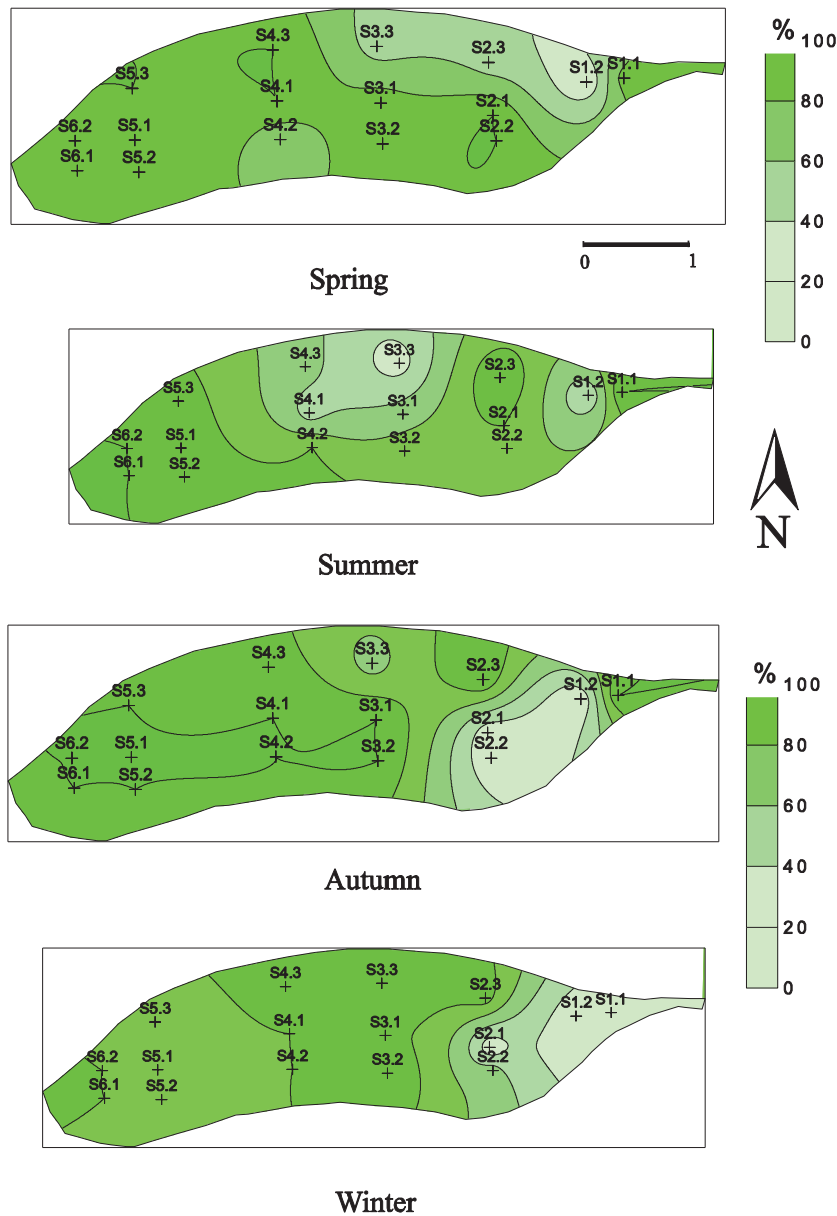
**Fig. 3: Percent cover (0 through 100%) of macrophytes in 2010. Points (crosses) S1.1 through S6.2 correspond to transects and samples: transect T1, sample 1 and transect T6, sample 2, respectively.**

Contrary to the species diversity and the dominant species, the biomass of macrophytes did not conspicuously change before and after the lagoon restoration, at least in spring and autumn (Tab. 3).

Year	Spring	Summer	Autumn	Winter
1996	1 147	514	1 143	274
2010	1 121	1 516	992	1 466
2011	1 024	1 072	1 535	1 561

**Tab. 3: Total biomass of macrophytes (metric tonnes of dry mass) in the Southern Tunis Lagoon, in 1996 (before the restoration plan; Shili et al., 2002) and in 2010 and 2011 (this work).**

Overall, the restoration program of the Southern Tunis lagoon has resulted in the improvement of the water circulation, the lessening of the mean residence time of the water, the decrease of the nutrient concentration, the reduction of the salinity range (Jouini *et al.*, 2005), the increase in marine influence, both abiotic and biotic, the increase in macrophytes species diversity and the decrease in species indicators of eutrophication.



**Fig. 4: Percent cover (0 through 100%) of macrophytes in 2011. Points (crosses) S1.1 through S6.2 correspond to transects and samples: transect T1, sample 1 and transect T6, sample 2, respectively.**

The restoration program of the Southern Tunis Lagoon must be regarded as a success story. The current state of the lagoon is obviously consistent with the welfare and requirements of the inhabitants of a large modern city, Tunis. However, it is worth emphasizing that the return to a hypothetical natural condition is far from being clear. Over the Holocene, then human, history of the lagoon, it continuously changed from a marine embayment to an open or closed lagoon, more or less brackish, more or less naturally eutrophic (Thornton, 1976). It is therefore uneasy to define 'natural' conditions. The current situation, which is a source of legitimate pride for managers and which enhances inhabitant welfare, tourism and fishery, is probably partly artificial.

### Acknowledgements

The authors are indebted to Société d'Etude et de Promotion de Tunis Sud (SEPTS) and Eco-Ressources International for scientific involvement and field assistance and to Michael Paul for improving the English text.

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## **DISTRIBUTION AND HABITAT REQUIREMENTS OF *ZOSTERA NOLTEI* ALONG THE NORTHERN COAST OF JERBA ISLAND (SOUTHERN TUNISIA, MEDITERRANEAN SEA)**

### **Abstract**

*The marine and brackish Magnoliophyta (seagrass) Zostera noltei is widely distributed in Tunisia. It thrives in sheltered bays and in brackish lagoons, in shallow waters where terrigenous inputs are relatively important. Here, we studied the distribution (mapping, biomass) and habitat requirements of Z. noltei in an embayment and a lagoon of the northern coast of Jerba Island (southern part of the Gulf of Gabès), one of the only two Mediterranean regions (with the northern Adriatic) where tide amplitude is conspicuous. Z. noltei meadows are either monospecific (from the seagrass point of view) or mixed, with Cymodocea nodosa. In comparison with northern Tunisia meadows (e.g. the Gulf of Tunis), in Jerba they are confined to shallower habitats, and exhibit a wider range of tolerance to a number of environmental parameters, e.g. tide, evaporation and salinity. In the absence of a baseline of the past distribution of the two seagrass species, possible changes in the balance between them, in relation with e.g. climate warming and resort development, cannot be evidenced.*

**Key-words:** *Zostera noltei*, *Cymodocea nodosa*, seagrass meadows, mapping, biomass

### **Introduction**

Jerba Island is located at the southern end of the Gulf of Gabès, Tunisia (Fig. 1). It exhibits a conspicuous marine biodiversity (*sensu* Boudouresque, 2011), of which the natural heritage value is high. It represents a wintering and nesting place for many migratory seabirds and a wintering (rarely a nesting) place for the sea turtle *Caretta caretta* (Linnaeus 1758) (ASSIDJE, 2000). *Posidonia oceanica* (Linnaeus) Delile seagrass meadows cover a wide area. This biodiversity is mainly due to: (i) Diversity of habitats (open sea, coastal lagoons, sabkhas, saltmarshes and islets). (ii) Occurrence of semi-diurnal tides with a range unusual in the Mediterranean Sea (up to 1.8 m), higher than in the northern Adriatic (1.5 m) and most of the Mediterranean (0.2-0.5 m) (Rouch, 1948). For these reasons, ASSIDJE (2000) and Ben Maïz, (2001) have proposed to include some Jerba sites (Ras Remel, Ras El Kastil, Ras El H'Mar and El Gatâaya Islets) within the SPAMIs (Specially Protected Areas of Mediterranean Importance) list.

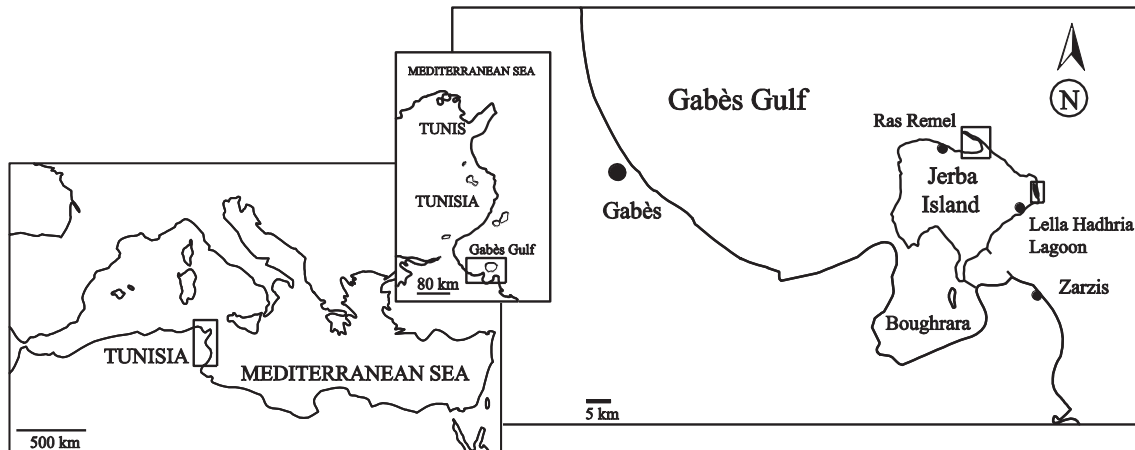
Published studies of the marine habitats and species of Jerba Island are few. They usually predate the peak of pollution of the Gulf of Gabès and the development of tourism on the northern and eastern coast of the island (Seurat, 1929, 1934; De Gaillande, 1970a, b) and poorly describe the current situation (Ben Maïz, 2001).

*Zostera noltei* Hornemann (= *Zostera noltii*, *Nanozostera noltii* (Hornemann) Tomlinson & Poluszny) is a euryhaline seagrass (Magnoliophyta) common in northern and eastern Tunisia, e.g. La Galite, Bizerte Lagoon, North and South Tunis Lagoon, Ghar El Melh and Kalaat El Andalouss lagoons and open sea shores and the shore of the Gulf of Gabès. It dwells in shallow and sheltered bays and in coastal lagoons widely opened to the sea, on sandy and muddy bottoms with relatively high terrigenous sediment supply (e.g.



Lovric, 1979; Boudouresque *et al.*, 2009; Shili *et al.*, 2010). As regards the seagrasses, the meadows can be monospecific, often adjacent to *Cymodocea nodosa* (Ucria) Ascherson meadows, or mixed with *C. nodosa*, more rarely with *Ruppia cirrhosa* (Petagna) Grande in some lagoons (Shili, 2008).

Here, we describe two habitats with more or less lagoon conditions, located on the northern coast of Jerba Island (Ras Remel and Lella Hadhria), which harbour stands of *Z. noltei* (Fig. 1).



**Fig. 1: Localisation of Jerba Island and the two study sites.**

### Material and methods

Ras Remel (Fig. 1) is a shallow embayment that, in a region subject to significant tides, experiences lagoon-like conditions. In addition, it is of great economic value for the artisanal and recreational fishery, the latter linked with the tourism economy. North of the sand spit, in the open sea, the *P. oceanica* meadow is relatively dense, although exhibiting, at its landwards limit, signs of degradation and of loss if compared with early 20<sup>th</sup> century data (SCET-TUNISIE, 1999).

The Lella Hadhria Lagoon (Fig. 1) is situated in a zone highly impacted by resort development. This shallow lagoon (less than 1-2 m depth), with sandy and muddy substrate, consists of two parts: **(i)** An eastern basin ~500 m long and ~320 m wide, which directly communicates with the sea *via* several natural passes opened through a rocky ridge; the height of this ridge is low, so that waves easily cross it during storms. This basin is used as a natural harbour by fishermen, which results in pollution; water movement (generated by the waves that break on the rocky ridge and by the entry of marine water through the passes) produces sediment re-suspension and therefore low water transparency, together with nutrient dissemination. **(ii)** A northern linear basin ('La Saguia'), which stretches over 2.0 km (3.4 km including the northernmost part, occasionally flooded), more or less parallel to the shoreline. Water movement (generated by wind) produces the same effects as in the eastern basin. Tides, either lunisolar (combined effects of the gravitational forces exerted by the Moon, the Sun and the rotation of the Earth) or meteorological (effects of barometric pressure, seiches, wind and storms), result in more or less long-lasting periods of drying. Off the Lella Hadhria Lagoon, in the open sea, the upper limit of the *P. oceanica* meadow is located at 8 m depth. The meadow is healthy and thick *banquettes* of dead leaves are cast ashore.

Study campaigns were organized in April-May 2009 to localize and quantify the *Z. noltei* meadows (together with *C. nodosa*) at Ras Remel embayment and in Lella Hadhria

lagoon. Their abundance (percentage cover) was measured by snorkelling along regularly spaced (500 m) transects. Depth, salinity and biomass (dry mass, DM; 0.25 m<sup>2</sup> quadrats, three replicates) were also measured.

### Results and discussion

In the Gulf of Gabès, the mean annual temperature is 19.4 °C, with wide seasonal variation: from 13 °C in winter to 26 °C in summer (SCET-TUNISIE, 1999). At the Ras Remel site, which is shallow and subject to high evaporation, the mean salinity (spring 2009) was 41. In summer, salinity could reach 44 at the head of the embayment, in relation with the southern Tunisian hot and dry climate. A minimum salinity of 36.6 was recorded near the seaward end of the sand spit. At the Lella Hadhria Lagoon, relatively high water temperatures could be observed in summer in the north of the shallow northern basin, with a decreasing gradient towards the eastern basin. Similarly, there was a strong salinity gradient (up to 14) between the north of the northern lagoon and the eastern basin mouth, near the passes, where salinity was close to that of the open sea, due to shallowness, strong evaporation and slow water circulation in the northern basin. Under low wind conditions, water transparency was good everywhere, while under strong wind conditions, transparency was lowest in the northern basin.

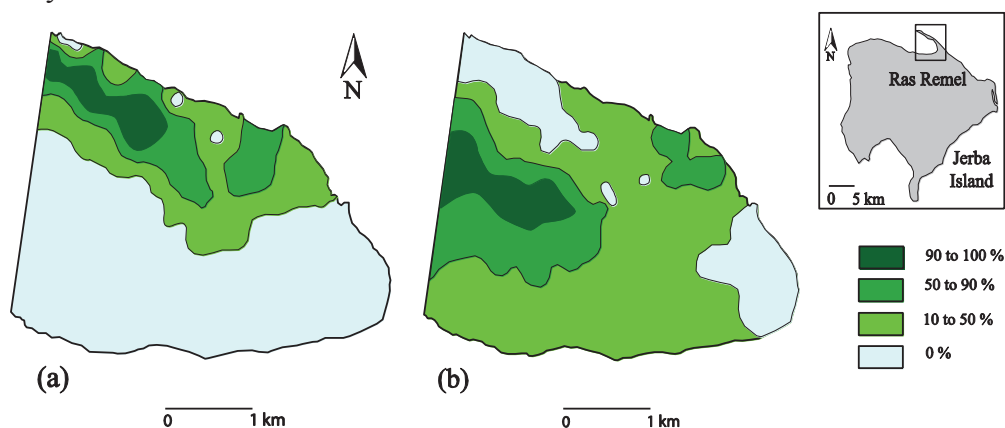
**Tab. 1: Surface area of the seagrasses (Magnoliophyta) *Z. noltei* and *C. nodosa* in Ras Remel (Jerba), as a function of their density (% cover) (Spring 2009). The two species may coexist in the same areas and therefore overlap (see Fig. 2).**

Cover (%)	Surface area of <i>Z. noltei</i> (km <sup>2</sup> )	Surface area of <i>C. nodosa</i> (km <sup>2</sup> )
10-50	2.60	8.20
50-90	1.70	2.81
90-100	0.70	1.14
<b>Total</b>	5.00	12.15

In the Ras Remel embayment, more or less dense *Z. noltei* meadows (50-100 %) were thriving in the NW, constituting monospecific stands or mixed stands with *C. nodosa* (Fig. 2). *Z. noltei* occurred at 20 to 90 cm depth (at low tide) on sandy and muddy substrate. For the whole area, low cover meadows (10 to 50 %) were the most common, *Z. noltei* being generally associated with *C. nodosa* (Fig. 2, Table 1). *C. nodosa* thrived from 10 to 100 cm depth and did well in salinity exceeding 41. The mean biomass (dry mass) of *Z. noltei* was of 219 g m<sup>-2</sup>, 162 g m<sup>-2</sup> and 22-28 g m<sup>-2</sup> in very dense (cover 90 to 100 %), dense (cover 50 to 90 %) and sparse (cover 50 to 10 %) meadow, respectively. Flowering of *Z. noltei* was observed in March-April, sometimes up to late May. Flowering of *C. nodosa* was also observed in spring, but less frequently. Outside the mapped area, *C. nodosa* constitutes monospecific stands, i.e. without *Z. noltei*, the latter species being hindered by water turbidity and a sandy substrate.

In the Lella Hadhria lagoon, *C. nodosa* is the dominant seagrass (Fig. 3). The eastern basin is under marine influence, while the northern basin (La Saguia), is under more marked continental influence. Taking into account the benthic primary producers, the Lella Hadhria Lagoon can be divided into four ecological zones (Fig. 3). (i) Zone 1 corresponds to the eastern basin. It constitutes the deeper part of the lagoon (0.5 to 2 m depth) and the direct marine influence, through the passes, is high. It is characterized by a mixed meadow of *C. nodosa* and *Z. noltei*. The bottom is carpeted by drifting *P. oceanica* leaves coming from the open sea. Along the western edge of this basin,

nitrophilous Chlorobionta (green ‘macroalgae’) are bioindicators of the organic pollution due to its utilisation as a port: *Ulva torta* (Mertens) Trevisan, *Ulva linza* (Linnaeus) J. Agardh, *Chaetomorpha linum* (O.F. Müller) Kützing, *Rhizoclonium* sp. and *Cladophora* spp.; *Codium bursa* (Olivi) C. Agardh and *Halimeda tuna* (Ellis & Solander) Lamouroux were also present. (ii) Zone 2 is shallow (<1 m) and corresponds to the beginning (some hundred metres) of La Saguia. The marine influence is still significant. The species diversity of macrophytes is relatively high, with a *C. nodosa* meadow or a mixed *C. nodosa* and *Z. noltei* meadow. The biomass of *C. nodosa* is high (414 g DM m<sup>-2</sup>). *Rhizoclonium* sp., *Chaetomorpha linum* and *Cladophora* sp. (Chlorobionta) are also present. (iii) Zone 3 corresponds to the greater part of La Saguia. Depth and water circulation are reduced. During lowest tides (lunisolar and/or meteorological), it can dry for more or less long-lasting periods. It harbours a *C. nodosa* and *Rhizoclonium* sp. meadow, the density of which conspicuously decreases from the centre to the shore. (iv) Zone 4 is the shallowest (< 30 cm at high tide) and the most subject to water turbidity. It can dry up for relatively long periods. These severe conditions, as a consequence of its confinement, prevent the development of seagrasses and eukaryotic seaweeds; species diversity is low; the only benthic primary producers are Cyanobacteria that constitute a crust layer on the sediment.



**Fig. 2: Map of the Ras Remel (North of Jerba) seagrass meadows (Spring 2009): percent cover. a: *Zostera noltei*; b: *Cymodocea nodosa*.**

Overall, in Jerba lagoons, *C. nodosa* is clearly dominant with respect to *Z. noltei*. The former better withstands unstable conditions, in particular changes in salinity, together with high salinity and high temperature. The latter is restricted to areas with definite marine influence and therefore less variable conditions. A question that arises is the possible role of herbivory in the competition between these two species. The herbivorous teleost *Sarpa salpa* (Linnaeus 1758) is common in the study area; we noticed bite scars on *C. nodosa* leaves, in contrast with the generally intact leaf apices of *Z. noltei*. The voracious herbivorous teleosts *Siganus* spp., introduced from the Red Sea, although present in Tunisia (Por, 1978; Boudouresque *et al.*, 2005; Sala *et al.*, 2011; Giakoumi, 2014), were not observed within the study lagoons. In the absence of an accurate study of the herbivory in Jerba lagoons, the authors' feeling is that herbivory does not constitute the main driver for seagrass distribution. Another question is the role of the current warming of Mediterranean waters (Moron, 2003), prone to favour *C. nodosa*, a species of warm affinities, at the expense of *Z. noltei* (Boudouresque *et al.*, 2009; Pergent *et al.*, 2012). The latter species has actually undergone a dramatic decline in the neighbouring

Boughrara Lagoon. Unfortunately, an accurate description of its past distribution, in the two northern Jerba coastal lagoons, is lacking. Finally, both species seem to have hitherto withstood the worrying human impact in the area (urbanisation, resort development, pollution).

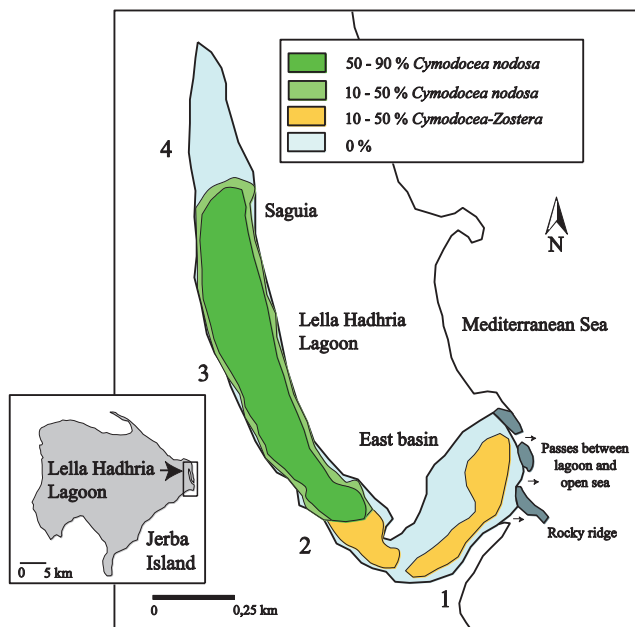


Fig. 3: Mapping of the seagrasses (Magnoliophyta) in the Lella Hadhria Lagoon (Spring 2009). Zones 1 through 4: see text.

If compared with northern Tunisia and other Mediterranean lagoons (Plus *et al.*, 2001; Bernard *et al.*, 2007; Shili, 2008), *Z. noltei* from Jerba seems to better withstand Salinity > 41; however, it is restricted to shallow habitats (< 1 m) due to its sensitivity to low water transparency. In contrast, *C. nodosa* seems to better withstand turbidity, high salinity (up to 44) and more or less long- lasting periods of drying up, depending upon lunisolar and meteorological tides that characterize the Gulf of Gabès area.

### Conclusions

The Gulf of Gabès, in particular Jerba Island, constitutes an area where the effects of human activities, climate change and invasive species can be evidenced and understood, due to its location at the borderline between the eastern and western Mediterranean, in the frontline area with regard to Lessepsian (i.e. Red Sea) species invasions and in an area where coastal development is relatively recent (in contrast with e.g. the French Riviera). A change in the balance between competing seagrasses, *C. nodosa*, *P. oceanica* and *Z. noltei*, can therefore be expected. The present work aims to constitute a baseline, currently unfortunately missing, for further monitoring of the possible changes.

### Acknowledgements

The authors acknowledge with thanks Michael Paul for improving the English text and Abdelmajid Kachkouch, President of the Association pour la Sauvegarde de l'île de Djerba), for help and facilities.

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## **MERCURY CONTAMINATION IN *POSIDONIA OCEANICA* IN A HARBOUR AREA OF THE EASTERN COAST OF TUNISIA**

### **Abstract**

*The seagrass *Posidonia oceanica* is considered to be an accurate bioindicator of past and present mercury contamination. Through the lepidochronology method, it is possible to have information on its life history traits, particularly its metal contamination. In the framework of MAPMED Project, we assess the temporal trend contamination by mercury investigations on dead sheaths of 45 samples of *P. oceanica* collected in the El Kantaoui area (Eastern Tunisian coast). Our results, first reported in this species inhabiting the Tunisian coasts, allowed comparing the mercury contamination in seagrass of El Kantaoui area with other Mediterranean ones. This study confirms the relevance of the use of *P. oceanica* as a biological indicator of metal contamination in coastal ecosystems. Thus the usefulness of *P. oceanica* as a tracer of temporal and spatial metal contamination and as a good tool for water quality health is highlighted.*

**Key-words:** Trace element; Seagrass; Mediterranean Sea; contamination; ICP-OES.

### **Introduction**

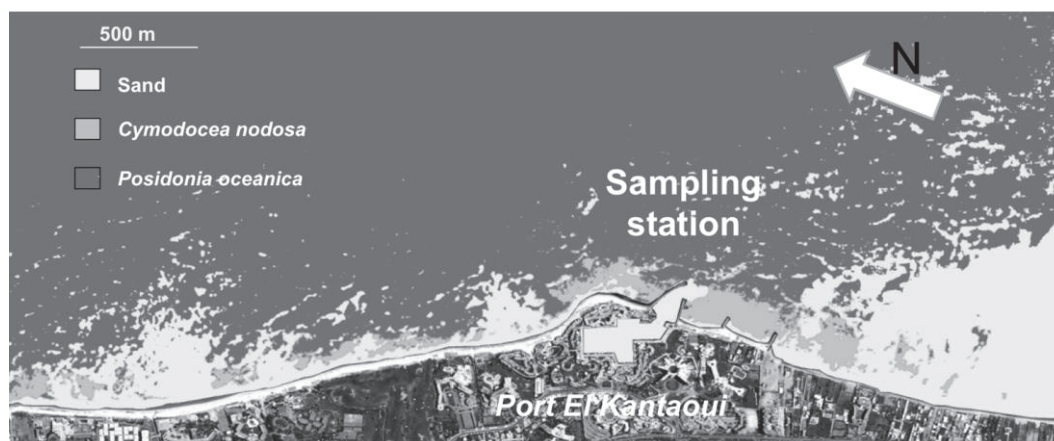
*Posidonia oceanica*, an endemic Mediterranean seagrass, forms dense infralittoral meadows, which are widely distributed along the Tunisian coasts from the surface to depths of 40m. *P. oceanica* meadows constitute an engineering ecosystem playing major ecological, geological and economic roles, but are sensitive to human disturbances such as coastal development and pollution (Boudouresque *et al.*, 2012). Like the majority of seagrasses, this plant is able to accumulate a range of trace metals (Costantini *et al.*, 1991; Malea *et al.*, 1994; Prange et Dennison, 2000; Gosselin *et al.*, 2006; Lafabrie *et al.*, 2007; Malea *et al.*, 2013).

Mercury contamination of the marine environment has long been recognized as a severe environmental concern. It is commonly accepted that human activities have artificially increased mercury loads in the atmosphere on a local, regional and even hemispheric scale, leading to the contamination of the environment (Thompson *et al.*, 1992). The enhanced atmospheric deposition of mercury is often the dominant source of mercury to the aquatic systems, which may reflect in seagrass mercury concentrations (Lafabrie *et al.*, 2007).

Several studies show that *P. oceanica* is a good bioindicator of coastal water mercury (Hg) contamination and lepidochronology can be used to follow historical changes up in the environment (Pergent-Martini, 1998; Lafabrie *et al.*, 2007). The aim of this work was to conduct a survey of mercury concentration in *Posidonia oceanica* at El Kantaoui touristic harbour located in the eastern coast of Tunisia and to compare the level of contamination with other sites around the Mediterranean Sea.

## Materials and methods

Study was conducted in September 2012 in the front of port El Kantaoui using scuba diving at 10 m depth (Fig. 1). Forty-five orthotropic shoots of *P. oceanica* were collected and conserved in -20 °C until the dissection in the laboratory.



**Fig. 1: Map of the sampling area in El Kantaoui port (Eastern coast of Tunisia)**

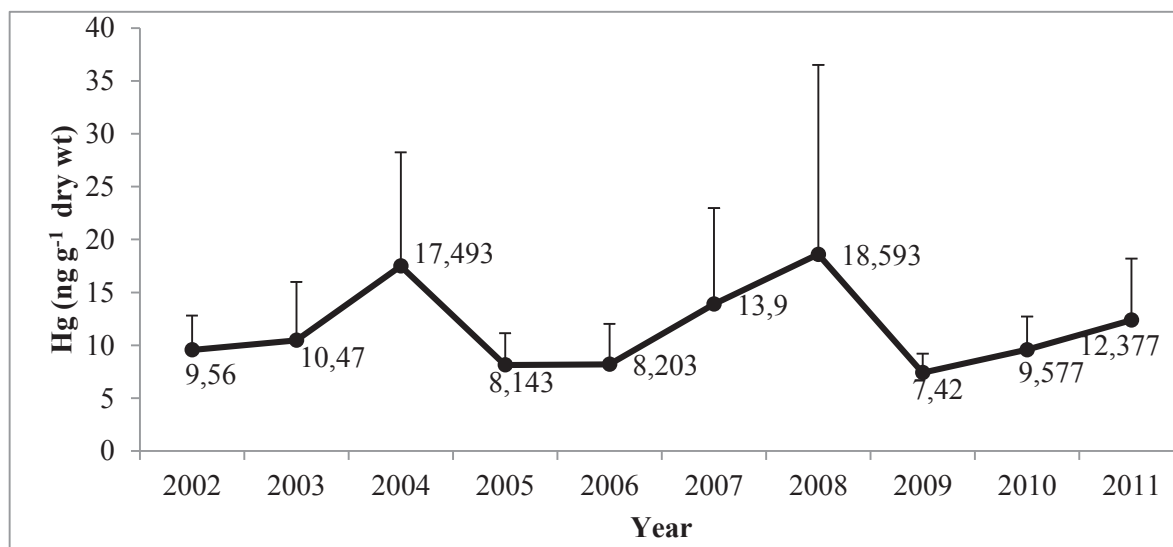
Three sub-samples of 15 shoots were considered for this study. For each pool of shoots, sheaths were carefully detached from the rhizome and ordered, according to their distical disposal, from the older (near the rhizome base) to the more recent (near the living leaves). The sheath thickness minima were identified during the dissection procedure to determine the limits of the annual cycles (Pergent, 1990). The pattern of the mercury concentration was studied over an annual cycle during the last decade (2002- 2011).

The sheaths of *Posidonia oceanica* were dried at 30°C for 48 h, milled to obtain an homogeneous sample. For Hg analyses, 500 mg of samples were placed in closed Teflon container to prevent any volatilisation of mercury (Matusiewicz *et al.*, 1989). Mineralisation was carried out using a mixture of 5ml of HNO<sub>3</sub> and 3ml of H<sub>2</sub>O<sub>2</sub>. The Teflon digestion vessels were sealed and placed in a microwave oven. Then, content was poured into 25ml volumetric flasks and diluted to volume with ultrapure water and then transferred to 60 ml polypropylene flasks. Mineralized samples were analysed using Inductively Coupled Plasma spectrometry (ICP-OES, model OPTIMA 2100 DV and PERKIN ELMER 8000).

One-way analysis of variance (ANOVA) was used to compare average concentration of mercury in the dead sheaths over years using XLSTAT software.

## Results

The annual variation of mercury concentration in the dead sheaths over a period from 2002 to 2011 is indicated in Fig. 2. Annual mean concentration varied between minima of  $7.42 \pm 1.79$  ng g<sup>-1</sup> dry wt in 2009 to a maximum of  $18.59 \pm 17.91$  ng g<sup>-1</sup> dry wt in 2008.



**Fig. 2: Temporal variation of the mercury concentration (ng g<sup>-1</sup> dry wt) in the dead sheaths of *Posidonia oceanica* in port El Kantaoui (mean ± SD)**

One-way ANOVA show that no significant difference on the mercury levels measured was observed between years.

The average mercury concentration of 10 years in El Kantaoui is compared with other studies around the Mediterranean Sea (Tab. 1). The mercury concentrations recorded in *P. oceanica* from El Kantaoui were lower than those of France and Italy.

**Tab. 1: Average mercury concentration of 10 years in different sites around the Mediterranean Sea**

References	Site	Mercury concentration
Pergent-Martini, 1998 Average of 10 years	Marseille-Coutiou (Bouches-du-Rhône, France)	127.5±44.1
	Calvi (Corsica, France)	16.0±3.4
	Island of Ischia (Bay of Naples, Italy)	24.9±3.6
Present study Average of 10 years	Port El Kantaoui (Tunisia)	11.57±3.9

### Discussion and conclusions

The present paper is the first study of mercury accumulation in sheaths of *P. oceanica* from El Kantaoui port. Mercury was present in detectable concentrations in the sheaths of *P. oceanica* during all years of the last decade in the studied area exhibiting lowest value versus the other sites in the Mediterranean Sea (Italy and France).

In the Mediterranean Sea, organic and trace metal pollution has increased over the last decades having a perceptible impact on marine environments (Kotnic et al., 2014). The seagrass *P. oceanica* is a sedentary species available in every season all over the Mediterranean coastal area and is easy to sample and identify. Consequently, *P. oceanica* has considerable potential as cosmopolitan biomonitors of trace metals in the Mediterranean. This seagrass can accumulate mercury from seawater in sheaths for a long period. This property allowed following the temporal change up of the mean mercury contamination of the environment (Pergent-Martini, 1998).

*P. oceanica* is located at the base of the food web in the Mediterranean and is probably the main source of metals for many animals grazing on its leaves. Therefore, the investigation of trace metal concentrations in the tissues of these species may provide useful information on the transfer of potentially toxic elements from abiotic compartments (water, sediments) to higher consumers, including man.

### Acknowledgments

The present study was funded by MAPMED project (I-B/2.1/395).

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# POSTERS

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## COMPOSITION OF THE FUNCTIONAL MACROALGAL GROUPS ON *POSIDONIA OCEANICA* (L.) DELILE LEAVES

### Abstract

This paper is designed to display the functional macroalgal groups classified according to their morphologies and the morpho-functional group numbers expressed as percentage on *Posidonia oceanica* leaf surface area (cm<sup>2</sup>) around Gökçeada Island (North Aegean Sea, Turkey) and finally interpret their relations with environmental factors.

**Key-words:** epiphytic functional macroalgal groups, *Posidonia oceanica*, North Aegean Sea

### Introduction

Ecological classifications of seaweeds came out from the recognition of the significance of their thallus morphology and life history (Orfanidis *et. al.*, 2001; Feldmann, 1951; Chapman & Chapman, 1976; Russel, 1977). To classify the marine coastal areas, marine algae and macrophytes can be used as a quality element because they respond directly to the abiotic and biotic changes in the aquatic environment (Orfanidis *et. al.*, 2001).

### Materials and methods

Three sampling locations from Gökçeada were selected between Spring 2009-Winter 2010 periods. *Posidonia oceanica* shoots were collected from the upper limits of the meadows for the identification of the epiflora and macroalgal species were classified according to their morphologies (Steneck & Dethier, 1994) then the number of morpho-functional macroalgal group numbers expressed as percentage on the leaf surface area per cm<sup>2</sup>.



**Fig. 1:** Sampling locations

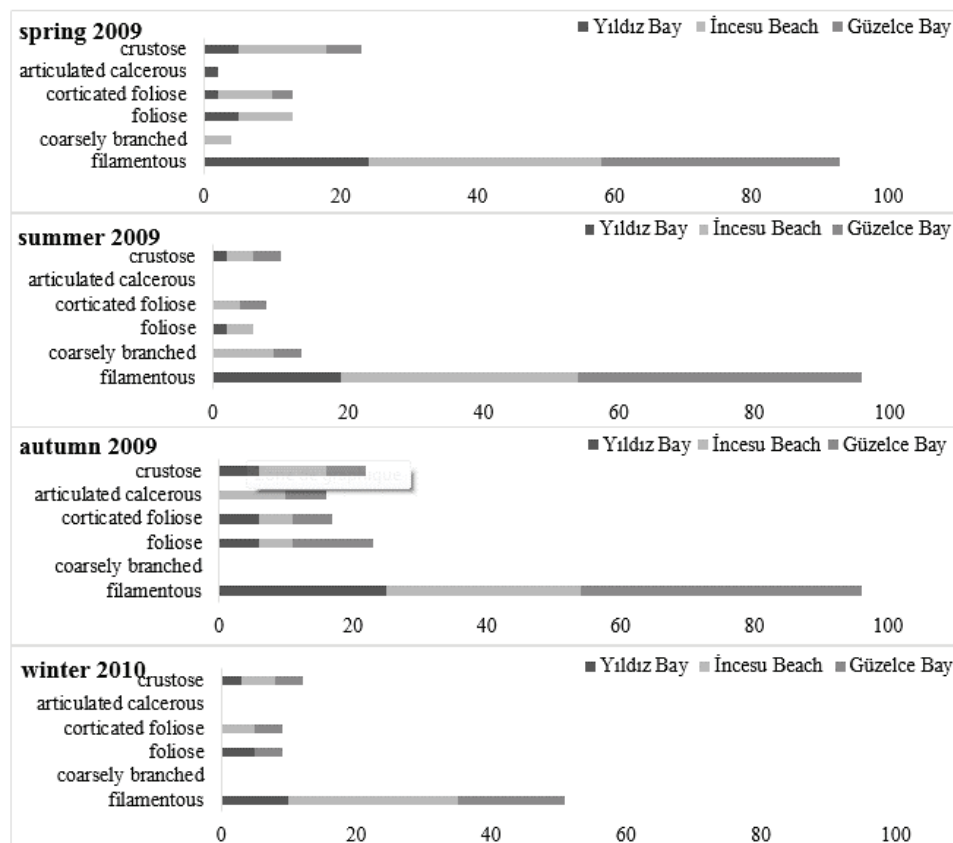
### Results

Filamentous species were dominant in all seasons especially settled in spring and showed maximum growth in summer. Foliose species followed them in all seasons but reached high coverage in autumn. Crustose species were more common on specific host species like seagrasses than others. The results of Kjøsterud (1997) and our study showed that crustose were found on leaves in every season. Therefore they supported the covering percentages of these groups (Tab. 1).

## Discussion and conclusions

Opportunistic species particularly involve filamentous and foliose groups and the level of these algal covers showed that the region has minor signs of disturbance whereas articulated calcereous species found in İncesu Beach showed that the south of the island is more pristine area. While observing environmental factors, morpho-functional epiphytic assemblages seemed appropriate to distinguish the water quality because when the epi-coverage rise due to water fluctuations, they cover high surface areas.

**Tab. 1: The seasonal variations of the morpho-functional epiphytic algal groups (percentages of species numbers (%) / cm<sup>2</sup> surface of leaves)**



## Acknowledgements

This work was supported by the Research Fund of Istanbul University (Project number 2149) and The Scientific and Technological Research Council of Turkey (Project number 109Y014).

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## **BIOACCUMULATION DU MERCURE, DU ZINC ET DU CUIVRE CHEZ *POSIDONIA OCEANICA* : COMPARAISON AVEC UN SITE DE REFERENCE**

### **Abstract**

*Les concentrations de Hg, Zn et Cu sont mesurées chez Posidonia oceanica provenant de deux sites situés dans la baie de Bou Ismaïl (Algérie, Sud de la Méditerranée). Les mesures effectuées dans différentes parties de la plante ont mis en évidence des teneurs importantes de ces trois métaux traces au niveau des rhizomes. Les limbes des feuilles adultes présentent des teneurs importantes de Hg et de Zn et les feuilles intermédiaires des teneurs importantes de Cu. La concentration en Hg est plus importante dans les échantillons issus du site anthropisé comparativement au site de référence, contrairement à celles du Zn et du Cu pour lesquelles l'écart est moins important.*

**Key-words:** Biomonitoring, Traces Metals, Bou Ismaïl, lepidochronology, *Posidonia oceanica*

### **Introduction**

*Posidonia oceanica* est utilisée depuis plus d'une décennie comme un bioindicateur de la contamination métallique du milieu marin (Pergent-Martini et Pergent, 2000; Lafabrie *et al.*, 2008). Dans ce cadre, pour la première fois et à un niveau local, les concentrations en Hg, Zn et Cu sont mesurées chez *P. oceanica*.

### **Matériel et méthodes**

45 faisceaux de *Posidonia oceanica* ont été prélevés tous les trois mois (entre mai 1996 et février 1997), à -10 m dans deux sites : Bou Ismaïl (BI), site pollué, et Kouali (AK), site de référence. Les feuilles sont séparées selon le protocole de Giraud (1979) : limbes des feuilles adultes (LFA), feuilles intermédiaires (FI) et pétioles. Les écailles et les rhizomes ont été obtenus par la méthode lepidochronologique (Pergent, 1990). Les échantillons ont été congelés, lyophilisés puis broyés. L'extraction des métaux a été réalisée selon le protocole de IAEA-MESL (1998) et le dosage par SAA. Les données ont été analysées en utilisant le test de Kruskal-Wallis et les résultats sont exprimés par la moyenne  $\pm$  S.E de trois répliquats pour chaque métal analysé.

### **Résultats et discussion**

Le Tab. 1 montre la distribution du Hg, Zn et Cu chez *Posidonia oceanica*. La répartition de ces métaux varie considérablement en fonction du tissu considéré. Concernant le Hg, les rhizomes présentent les concentrations les plus élevées, suivis des écailles, puis les LFA et les FI et enfin les pétioles et ce au sein des deux sites. Les concentrations en Hg dans les LFA et les FI sont équivalentes dans les deux sites. Dans les sites dont les niveaux d'anthropisation sont faibles, la différence entre les concentrations en Hg des LFA et des FI est moins marquée que dans les sites anthropisés, où les feuilles adultes, tissus plus âgés, présentent des valeurs plus élevées. Selon la grille d'évaluation proposée par Pergent (2007) pour les feuilles adultes, la qualité des eaux de AK est classée «bonne» (0,035 à 0,053  $\mu\text{g/g}$ ) et celle de BI «moyenne» (0,053 à 0,067  $\mu\text{g/g}$ ). Le rhizome représente également le tissu qui a concentré des taux importants de Zn et de Cu.

**Tab. 1. Concentration moyenne annuelle ( $\pm$  SE) du Hg, Zn et Cu ( $\mu\text{g/g}$  de poids sec) dans les différents tissus de *Posidonia oceanica* dans les deux sites.**

Tissus	Sites	
	AK	BI
<b>Hg</b>		
LFA	<b>0,04 <math>\pm</math> 0,01</b>	<b>0,06 <math>\pm</math> 0,01</b>
FI	<b>0,04 <math>\pm</math> 0,01</b>	<b>0,06 <math>\pm</math> 0,01</b>
Pétioles	<b>0,03 <math>\pm</math> 0,01</b>	<b>0,05 <math>\pm</math> 0,01</b>
Ecailles	<b>0,05 <math>\pm</math> 0,01</b>	<b>0,10 <math>\pm</math> 0,02</b>
Rhizomes	<b>0,07 <math>\pm</math> 0,01</b>	<b>0,11 <math>\pm</math> 0,03</b>
<b>Zn</b>		
LFA	<b>106,0 <math>\pm</math> 19,3</b>	<b>130,6 <math>\pm</math> 21,3</b>
FI	<b>90,0 <math>\pm</math> 15,5</b>	<b>108,5 <math>\pm</math> 13,5</b>
Pétioles	<b>21,1 <math>\pm</math> 3,2</b>	<b>25,5 <math>\pm</math> 3,9</b>
Ecailles	<b>22,0 <math>\pm</math> 3,2</b>	<b>31,2 <math>\pm</math> 3,3</b>
Rhizomes	<b>111,5 <math>\pm</math> 13,2</b>	<b>128,2 <math>\pm</math> 10,7</b>
<b>Cu</b>		
LFA	<b>8,1 <math>\pm</math> 1,0</b>	<b>8,7 <math>\pm</math> 0,9</b>
FI	<b>11,6 <math>\pm</math> 1,3</b>	<b>11,6 <math>\pm</math> 1,0</b>
Pétioles	<b>9,7 <math>\pm</math> 1,2</b>	<b>9,9 <math>\pm</math> 1,3</b>
Ecailles	<b>6,0 <math>\pm</math> 0,7</b>	<b>6,8 <math>\pm</math> 0,6</b>
Rhizomes	<b>11,3 <math>\pm</math> 2,1</b>	<b>12,3 <math>\pm</math> 2,5</b>

L'accumulation du Zn est également plus importante dans les LFA et les FI que dans les pétioles et les écailles. Au niveau de la partie épigée, les concentrations en Zn sont plus importantes au niveau des LFA. En revanche, le Cu semble s'accumuler préférentiellement dans les FI. La comparaison entre les deux sites montre que quel que soit le tissu considéré, les concentrations en Hg relevées à BI sont toujours supérieures à celles de AK (Kruskal-Wallis,  $p < 0,05$ ), en particulier au niveau des écailles et des rhizomes. Ces résultats confirment l'existence d'une corrélation entre les teneurs en Hg dans l'eau ou dans le sédiment et celles accumulées par la plante. En revanche, les teneurs en Zn et en Cu, métaux entrant dans le métabolisme de *P. oceanica*, n'ont montré de différence qu'au niveau des FI et des écailles pour le Zn et des écailles pour le Cu.

### Conclusion

Le rhizome semble constituer le réservoir d'accumulation préférentielle pour les trois métaux traces et pourrait donc constituer le marqueur privilégié dans le monitoring de contamination métallique en Méditerranée, notamment sur le long terme. L'évaluation du Hg dans les feuilles adultes a permis de conclure que les eaux du site de référence sont de «bonne» qualité, alors que celles du site anthropisé ont une qualité «moyenne».

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## **NEW DATA ABOUT THE PHENOLOGY AND THE DISTRIBUTION OF *DICTYOTA CYANOLOMA* TRONHOLM *ET AL.* IN THE MEDITERRANEAN IBERIAN COASTS**

### **Abstract**

*Dictyota cyanoloma* (Dictyotales, Phaeophyceae) was described by Tronholm *et al.* (2010). But little is known about the phenology and distribution of this species. Therefore, a detailed phenologic study was carried out in Palamós (Girona, Spain), where the type is from, and also a distribution study along the entire Mediterranean Iberian coast. In Palamós *D. cyanoloma* is present all the year with a maximum development in winter and a minimum in summer. Fertile specimens are found all the months except in summer (July and August). Sporophytes are dominant from January to June and gametophytes only occur in February, March and June. Information about the antheridia, which has never been described before, is provided. At present *D. cyanoloma* occurs along all the Mediterranean Iberian coasts, in 19 of 32 localities sampled. It is found mostly in harbours and the increase of its distribution suggests that *D. cyanoloma* could be an introduced species in the Mediterranean Sea.

**Key-words:** *Dictyota*, phenology, distribution, Iberian Peninsula, Mediterranean Sea

### **Introduction**

*Dictyota cyanoloma* Tronholm, De Clerck, Gómez Garreta *et* Rull Lluç was recently described by Tronholm *et al.* (2010) being the original description and some morphological data the unique information available until now. Therefore, the main aim of this study was to carry out a detailed phenologic study of this species, as well as to describe the antheridia, which were unknown up until today. So far, *D. cyanoloma* has been reported from Canary Islands, Madeira, Azores, Portugal, Adriatic Sea (Tronholm *et al.*, 2010) and Turkey (Taskin, 2013). Most of these citations correspond to harbours and surroundings. Hence, the other aim of this study was to survey the most important harbours of the Mediterranean coast of the Iberian Peninsula in order to know the present distribution of *D. cyanoloma* in our coasts.

### **Materials and methods**

To study the distribution of *D. cyanoloma* in the Mediterranean peninsular coasts, a total of 29 harbours were explored from Llançà (Girona) to Algeciras (Cádiz) mainly during winter 2013 and spring 2014. The presence or absence of *D. cyanoloma* in each harbour was assessed, collecting some specimens when the species was present. The phenological study was carried out in Palamós harbour (Girona). Sampling was monthly, from December 2012 to November 2013, collecting two samples of 10 x 10 cm each month. For each specimen of *D. cyanoloma* the thallus length, the number of dichotomies and the width of the third dichotomy were measured. The reproductive stage was also assessed, and the abundance of the specimens of each reproductive type was quantified (coverage in cm<sup>2</sup>).

## Results

At Palamós *D. cyanoloma* was observed throughout the year. In summer the specimens showed the lowest mean values in thallus length, number of dichotomies and with of the third dichotomy, as well as in total cover. The maximum mean values were found in winter-spring. Sterile specimens were found all the year except in February, March and April. Fertile sporophytes were also found almost all the year but July and August. Fertile gametophytes were found only in December (male), February (male and female) and June (female). Fertile plants (mainly sporophytes) were dominant from January to June, whereas the sterile ones dominated from July to December. In July and August all the specimens were sterile, while in April all were sporophytes. Antheridia are reported for the first time; they are grouped in ellipsoidal sori in both sides of the frond; sori measure 213-1085(1651) x 100-450(580)  $\mu\text{m}$ , are indusiate and surrounded by 3-4 (5) layers of unicellular paraphyses; there are 82-247 antheridia per sori; antheridia measure (31)40-78 x 10-31  $\mu\text{m}$  and they are supported by one stalk cell. Concerning the distribution of *D. cyanoloma* in the Mediterranean Iberian coasts, the species was found along the entire studied area, in 19 of the 32 harbours visited.

## Discussion

*D. cyanoloma* (as *D. ciliolata* Sonder) was firstly reported in the Mediterranean Sea by Rull Lluich *et al.* (2007), who raised the possibility that the species had been recently introduced to the area. The present expansion of this species on the Iberian coasts, as well as its localization mostly in harbours, also suggests that it could be an introduced species. Molecular studies will provide more information to reveal its origin. At Palamós *D. cyanoloma* occurs along all the year showing a clear seasonal cycle, with a maximum development in winter and minimum in summer. Reproductive phenology also follows this same pattern: fertile specimens dominating in winter while sterile ones were more abundant in summer. Other Mediterranean *Dictyota* species, such as *D. spiralis* and *D. dichotoma*, also occur along the year (Polo, 1978) but unlike *D. cyanoloma* they are more abundant in spring and summer, being usually fertile in the later period (Polo, 1978; Feldmann, 1937). Concerning antheridia, our data agree with those available for the other Mediterranean *Dictyota* species (Cormaci *et al.*, 2012).

## Acknowledgments:

Financial support was provided by Fundación Biodiversidad. We thank all the harbour authorities for sampling permissions.

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## RESEARCH OF CIRCULAR PATTERN DAMAGE TO *POSIDONIA* MEADOWS ALONG THE EASTERN ADRIATIC SEA

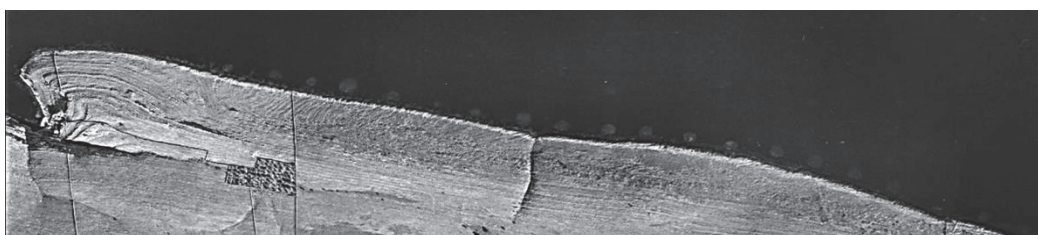
### Abstract

Remote aerial or satellite images of the Croatian Adriatic have revealed several locations of circular damage to *Posidonia* meadows extending back in time over a decade. In some locations these circles exist as a regular linear series on remote island shores, and at other locations they appear haphazardly positioned and sometimes overlapping. Our aims are to determine possible causes of this damage, obtain estimates of the time of damage, and to monitor the rate of *Posidonia* recovery. The damage circles are likely caused by submarine explosions, for example from blast fishing or military shelling. We expect that more than one type of human activity will be required to explain all the *Posidonia* damage circles in Croatia.

**Key-words:** circular pattern damage, submarine explosions, *Posidonia oceanica*, Croatia

### Introduction

*Posidonia oceanica* (L.) Delile meadows represent priority habitat according to the EU Habitats Directive, and they are legally protected in Croatia. They inhabit shallow coastal areas in which numerous human activities threaten them (Bakran-Petricioli, 2011). On aerial photos of the Croatian part of the Adriatic, we recorded several locations with a circular pattern damage to *Posidonia* meadows, some older than 10 years. At some locations the damage appears in regular series along the coast of islands, even in protected areas (Fig. 1); at others they are random and overlapping. We suspect that damage circles are likely caused by submarine explosions, for example from blast fishing or military shelling. The recovery of meadows after a bomb blast could take decades (Meinesz & Lefevre, 1984). The aims of our research are to determine possible causes of recorded damage, obtain estimates of the time of damage, and to monitor the rate of *Posidonia* recovery.



**Fig. 1:** The circular pattern damage to *Posidonia* meadow along the NE coast of Kornat Island, Kornati National Park, Croatia (from Bakran-Petricioli *et al.*, 2009).

### Materials and Methods

As a first step, we initiated an inventory of all locations of circular *Posidonia* damage in Croatia. For this purpose we used the publicly available service ARKOD Preglednik, established by the Croatian Ministry of Agriculture as a part of the Land Parcel Identification System, which contains 1:5000 resolution aerial orthophotos of the entire

Croatian coast, including subtidal areas to a depth of approximately 15-20 meters. From these images several characteristics of each visible damage circle, including diameter, area and depth of damage, distance to the nearest damage circle, to the shoreline, and to the nearest human settlement were measured. Also, slope of the bottom, aspect, and degree of exposure to wind and waves were recorded.

### Results

The first result is a geo-referenced index of all known *Posidonia* damage circles in Croatia and their most important features. Our second step, which we are currently working on, is a statistical analysis of the dataset to uncover patterns, including the correlation between features of the damage circles and the local socio-environmental context. The results of the analysis will be shown on the poster.

### Discussion and Conclusion

We assume that analysis will reveal that the size and spacing of damage circles will be related to the distance to human settlements and to the proximity of certain kinds of human activities, which will provide evidence of the causes of the damage (e.g. illegal blast fishing or war activities). The results of the second step will direct our third stage of study, namely *in situ* observation of the degree of damage and recovery in which we are planning to use boat based geo-referenced underwater videography along with diving surveys (Bakran-Petricioli *et al.*, 2011). We expect that more than one type of human activity will be required to explain all the *Posidonia* damage circles in Croatia.

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**APPLICABILITY OF MEDITERRANEAN BASELINES OF *POSIDONIA OCEANICA* DESCRIPTORS FOR THE ASSESSMENT OF ITS CONSERVATION STATUS ALONG THE EASTERN PART OF THE ADRIATIC SEA**

**Abstract**

*Ecological differences along the Adriatic Sea (climatic differences along the NW-SE axis, higher eutrophication in the North Adriatic, and higher natural eutrophication in the area between the islands or inside deep indented bays) influence *Posidonia oceanica* distribution and the appearance of its meadows. As the baselines of descriptors for *Posidonia* meadows for a particular area in the Croatian Adriatic have not yet been established we argue that non-critical and untested use of baselines of descriptors and classification methods that exist for *Posidonia* meadows in the NW Mediterranean could lead to erroneous conclusions and an incorrect assessment of meadow conservation status.*

**Key-words:** descriptor baselines, conservation status assessment, *Posidonia oceanica*, Croatia

**Introduction**

The Adriatic Sea is the northernmost part of the Mediterranean. There are notable ecological differences along the Adriatic such as climatic differences along the NW-SE axis, as well as higher eutrophication in the North Adriatic, and higher natural and human impacted eutrophication in the area between the islands or inside deep indented bays (for overview see IOF Split, 2012). Preliminary research indicated that those differences influence *Posidonia oceanica* distribution and the appearance of its meadows (Bakran-Petricioli & Schultz, 2010). Although *Posidonia* meadows are common along the Adriatic coast, their exact status is partially known and precise distribution maps are still missing. There are a number of different descriptors used for characterisation of Mediterranean *Posidonia* meadows (Pergent-Martini *et al.*, 2005) and there is a growing pressure to standardize mapping and monitoring methods across the whole Mediterranean (UNEP-RAC/SPA, 2011; Guala *et al.*, 2012; Guala *et al.*, 2014). Also, *Posidonia* conservation status classification has been proposed and it has been suggested that it should be used for the interpretation of monitoring data (UNEP-RAC/SPA, 2011).

**Materials and Methods**

In this contribution we compared available data for some generally and widely used descriptors like shoot density, meadow bathymetric extension, leaf biometry, and estimated primary production for the Adriatic (Bakran-Petricioli & Schultz, 2010; Bakran-Petricioli, unpublished data; Guala *et al.*, 2012; IOR Split, 2012) with proposed baselines for the Mediterranean (UNEP-RAC/SPA, 2011).

**Results**

The results (due to the lack of space the tables will be shown on the poster) show that, if the above baselines are applied for Croatia, even non-impacted meadows exhibit poor or



bad conservation status. This is especially true for the shoot density descriptor, number of shoots per m<sup>2</sup>, which is highly sensitive to natural eutrophication and/or other natural environmental stressors (Bakran-Petricioli & Schultz, 2010).

### Discussion

The baselines of descriptors for *Posidonia* meadows for a particular area in the Croatian Adriatic have not yet been established and, based on our previous research, we expect that they will be different from elsewhere in the Mediterranean. Non-critical and untested use of baselines of descriptors and classification methods that exist for *Posidonia* meadows in the NW Mediterranean (especially those including data from the southern Mediterranean; UNEP-RAC/SPA, 2011) could lead to erroneous conclusions and an incorrect assessment of meadow conservation status in the Croatian Adriatic. Similarly, the state of *Posidonia* meadows occupying open waters of the Adriatic (exposed to southeast, south, and southwest) cannot be used as a baseline target for all *Posidonia* beds in the Croatian Adriatic.

### Conclusion

The monitoring protocols developed for other Mediterranean countries should not be simply copied verbatim for use with Adriatic *Posidonia* meadows. The wide use of a method or descriptor in many or all Mediterranean countries does not imply that baselines are identical in every country, or even at every location within a single country. Such assumptions should be replaced with critical thinking and basic scientific field research that includes estimates of temporal trends of descriptors.

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## **VERS UNE NOUVELLE AIRE MARINE PROTEGEE MAROCAINE EN MEDITERRANEE : LE CAP DES TROIS FOURCHES**

### **Résumé**

*Dans le cadre du Projet MedMPAnet, une priorisation des Sites d'Intérêt Biologique et Ecologique (SIBE), préalablement identifiés sur la façade méditerranéenne du Maroc, a révélé que le site « Cap des Trois Fourches », est le mieux approprié pour être érigé en Aire Marine Protégée (AMP). Ceci a été confirmé par les prospections sous-marines, réalisées en 2012 et 2013. En effet, le site présente un excellent état environnemental et une grande valeur écologique liée à la présence de communautés bien structurées et de nombreuses espèces d'intérêt pour la conservation en Méditerranée. Il se caractérise également par une grande diversité d'habitats remarquables (coralligène, herbiers de phanérogames marines, maërl, grottes, etc.). La mise en place d'une AMP dans cette zone servira certainement à améliorer la connectivité entre les diverses AMPs en mer d'Alboran et à promouvoir l'économie locale dans une perspective de durabilité des ressources.*

**Key-words:** Cap des Trois Fourches, Morocco, Marine Protected Area, Mediterranean, conservation.

### **Introduction**

En Méditerranée marocaine, le Haut-Commissariat aux Eaux et Forêts et à la Lutte Contre la Désertification a collaboré, dans le cadre du Projet MedMPAnet, avec le Centre d'Activités Régionales pour les Aires Spécialement Protégées en vue d'une caractérisation des sites marins méritant d'être érigés en nouvelles Aires Marines Protégées (AMP). A l'issue d'une priorisation des Sites d'Intérêt Biologique et Ecologique, préalablement identifiés sur la façade méditerranéenne du Maroc, le Cap des Trois Fourches (35°26' N - 2°59' W) s'est révélé le site pilote le mieux approprié pour être érigé en future AMP. La présente étude expose les principaux résultats issus des prospections marines menées dans le cadre d'une caractérisation écologique visant à mettre en exergue les potentialités du site 'Cap des Trois Fourches' en tant que site d'intérêt de conservation méritant d'être érigé en future AMP.

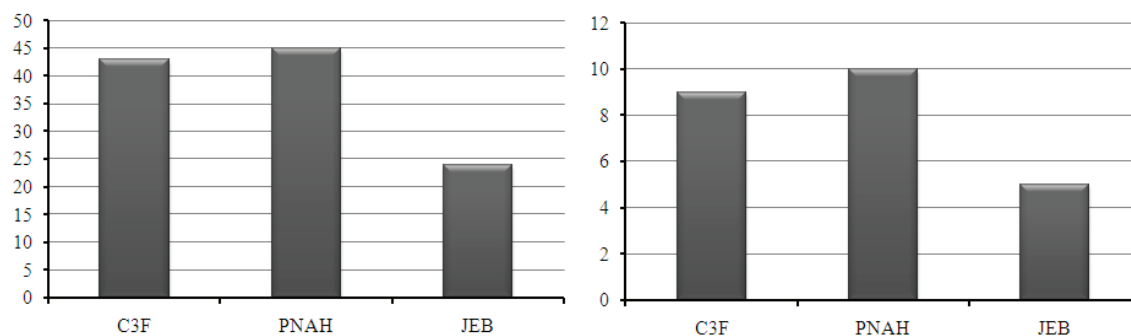
### **Matériels et méthodes**

Menées en septembre 2012 et 2013, les prospections sous-marines ont concerné un total de 95 stations. Elles ont été réalisées à l'aide de plongée en scaphandre autonome et/ou à l'aide d'un ROV. Les séquences vidéo et les photographies prises ont été analysées ultérieurement sur ordinateur en vue d'identifier les espèces présentes dans chaque point. Le type de communauté

dominante a été défini en utilisant la classification de Calvin-Calvo (2000) avec des adaptations au contexte local.

### Résultats et discussion

Les investigations sous-marines ont permis de recenser un total de 180 espèces et dix types de communautés benthiques (PNUE-CAP/ASP 2013a et b). L'évaluation de la valeur écologique du site, en considérant les espèces et les habitats d'intérêt pour la conservation en Méditerranée, confirme que le site mérite d'être érigé en AMP. En effet, 43 espèces et 9 habitats remarquables à l'échelle du bassin méditerranéen y ont été recensés (Fig. 1); ce qui confère au site une valeur écologique remarquable et en fait un site sensible d'intérêt pour la conservation en Méditerranée. Aussi, les valeurs de l'indicateur 'Etat de Conservation des espèces et habitats remarquables dans les AMP' (% des états favorables) sont de manière générale plus élevées au Cap des Trois Fourches (28% pour les espèces et 22% pour les habitats) qu'au Parc National d'Al Hoceima (14% pour les espèces et 8% pour les habitats) (PNUE-CAP/ASP 2013b).



**Fig.1 : Nombre d'espèces (à gauche) et d'habitats (à droite), d'intérêt pour la conservation en Méditerranée, recensés au Cap des Trois Fourches (C3F) et comparaison avec le Parc National d'Al Hoceima (PNAH) et le SIBE 'Cirque de Jebha' (JEB).**

Le Cap des Trois Fourches est un excellent 'site potentiel' qui présente toutes les qualités scientifiques pour être érigé en AMP, notamment en raison de sa grande qualité environnementale (présence de nombreuses espèces bio-indicatrices d'eaux propres) et de sa diversité biologique remarquable à l'échelle de la Méditerranée (espèces et habitat du protocole ASP/DB). La mise en place d'une AMP dans cette zone servira certainement à améliorer la connectivité entre les divers AMP en mer d'Alboran et à promouvoir l'économie locale dans une perspective de durabilité des ressources.

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## **THE COMMON PROCESS OF IDENTIFICATION OF ECOLOGICALLY OR BIOLOGICALLY SIGNIFICANT MARINE AREAS IN THE RAMOGE AREA – A PILOT INITIATIVE OF TRANSBOUNDARY COOPERATION**

### **Abstract**

*Integrated coastal zone management requires adequate information on the values characterizing a specific coastal area, starting from the identification of the possible presence of areas of high conservation value. In the framework of the RAMOGE Agreement it was decided to create an inventory of sites of particular ecological or biological interest. The objective of this paper is to present this pilot initiative describing the applied approach and criteria, and the results of the analysis. The sites identified and evaluated are 67: 35 for the Ligurian/Italian waters, 6 for Monaco and 25 for the French PACA region. A specific GIS layer was created and the application of the “kml” format allowed to visualize all the surfaces as well as the score assigned to the identified areas on Google Earth<sup>®</sup>.*

**Key-words:** EBSAs, Mediterranean, GIS.

### **Introduction**

The RAMOGE Agreement is a framework of scientific, technical, legal and administrative cooperation by which France, Monaco and Italy are taking actions towards integrated coastal zone management. One major aspect of this objective is the creation of an inventory of areas of high ecological or biological values, identified applying common criteria, considering that the Mediterranean Sea requires the management of environmental problems in an integrated and cooperative vision through transboundary cooperation. A specific Mediterranean CBD workshop for the identification of ecologically or biologically significant marine areas (EBSAs) (Malaga, 7-11 April 2014) estimated that the whole Northern part of the Western Mediterranean is hosting two wide EBSAs, the “North-Western Mediterranean Pelagic Ecosystems” and the “North-Western Mediterranean Benthic Ecosystems”, partially overlapped (UNEP/CBD, 2014). Starting from this general assessment, it was decided to create an inventory of the areas of high ecological or biological values, applying an approach similar to the one proposed by the CBD for the EBSAs (CBD, 2008), but better tailored to the RAMOGE spatial context.

### **Materials and methods**

The identification of the areas meeting scientific criteria for EBSAs and their evaluation were undertaken using the scientific available information on the ecological features and biological characteristics of the RAMOGE area, from Marseille to La Spezia, and more precisely, from the mouth of the Rhone to the mouth of the river Magra. The criteria identified to select and classify the areas, assuming that an area should meet several but not necessarily all the criteria, were the following: 1- Uniqueness or Rarity; 2- Ecological/Functional significance; 3-Importance for threatened/declining species and

habitats/biotopes; 4- Vulnerability, fragility, sensitivity or slow recovery; 5-High natural biological diversity; 6- Naturalness. For each Criterion a “4 level” ranking evaluation was applied (0= No specific information; 1 Low; 2 Medium; 3 High relevance).

The areas considered were: all the Natura 2000 Sites of Community Importance (SCIs) (mainly focusing on *Posidonia* meadows or coralligenous), other marine protected areas (MPAs), and other sites for which scientific information substantiate their biological and/or ecological importance such as nurseries areas, canyons, dark habitats etc.

## Results

Overall the sites identified and evaluated on the basis of the present analysis are: 35 for the Ligurian/Italian waters, 6 for Monaco and 25 for the PACA French region.

The Italian sites are mainly coastal SCIs and MPAs (27), followed by canyons (6), escarpments (1) and nursery areas (1). In Monaco, 2 sites are MPAs (*Posidonia* meadows and red coral), and the others sites were mainly identified considering the criteria of rarity, vulnerability, fragility, sensitivity and slow recovery. They are mostly coralligenous habitats (3) and a *Lithophyllum* “trottoir” (1).

The French sites are mainly MPAs and coastal. 2 sites are situated offshore to take into account the observations of the MEDSEACAN expedition (deep corals).

A specific Geographic Information System (GIS) layer was created and the application of the “kml” format allowed to visualize all the surfaces as well as the score assigned to the identified areas on Google Earth<sup>®</sup>.

## Discussion

The present pilot initiative allowed to create an early inventory of areas of high ecological or biological values in the RAMOGE zone, giving a first general view about their distribution and coverage in the three Countries, starting from the identification of common criteria and confirming that, in the Mediterranean sea, environmental topics must be managed with an integrated and cooperative vision through transboundary cooperation. Nevertheless the process of describing areas meeting the EBSA criteria is an open and ongoing process, this first result is particularly relevant both to support ICZM (EEA, 2010), and because it allows comparisons among the conservationist priorities identified in the different Countries. Moreover this product supports the prioritization of new research activities focusing on data sets useful to strengthen the available information. In Italy, for example, relevant and multidisciplinary information is available for SCIs and MPAs, while for the other selected sites, i.e. canyons and escarpments, new, additional information is needed. All these activities are fully in line with the implementation of the new Action Plan for the conservation of Habitats and Species associated with seamounts, underwater caves and canyons, aphotic engineering benthic invertebrates and chemo-synthetic phenomena, in the Mediterranean Sea (Dark Habitats Action Plan) adopted by the Contracting Parties to the Barcelona Convention in 2013.

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## **MONITORING *POSIDONIA OCEANICA* IN THE GULF OF IZMIR BETWEEN 2000 AND 2003: THE KARABURUN PENINSULA AS A REFERENCE AREA**

### **Abstract**

*The Gulf of İzmir is one of the best places on the coasts of the Aegean Sea to observe Posidonia beds, which are under pressure from many sources. The area shows a gradient of harmful factors from the outer to the inner gulf. The lower limit for beds rises from 33 m to 11 m in the mid-gulf, and finally to 8 m at the edge of the inner gulf. Density values also decline towards the inner gulf. A principal factor is domestic and industrial pollution: the inner gulf is in the centre of the city, and Posidonia beds have entirely disappeared from there. Fish farming has further increased the nutrient load, and together with mechanical damage caused by fishing activities has had a negative effect on plant health. The area, where plants are progressively healthier from the inner to the outer gulf, and particularly the Karaburun peninsula, which is least affected, is suitable as an area of protection and reference. The Karaburun peninsula can be a pioneering reference point for others to be chosen later, enabling comparison of Posidonia meadows in areas in the inner parts of the gulf and the whole Aegean coast.*

**Key-words:** *P. oceanica*, Biotic index, Water quality, Reference area, Karaburun

### **Introduction**

*P. oceanica* forms lush meadows in the Mediterranean to a depth of 50 m, and has ecological and economic importance. It is very sensitive to the pollution of coastal waters from human domestic and industrial sources, and is used as a bioindicator (Pergent *et al.*, 1995). Recently, *Posidonia* beds have played a role in obtaining a sound and speedy result as a “biological quality element” (BQE) when the Mediterranean coastal waters were determined to be “in good ecological status” until 2015 under the water policy adopted by the EU nations in 2000. It was felt that the BiPo index was the most practical and soundest method for this purpose among several developed, and so this was used to determine the distribution of *Posidonia* beds in the Gulf of İzmir to a depth of 33 m in order to evaluate their state of health and thus to estimate the water quality of the gulf.

### **Material and methods**

All *P. oceanica* meadows in the Gulf of İzmir were studied: distributions were shown with 52 transects by scuba and snorkel dives. In evaluating data, the basic descriptors (lower limit, lower limit type, shoot density, meadow cover and leaf phenology) were used, and to estimate water body quality, BiPo (Lopez y Royo *et al.*, 2010) was developed to evaluate the status of coastal waters based on the *P. oceanica* BQE.

### **Results and Discussion**

*Posidonia* meadows in the Gulf of İzmir were studied in four zones in 2000-2003. Tab. 1 shows water quality values of the gulf according to the condition of the *Posidonia* population: domestic and industrial pollution in the inner gulf and secondary threats in the middle gulf such as sedimentation, fishing, shipping traffic and fish farming lead to

low EQ values. In zone 3, health hier plants are found, but this area is under heavy pressure from the large numbers of fish farms set up along the coast since 2000. Although zone 4 is open sea, it is affected by a large human population and intense tourism activities, so that the EQ values of the area are 'good' or 'moderate'. Only in zone 2 do lower limit and density values indicate a healthy plant population, supported by EQ values. In 2000-2003 according to the basic descriptors, the water quality in the gulf showed a gradient. Today according to the WFD, following work to improve water quality, there is a need to compare the various indices which have been developed on *Posidonia* descriptors which have been accepted since 2000 as BQE. In this study, the BiPo index was preferred as it largely agreed with the water quality estimates of the last decade. These results strengthen the case for the Karaburun stations to be chosen as a reference area in the Aegean Sea, and show that there would be great benefit from taking new samples and comparing them with previous findings.

**Tab.1: Lower limits and plant density (shoots m<sup>2</sup>) of *Posidonia oceanica* at stations in the Gulf of Izmir, and variations in the Ecological Quality Ratio of the water body found by the European Water Framework Directive.**

	ZONE 1							ZONE 3			
	Asıklar Çeşmesi	Hastane Island	Urla Iskele	Pirnalı Island	Yassica Island	Esek Island	Hekim Island	Kumburnu	Mentes	Inceburun	Dalyanburnu
Density (shoot.m <sup>-2</sup> )	96	112	368	176	112	208	352	240	272	144	416
Lower limit (m)	8.5	12	13	13.1	13.8	15.7	17.5	17	18	19	22.4
EQR	0.25	0.225	0.433	0.318	0.274	0.296	0.484	0.421	0.516	0.464	0.470
Class	Poor	Poor	Moderate	Poor	Poor	Poor	Moderate	Moderate	Moderate	Moderate	Moderate

	ZONE 4							ZONE 2					
	Orak Döküntü	Fenerburnu	Fener (1)	Hayırsız (1)	Fener (2)	Hayırsız (2)	Orak Island	Büyükada	Akburun	Burgaz	Domuzlukaya	Kanlıkaya	Değirmentepe
Density (shoot.m <sup>-2</sup> )	464	176	352	304	432	560	480	768	416	528	288	400	832
Lower limit (m)	13	14.4	16.7	17.3	24.6	25.5	26.3	31.2	31.7	32	32.3	32.6	32.7
EQR	0.586	0.458	0.601	0.539	0.633	0.700	0.680	0.854	0.769	0.797	0.808	0.778	0.881
Class	Good	Moderate	Good	Moderate	Good	Good	Good	High	Good	High	High	High	High

### Acknowledgements

This study is a part of a project funded by TUBITAK project number 197T032. Thanks to Gerard Pergent for his ideas and suggestions, and to Alec Rylands for translating the paper.

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## **CONTRIBUTION A L'ETUDE DES CRUSTACES PERACARIDES ASSOCIES A L'ALGUE *CORALLINA ELONGATA* ET AUX HERBIERS DE *CYMODOCEA NODOSA* AU CAP DES TROIS FOURCHES (MAROC, MEDITERRANEE)**

### **Résumé**

*L'hétérogénéité spatiale des habitats de l'algue *Corallina elongata* et des herbiers de *Cymodocea nodosa* au niveau du Cap des Trois Fourches a été approchée par une analyse des assemblages des crustacés pércarides. Les habitats de *C. elongata* du secteur Nord du Cap se sont révélés plus riches et mieux structurés que ceux du secteur Ouest ; probablement en raison de l'hydrodynamisme différentiel entre ces deux secteurs. Les deux herbiers de *C. nodosa* prospectés dans le secteur Ouest du Cap montrent une forte hétérogénéité, qui se traduit plutôt par un assemblage différentiel des espèces que par leur nombre et abondance. Les patrons de distribution des pércarides dans les deux habitats végétalisés considérés ici révèlent une hétérogénéité spatiale remarquable qui doit être prise en considération dans toute perspective de conservation du site.*

**Mots-clés :** Cap des Trois Fourches, *Corallina elongata*, *Cymodocea nodosa*, Percarid.

### **Introduction**

Les Crustacés Pércarides représentent un groupe très diversifié, colonisant une grande variété de substrats (algues, herbiers, bryozoaires, etc.). Ces organismes macrobenthiques sont considérés comme des bioindicateurs de la qualité et de l'état de santé du milieu marin (Guerra-Garcia *et al.*, 2009). Ce travail, réalisé dans le cadre du Projet MedMPAnet, se propose de contribuer à l'évaluation de l'hétérogénéité spatiale des habitats à *Corallina elongata* et des herbiers à *Cymodocea nodosa*, et ce par l'analyse des assemblages des pércarides associés à ces habitats.

### **Matériels et méthodes**

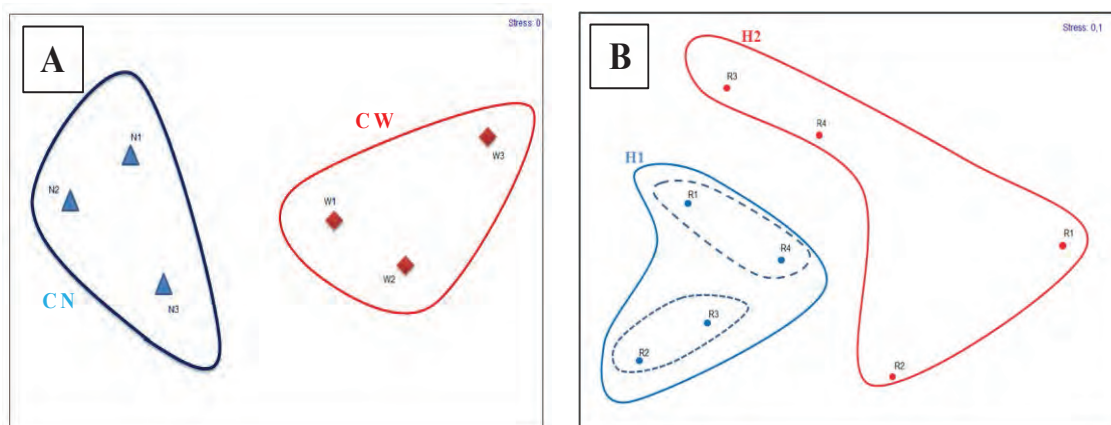
Les prospections de terrain ont été réalisées en septembre 2012. L'échantillonnage de l'algue *C. elongata* (grattage intégral jusqu'à la roche) a été réalisé par plongée libre et a concerné les secteurs Nord et Ouest du Cap des Trois Fourches (Trois stations par secteur). Les deux herbiers de *C. nodosa*, localisés à l'Ouest du Cap à des profondeurs autour de 12 m, ont été prospectés par scaphandre autonome et les prélèvements (quatre stations par herbier) ont été effectués à l'aide d'un carottier manuel, de 12,5 cm de diamètre, jusqu'à une profondeur de 20 cm.

Dans les deux cas, les prélèvements ont été tamisés sur place en utilisant un tamis de 0,5 mm de vide de maille et conservés dans l'alcool. Au laboratoire, les espèces de pércarides ont été identifiées et comptées. L'hétérogénéité des assemblages de pércarides a été approchée par des analyses univariées (richesse spécifique, abondance,

indices de diversité) et par des analyses multivariées (classification ascendante hiérarchique (CAH), analyse de proximité (MDS)). Ces analyses ont été réalisées à l'aide du logiciel PRIMER® (Clarke & Gorley, 2003).

### Résultats et discussion

L'analyse des assemblages de pécaricides associés à l'algue *C. elongata* a permis de recenser un total de 21 espèces (9 Gammaridés, 5 Caprellidés, 6 Isopodes et 1 Tanaïdacé). L'isopode *Coxicerberus remaniest* est signalé ici pour la première fois au Maroc. Les analyses multivariées ont permis d'identifier deux assemblages, le premier regroupant les prélèvements effectués dans la station Nord, et le second correspond à ceux effectués dans l'Ouest du Cap (Fig. 1-A). La station Nord du Cap s'est révélée plus riche et mieux structurée que la station Ouest. Cette hétérogénéité peut être liée à l'hydrodynamisme différentiel entre les deux zones. Par ailleurs, cette faune s'apparente fortement à celle observée dans des habitats similaires du Déroit de Gibraltar (Guerra-Garcia *et al.*, 2009).



**Fig. 1 : Ordination en deux dimensions (analyse de proximité MDS) des assemblages de pécaricides associés à *C. elongata* (A) et à *C. nodosa* (B) au Cap des Trois Fourches. Les traits délimitent les groupes identifiés par la classification automatique hiérarchique. C : *Corallina elongata* (W : secteur ouest, N : secteur Nord) ; H : Herbiers à *Cymodocea nodosa*.**

L'étude des deux herbiers de *C. nodosa* de la partie Ouest du Cap a permis de recenser un total de 41 espèces dont cinq sont endémiques de la Méditerranée. Les analyses univariées n'ont pas montré de différences significatives entre les communautés des deux prairies alors que les analyses multivariées ont montré une forte hétérogénéité au niveau de chaque prairie (Fig. 1-B). L'hétérogénéité observée se traduit plutôt par l'assemblage différentiel des espèces que par leur nombre et abondance.

En définitif, les patrons de distribution des pécaricides dans les deux habitats végétalisés étudiés révèlent une hétérogénéité spatiale remarquable qui doit être prise en considération dans toute perspective de conservation du site.

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## **DESIGNING AND ESTABLISHING OF MPA IN SALLUM AREA (SOCIOECONOMIC PERSPECTIVE)**

### **Abstract**

*The conservation of the western Mediterranean coast of Egypt with its natural resources has been recognized as a top priority on the biodiversity conservation agenda of Egypt. The utilization of a site based conservation approach, such as protected areas, is the most appropriate means to conserve the natural heritage of the Egyptian western Mediterranean coast. UNEP RAC/SPA has identified 26 priority sites for protection on the North African coast, of which the Gulf of Sallum, Egypt (31°31'N, 25°10'E) is one. The Gulf of Sallum supports a wide range of ecosystems, from the rich sea grass meadows and rocky reefs of the coastal zone, to little seamounts. It is very essential in designating a Marine Protected Area (MPA) to understand the socioeconomic conditions of the site and disseminate the basic information and knowledge of marine conservation among the local population in order to ensure the participation of all stakeholders in the process and to enhance the future support of the local population at the new marine area management. An Egyptian NGO, in collaboration with IUCN-Med had undertaken an awareness campaign in the Gulf of Sallum. They initiated a public awareness and information dissemination campaign in order to sensitize local people and relevant authorities of Sallum to the potential importance of marine protected areas. The target audiences in these events were not only fishermen, but government representatives who may not have a full appreciation of impacts of damaging practices on the area. To achieve the goals of this awareness campaign, meetings with the local populations and governmental officials were arranged, in addition to delivering presentations and ecologically friendly materials to school students, fishermen and officials.*

**Key-words:** Marine Protected Areas, Sallum, Mediterranean Sea, Egypt, Awareness.

The conservation of the western Mediterranean coast of Egypt with its natural resources has been recognized as a top priority on the biodiversity conservation agenda of Egypt. There was also a great urgency to take effective conservation measures, as biodiversity resources are rapidly being degraded and destroyed because of the intensive development pressure in this region. The utilization of a site based conservation approach, such as protected areas, is the most appropriate means to conserve the natural heritage of the Egyptian western Mediterranean coast. This has been proposed and supported by international and national organizations. UNEP RAC/SPA has identified 26 priority sites for protection on the North African coast, of which the Gulf of Sallum, Egypt (31°31'N, 25°10'E) is one.

The Sallum area is a part of the western Mediterranean coastal region of Egypt, located about 600 km from Alexandria in the East and the Libyan border in the West. Near the town of Sallum, the shoreline changes its east-west facet, to a northerly direction in a rather sharp bend, forming the Gulf of Sallum. The Gulf of Sallum supports a wide range of ecosystems, from the rich sea grass meadows and rocky reefs of the coastal zone, to little seamounts, Sea cliffs are formed further north.

From the national standpoint, Sallum area is especially suitable for protection and of high conservation priority, not only because of its high natural value, but also because it is



nearly the only section of the western Mediterranean coast of Egypt remaining fairly intact.

Stress factors causing sharp decline in resource quality include the following: Insufficient regulatory enforcement, over-harvesting (trawlers), fishing during breeding season, destructive fishing techniques, and urban pollution. Many steps were done (scientifically, socially and economically) to ensure the provision of proper protection of wildlife resources that exist in the region, for the benefit of local populations and the nation at large. These efforts ended with the declaration of the Gulf of Sallum as the first marine protected area in the Mediterranean Sea of Egypt in 2010 by the Egyptian Prime Minister's decision No. 533.

Some of the initial steps in designating a Marine Protected Area (MPA) are to:

- Understand the socioeconomic aspects of the site and develop marine managed zones that take these issues into consideration.
- Disseminate the basic knowledge of marine conservation among the local population to ensure the participation of stakeholders in the process and to enhance future support of local population for the new MPA management.

For achieving this aim, one of Egypt's non-governmental organizations (NGO), in collaboration with IUCN Centre for Mediterranean Cooperation conducted an awareness campaign in Sallum about "Protected Areas". They initiated a public awareness campaign targeting the local people and relevant authorities of Sallum and Marsa Matrouh to the importance of marine protected areas. The target audiences in these events were not only fishermen, but government representatives who may not have a full appreciation of actual impacts of damaging practices on the area. It is important to repeat things that the fishermen already know, because the coast guard and/or other local government bodies may not know. In pursuit of this approach the following was conducted:

- Undertake a reconnaissance visit to Sallum to familiarize with the project area and meet head of the Fishermen's Association, in addition to the head of the city council to introduce and plan awareness raising events.
- Meet with head of the local school board, followed by meetings with school principals to discuss potential activities and materials for both teachers and students to introduce the idea of Nature Day.
- With input from Dr. Yasser Geneid of the National Institute for Oceanography and Fisheries, produce a PowerPoint presentation, explaining the importance of MPAs and why Sallum, to be used during the event.
- Select and design appropriate awareness raising materials tailored to members of Fishermen's Association and relevant government stakeholders.
- Select and design, with input from Dr. Yasser and graphic company, appropriate awareness raising materials tailored to school teachers and children.
- Regroup with stakeholders to receive input on materials.
- Hold awareness raising event with local fishermen and selected stakeholders and distribute materials.

Recommended guest list for these events were:

- Members of the Fishermen's Association, with their head (sheikh el sayadeen).
- Head of the City Council / Matrouh and local government representatives.
- Representatives from the local Bedouin community of the region.
- Representatives of Coast Guard officials.
- Representatives of Egyptian Environmental Affairs Agency.
- Head of School Board and selected school principals.

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## **DOES *ZOSTERA MARINA* EXIST IN GREECE?**

### **Abstract**

*The occurrence of the seagrass Zostera marina Linnaeus (Alismatales, Angiospermae) in Greece is obscure. Based on a detailed review on Greek marine vegetation literature, we detected 14 Z. marina records from the Greek coasts. 10 of them are dating from the 19<sup>th</sup> century until the 1950s, but all lacking a detailed description of the species. Since the mid 20<sup>th</sup> century, records of Z. marina found in Greece decline (4 records in total but none of them confirmed), despite the higher scientific effort. In conclusion, we state that two scenarios are possible: a) either Z. marina records from Greece are actually misidentifications of another seagrass species or b) the species existed until the 1950s and its population drastically decline since then, possibly due to a epidemic disease effect, similar with that reported in Northern Europe populations during that time. Since no herbarium specimens of Z. marina from Greece exist it will remain unknown if the species ever occurred in the Greek seas. However, we cannot exclude the possibility that Z. marina might occur locally through very small populations even today in unexplored areas of the Greek coasts.*

**Key-words:** seagrasses, Mediterranean, marine angiosperm

### **Introduction**

Despite commonly found in North European shores, the seagrass *Zostera marina* Linnaeus is very rare in the Mediterranean Sea, presenting extremely local populations in the northern limits of the basin, such as the northern Adriatic Sea (Curiel *et al.*, 1997), while its occurrence in the Eastern Mediterranean, including Greece, remains obscure. The present study aims to address the possible occurrence of *Z. marina* along the Greek coasts.

### **Materials and methods**

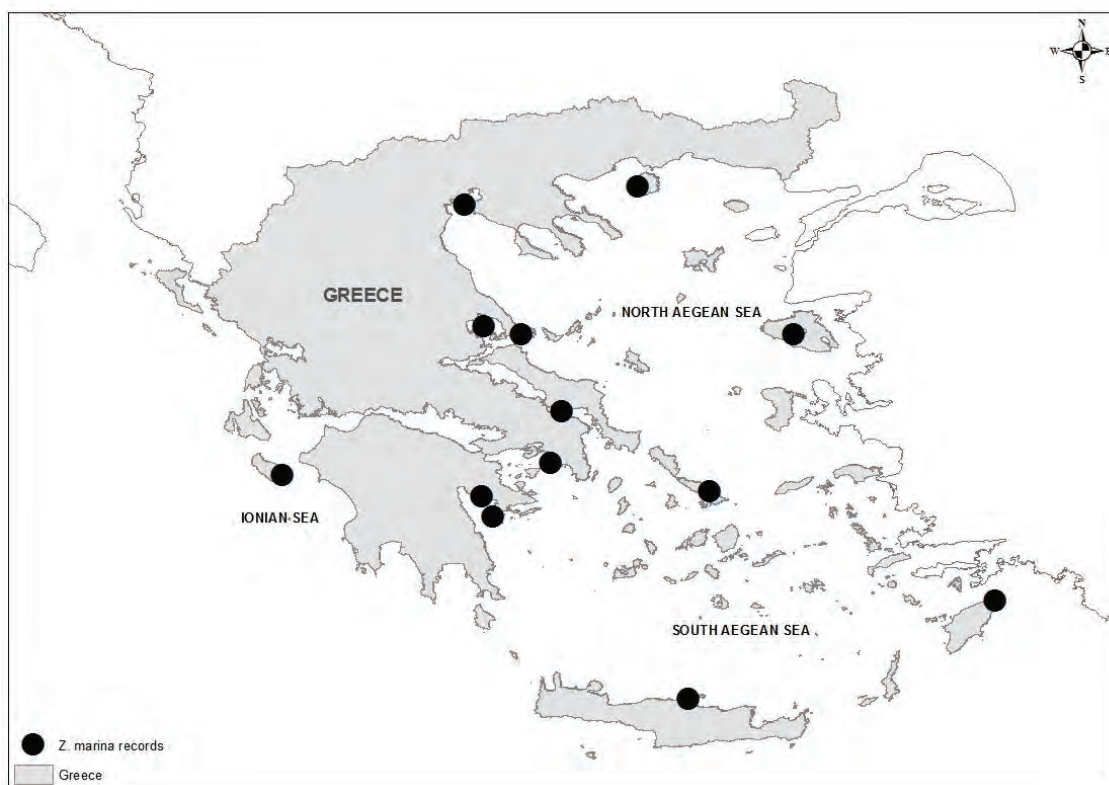
A detailed review on marine vegetation literature from Greece was carried out in order to detect and evaluate *Z. marina* records originating from the Greek coasts, from the early 19<sup>th</sup> century until today.

### **Results**

14 *Z. marina* records from the Greek coasts were detected (Fig. 1). The species was first reported in 1832 from Peloponnesus (Bory 1832), followed by several other records from various locations, most of them dating from the early 19<sup>th</sup> century until the 1950s (10 records in total). However, all of them lack a detailed description of the species and we state the high possibility of misidentification with another seagrass species sharing similar morphology, e.g. *Cymodocea nodosa*. Nevertheless, many of the references mentioning *Z. marina* also cite other Mediterranean seagrass species (*C. nodosa*, *Posidonia oceanica*, *Zostera noltei*). Since the mid 20<sup>th</sup> century, records of *Z. marina* found in Greece decline (4 records in total and none of them confirmed), despite the higher number of scientific efforts.

### Discussion and conclusions

Two scenarios are possible: a) either *Z. marina* records from Greece are actually misidentifications of another seagrass species or b) the species existed until the 1950s and its population drastically decline since then, possibly due to an epidemic disease effect, similar with that reported in Northern Europe populations during that time (1930s). Since no herbarium specimens of *Z. marina* from Greece exist it will remain unknown if the species ever occurred in the Greek seas. However, we cannot exclude the possibility that *Z. marina* might occur locally through very small populations even today in unexplored areas of the Greek coasts.



**Fig 1: *Zostera marina* records from the Greek coasts. Due to space limitation, corresponding references are not provided but they are available from the authors on request.**

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## TRACE ELEMENT BIOACCUMULATION AND COMPARTMENTALIZATION IN THE INVASIVE ALGAE *CAULERPA RACEMOSA* VAR. *CYLINDRACEA* FROM THE CALVI BAY, CORSICA

### Abstract

Very little is known regarding *Caulerpa racemosa* biochemistry and its natural ability to bioaccumulate trace elements (TEs). To fill this lack of knowledge, *Caulerpa racemosa* var. *cylindracea* (Sonder) were sampled in August 2011 along two depth gradients in the Calvi Bay (northwestern Corsica). 19 TEs were measured in the main compartments of the algae, i.e. fronds, stolons and rhizoids. TE bioaccumulation in *C. racemosa* ranged from  $10^{-3}$  to  $10^2 \mu\text{g g}_{\text{DW}}^{-1}$ , depending on the element. In accordance with the algae physiology (coenocytic algae), similar TE levels were measured in fronds and stolons. However, all TEs except Cd and As displayed higher concentrations in buried rhizoids. Some anthropogenic disturbances along one of the investigated depth gradient, i.e. the discharge of Calvi city Fe-treated wastewater and the presence of a the wreckage of a B17 aircraft, could explain the Fe and Bi level increases in the algae compartments with increasing depth. Thus, this study showed that poorly studied TE bioaccumulation and compartmentalization processes in *C. racemosa* var. *cylindracea* are under the influence of natural and anthropogenic factors that require further investigations.

**Key-words:** *Caulerpa racemosa* var. *cylindracea*, trace element, bioaccumulation, compartmentalization

### Introduction

The algae *Caulerpa racemosa* var. *cylindracea* (Sonder) was first observed along Corsican coasts in 2002 (Ruitton *et al.*, 2005), and more recently in the Calvi Bay (northwestern Corsica) in 2008. Numerous works have studied the distribution of that invasive species and the factors driving its rapid expansion, as well as the resulting ecological impacts on native coastal communities (Klein and Verlaque, 2008). However, very little is known regarding *C. racemosa* biochemistry and its natural ability to concentrate trace elements (TEs). In the framework of the STARECAPMED project, this work therefore aimed to study the bioaccumulation and the compartmentalization of 19 TEs in this macroalgae.

### Material and methods

*Caulerpa racemosa* var. *cylindracea* were sampled by SCUBA diving in August 2011 along two depth gradients in the little anthropized Calvi Bay: station B17 (at 10, 15, 20, 22 and 28 m depth) near Calvi city and station Punta Bianca (at 18 and 42 m depth) at the western limit of the Bay. Collected algae were sorted in fronds, stolons and rhizoids for compartmentalization analysis, lyophilised, and ground to powder. 19 TEs: Al, V, Fe, Cr, Mn, Co, Ni, Cu, Zn, Se, Ag, Cd, Sn, Sb, Mo, As, Be, Pb and Bi, were measured by DRC-ICP-MS in the algae compartments following acid digestion ( $\text{HNO}_3/\text{H}_2\text{O}_2$ ). Analytical accuracy was checked through the analysis of certified reference materials.

## Results

Median TE concentrations measured in the compartments of *C. racemosa* var. *cylindracea* sampled along the two depth gradients in the Calvi Bay are given in Tab. 1.

**Tab. 1: Trace element median concentrations ( $\mu\text{g g}_{\text{DW}}^{-1}$ ) in fronds (n = 3-10; N=38), stolons (n = 4-10; N = 44) and rhizoids (n = 1-4; N=20) of *Caulerpa racemosa* var. *cylindracea* sampled in August 2011 along two depth gradients (total number of sampling depths = 7) in the Calvi Bay. \*, \*\* and \*\*\* values represent Se and/or Be concentrations below the quantification limit, the detection limit or the detection decision, respectively.**

	Al	V	Fe	Cr	Mn	Co	Ni	Cu	Zn	Se
fronds	47	1.65	79	0.140	7.45	0.118	0.99	2.13	20.0	0.114**
stolons	75	2.32	91	0.184	6.68	0.105	1.20	2.49	25.2	0.135*
rhizoids	479	3.20	354	0.910	8.85	0.131	1.58	4.26	28.5	0.187**
	Ag	Cd	Sn	Sb	Mo	As	Be	Pb	Bi	
fronds	0.158	0.151	0.058	0.0152	0.181	7.44	0.0049***	1.44	0.0054	
stolons	0.178	0.132	0.053	0.0166	0.222	6.29	0.0050**	1.77	0.0058	
rhizoids	0.219	0.053	0.201	0.0421	0.560	3.63	0.0326*	4.76	0.0219	

## Discussion and conclusion

To our knowledge, this is the first study on the bioaccumulation of 19 TEs in the invasive algae *Caulerpa racemosa* var. *cylindracea* in the northwestern Mediterranean. TE concentrations ranged from  $10^{-3} \mu\text{g g}_{\text{DW}}^{-1}$  for Be and Bi in fronds and stolons to  $10^2 \mu\text{g g}_{\text{DW}}^{-1}$  for Al and Fe in rhizoids (Tab. 1). In accordance with the algae physiology (coenocytic algae), similar TE levels were measured in fronds and stolons. However, since all TEs except Cd and As displayed higher concentrations in buried rhizoids, this part of the algae could be regarded as the preferential route of TE uptake from the sediment and/or as a storage compartment (Richir *et al.*, 2013). Only Fe, used as a chemical reagent in the treatment of wastewater, and Bi, a urban sewage tracer, showed in station B17 a clear increase of their concentrations with increasing depth in the algae 3 compartments (data not shown). This could result from the discharge of Calvi city Fe-treated wastewater at 40 m depth in the vicinity of that station and to the presence of the wreckage of a B17 aircraft along that transect at 27 m depth. In conclusion, this study showed that bioaccumulation processes in *C. racemosa* var. *cylindracea* are under the influence of natural and anthropogenic factors that require further investigations.

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## **ON TWO *PADINA* SPECIES UNNOTICED IN THE BENTHIC MARINE FLORA OF THE IBERIAN PENINSULA**

### **Abstract**

*Padina distristromatica* Ni-Ni-Win & H. Kawai and *Padina pavonicoides* Ni-Ni-Win & H. Kawai, two species of Dictyotales (Phaeophyceae) recently described from the Mediterranean Sea by Ni-Ni-Win et al. (2011) have been found in the coasts of the Iberian Peninsula and the former also in the Balearic Islands. In this paper new morpho-anatomic details of both vegetative and reproductive structures of these species are provided. Among them it can be noticed the longitudinal aperture of the indusium in the sporangial sori of *P. distristromatica*, as well as the lack of stoloniferous proliferations at the base of the specimens of this species. Both features are useful for distinguish *P. distristromatica* from *P. pavonica*, the widest distributed *Padina* species in the Mediterranean Sea, and they are proposed as new taxonomical characters for *P. distristromatica*.

**Key-words:** *Padina*, Dictyotales, Phaeophyceae, Iberian Peninsula, Mediterranean Sea

### **Introduction**

Recently, Ni-Ni-Win et al. (2011) described *P. distristromatica* Ni-Ni-Win & H. Kawai and *P. pavonicoides* Ni-Ni-Win & H. Kawai as two new species for the Mediterranean. Until then, several *Padina* species had been reported from this area (Ribera et al., 1992): *P. boergesenii* Allender & Kraft, *P. boryana* Thivy, *P. gymnospora* (Kützinger) Sonder, *P. tenuis* Bory de Saint-Vincent and *P. pavonica* (Linnaeus) Thivy. However, Ni-Ni-Win et al. (2011) and Cormaci et al. (2012) consider that most of these records correspond to misidentifications with *P. pavonica* and consequently, they propose that only 3 *Padina* species occur at present in this area: *P. pavonica*, *P. pavonicoides* and *P. distristromatica*. In the framework of the *Flora phycologica iberica* project, we revised several herbarium specimens of *P. pavonica* from the Spanish Mediterranean coasts in order to know if any of the two recently described species had passed unnoticed.

### **Material and Methods**

The study was carried out on the basis of herbarium specimens from the Mediterranean coast of the Iberian Peninsula (Girona to Cádiz) and the Balearic Islands held in the BCN-Phyc Herbarium (Centre de Documentació de Biodiversitat Vegetal de la Universitat de Barcelona). For the anatomical study of the specimens, frond sections were made by hand with a razor blade.

### **Results**

Of the 142 herbarium samples studied, 111 corresponded to *P. pavonica*, 28 to *P. distristromatica*, and 3 to *P. pavonicoides*.

Specimens of *P. distristromatica* consists of erect, flabelliform to semicircular, often segmented blades up to 11.5 cm high, usually having a stupose rhizoidal basal disc of 0.5–2 cm in diameter; blades moderate to heavily calcified on both surfaces; concentric hair lines common on the

external surface and less frequent on the internal one. Blades 92.9-143.9  $\mu\text{m}$  thick, consisting of an alternation of 2 to 3 cell layers in the middle and basal parts. Cells square in cross section, 15-27(48) x 14-29(38)  $\mu\text{m}$ , and rectangular in longitudinal section, 56-89(102) x 15-30  $\mu\text{m}$ ; in areas with 2 cell layers, the internal cell higher than the external one, of 19-53  $\mu\text{m}$  high. Sporangia occur in sori between the hair lines, isolated or sometimes grouped forming discontinuous bands; indusium present and persistent after aperture, with a clear longitudinal opening; sporangia obovate, (72)108-144 x (57)72-92  $\mu\text{m}$ , supported by a single cell pedicel of 32-56 x 14-28  $\mu\text{m}$ . The specimens were collected in Catalonia (BCN-Phyc 6235, 6240), in the Valencian Country (BCN-Phyc 1612, 3410) and in the Balearic Islands (BCN-Phyc 3405, 3420, 3424) occurring from the surface to a depth of 7(15) m.

Specimens of *P. pavonicoides* consist of erect, semicircular, rarely segmented blades up to 3 cm high, usually having a stupose rhizoidal base without forming a disc; blades slightly calcified on the internal surface and no or little calcified on the external one; concentric hair lines on both blade surfaces, but less frequent on the internal one. Blades consisting of 3 cell layers in the middle and basal parts, the central one being double the height of the other two. External cells square in cross section, 21-27 x 23-36  $\mu\text{m}$ , and rectangular in longitudinal section, 60-112 x 24-31  $\mu\text{m}$ ; central cells 48-62  $\mu\text{m}$  high. Reproductive structures not observed. The specimens were collected in Catalonia (BCN-Phyc 6241), occurring between 10 and 25 m depth.

### Discussion

Our specimens of *P. ditristromatica* and *P. pavonicoides* are consistent with the descriptions of these species provided by Ni-Ni-Win *et al.* (2011). Concerning *P. ditristromatica*, in the present paper, we provide new morphological and anatomical information, as well as new data on the description of both sori and sporangia. It can be noticed that the indusium is persistent and it tears longitudinally, their remnants remaining perpendicular to the hair lines. This feature clearly distinguishes *P. ditristromatica* from *P. pavonica*, in which the indusium splits transversally and their remnants remain parallel to the hair lines. Another useful character proposed to distinguish both species is the lack of a stoloniferous base in *P. ditristromatica*. Regarding *P. pavonicoides*, one of the main features defining this species is the fact that the central layer of the blade (which consists of 3 cell layers) has double the height of the exterior layers. Our observations in *P. pavonica* showed that this species can also present a taller central layer in the part of the blade with 3 cell layers, and therefore we consider that this feature should be reinvestigated.

### Acknowledgments

This study was supported by project CGL2009-09589-BOS of the Spanish Government.

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## INFRASPECIFIC GEOGRAPHICAL VARIABILITY IN THE FLAVONOID CONTENT OF *ZOSTERA NOLTEI*

### Abstract

*The geographical variability in the flavonoid content of Z. noltei from different Atlantic and Mediterranean Sea regions was evaluated. Zostera noltei leaves were collected at thirteen localities and extracts were prepared by maceration in water. The flavonoid content was fully characterized using NMR, UV and LC-MS spectroscopy. Analysis of the results showed that the 3 populations from the French Atlantic coast and the 7 populations from the Mediterranean Sea share the same flavonoid pattern, largely dominated by diosmetin-7-sulphate. In contrast, the three populations grown in the sub-basin of the North Atlantic nearest to the Strait of Gibraltar are chemically distinct, with apigenin-7-sulphate being the dominant. This is the first large-scale study of the Zostera noltei flavonoid fingerprint.*

**Key-words:** *Zostera noltei*, flavonoid content, chemotype, geographical chemodifferentiation.

### Introduction

*Z. noltei* Hornem. (common name Dwarf eelgrass, Zosteraceae family) occurs along European and North African coasts (Green & Short, 2003). Until recently, *Zostera* taxonomy was only based on morphology. The development of DNA-based molecular markers has led to an abundant literature on seagrass genetics over the last decade. The existence of geographically distinct populations of *Z. noltei* throughout its biogeographic range has been reported. As yet, the factors underlying this geographical genetic variability are poorly understood. Their possible consequences for the secondary chemistry of *Z. noltei* have not been considered despite the role of these substances as chemical defences. Only a few studies have investigated the phenolic content of *Z. noltei*, aside from our papers (Achamlale et al. 2009a,b; Grignon-Dubois et al. 2012a). Concerning the flavonoids, we have recently reported the first evidence of a chemodifferentiation between *Z. noltei* population from Arcachon Bay and Cadiz Bay (Grignon-Dubois et al, 2012b), and of their role in allelochemical relations (Laabir et al., 2013). Documenting the presence of flavonoids in *Z. noltei*, and how these compounds vary in abundance between seagrass meadows across large geographical scales is crucial to understanding the landscape-level adaptability of the plant to environmental factors. Our aim was to fully characterize the flavonoid profile of *Z. noltei*, and examine how these compounds vary among seagrass meadows located throughout the Atlantic and Mediterranean.

### Materials and methods

*Z. noltei* leaves were collected from 13 seagrass beds representative of 3 types of coastal ecosystems: mesotidal bays on the Atlantic coast, Mediterranean coastal lagoons, and open-sea Mediterranean *Zostera* bed (Tab. 1). Aqueous extracts were prepared from the plant material and analyzed for both the identity and quantity of phenolics present using NMR, HPLC and LC/MS.

**Tab. 1: Collections sites**

Country	Collections sites	Sea Regions	Geographical coordinates
	Gulf of Morbihan	French Atlantic coast	47° 60 N, -2 ° 82 W
	Bay of Arcachon		44° 59 N, -1°24 W
	Hossegor Lake		43° 66 N, -1°42 W
France	Salses-Leucate	Western Mediterranean Basin	42° 85 N, 2 °90 E
	Lagoon		43° 45N, 3° 65 E
	Thau Lagoon		43° 43N, 5° 10 E
Tunisia	Bizerte Lake		37° 24 N, 9° 85 E
	Alfac Bay		40° 62 N, 0° 74 E
Spain	Menorca		39 ° 92 N, 4° 05 E
	Cadiz Bay		36 ° 51 N, -6 ° 19 W
Portugal	Ria Formosa	Gulf of Cadiz	37° 00 N, -7 ° 92 W
Morocco	Merja Zerga		34 ° 83 N, -6 ° 26 W
Turkey	Izmir Bay	Eastern Mediterranean Basin	36° 12 N, 26° 24 E

## Results

At each location, the flavonoid profile was dominated by a single product, which was identified as apigenin-7-sulfate (Cadiz, Faro, and Morocco's rif) and diosmetin-7-sulfate (all other sites). These assignments were confirmed by acid hydrolysis of the crude extracts, which led to apigenin (Cadiz, Faro, and Morocco's Rif) or diosmetin (all other sites). In addition, authentic samples of the 7-sulphated-flavonoids were synthesized by sulfation of luteolin, apigenin and diosmetin.

## Discussion and Conclusion

The results showed that the populations from the French Atlantic coast and the populations from the Mediterranean Sea share the same flavonoid fingerprints, largely dominated by diosmetin-7- sulphate. In contrast, the Atlantic populations nearest to the Strait of Gibraltar are chemically distinct from all the others. This reveals unknown features about the chemical plasticity and patterns of flavonoid composition in *Z. noltei*. Understanding the underlined causes of the geographical flavonoid variability and its possible link with ecological factors appears crucial to elucidating the functioning of *Z. noltei* communities, and to the monitoring and management of *Zostera* beds.

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## PHENOLIC CHEMISTRY OF THE NATIVE MEDITERRANEAN SEAGRASSES

### Abstract

The phenolic fingerprints of the four native Mediterranean seagrasses *Posidonia oceanica*, *Cymodocea nodosa*, *Zostera marina*, *Zostera noltei*, were unambiguously established using a panel of reliable techniques of analysis (NMR, analytical and quantitative HPLC with DAD-detection, LC/MS). *Z. noltei* and *Z. marina* are characterized by the same phenolic acid pattern, namely zosteric-, caffeic- and rosmarinic acid. In contrast, they differ in their flavonoid content, which is dominated by sulphates of luteolin- and chrysoeriol in the case of *Z. marina* and diosmetin-7-sulphate in the case of *Z. noltei*. In contrast, *P. oceanica* and *C. nodosa* were found to share the same phenolic signature, dominated by caffeic tartrates, along with low amounts of coumaric- and ferulic tartrates. The virtual lack of significant concentration of flavonoid in *P. oceanica* and *C. nodosa* contrasts with *Z. marina* and *Z. noltei*.

**Key-words:** *Posidonia oceanica*, *Cymodocea nodosa*, *Zostera marina*, *Zostera noltei*, phenolic fingerprint.

### Introduction

Seagrasses constitute a group of about 60 species, which form the most widespread and productive coastal systems in the world. Of the five species of strictly marine found in the Mediterranean Sea, one is endemic (*Posidonia oceanica*), three are also found in the Atlantic Ocean (*Cymodocea nodosa*, *Zostera marina* and *Zostera noltei*) and one is a Lessepsian immigrant (*Halophila stipulacea*).

The native Mediterranean endemic seagrasses form extensive and highly productive meadows, which play important ecological roles. They are subject to many threats, both natural and anthropogenic and their conservation is a priority. Phenolic signatures should constitute good candidates as early indicators of the level of seagrass environmental stress. Phenolics play several important functions in plants and they are common in marine ecosystems. Many of them are antimicrobial agents, presumed to protect marine macrophytes against pathogen attacks, harmful ultraviolet radiation, and herbivores. The comprehensive identification of phenolic compounds is a crucial starting point for assessing the seagrass's secondary chemistry and its possible role in the plant's chemical communication and defence mechanisms. Our aim was to determine and compare the phenolic fingerprint of the four native Mediterranean seagrasses (ie *P. oceanica*, *C. nodosa*, *Z. marina* and *Z. noltei*).

### Materials and methods

Plant material was collected throughout the Mediterranean Sea (Tab.1). Dried ground leaves were extracted at room temperature with aqueous methanol. Phenolic compounds were identified, based on NMR spectra, UV-visible spectra, HPLC retention time and on-line DAD spectra, MS molecular ions in comparison with authentic standards.

**Tab. 1: Species and Collection sites**

Species	Collection sites
<i>Z. noltei</i>	Spain (Menorca, Alfacs Bay), France (Salses Leucate Lagoon, Thau Lagoon, Berre Lagoon, Turkey (Izmir Bay), Tunisia (Bizerte Bay)
<i>Z. marina</i>	France (Salses Leucate lagoon, Thau lagoon)
<i>P. oceanica</i>	Spain (Port Lligat), France (Lerins Islands), Tunisia (Carthage, Mahdia)
<i>C. nodosa</i>	Spain (Alfacs Bay), Turkey (Izmir Bay), Tunisia (Monastir)



## Results

The combination of reliable and complementary analytical tools allowed a clear understanding of the phenolic content of the four Mediterranean native seagrasses. *P. oceanica* and *C. nodosa* share the same phenolic signature, largely dominated by chicoric acid, accounting for 80-89 % of the total phenolic detected. Caffeic acid is the second phenolic in abundance, but in much lower concentration, along with traces of ferulic and coumaric acid. Noticeable is the virtual lack of significant concentration of flavonoid. Only small amounts of flavonol were detected and other forms of flavonoids such as flavones or flavanones were undetectable.

The similarity of the phenolic pattern is so great that it makes it difficult to tell the difference between *P. oceanica* and *C. nodosa* on the sole basis of their HPLC-DAD chromatograms.

In contrast, *Z. noltei* and *Z. marina* exhibit a greater phenolic diversity including phenolic acid derivatives as well as gluco- and sulphated flavonoids. Both contain the same phenolic acids. Rosmarinic acid is the most abundant, while caffeic and zosteric acid are present in lower concentration. In contrast, the two species greatly differ in their flavonoid content. The individual compounds identified were the 7-sulphate of luteolin, diosmetin and chrysoeriol and the 7,3'-disulphate of luteolin for *Z. marina*, this last one being the major product, whereas only the 7-sulphates of luteolin and diosmetin are found in *Z. noltei*, with this last one being dominant.

## Discussion and conclusion

Phenolics arise biogenetically from phenylalanine through the shikimate-phenylpropanoids-flavonoids pathways. The p-coumaroyl CoA is at the crossroads of the metabolic routes leading either to flavonoids or to phenolic acids. The metabolic flux entering these two routes are respectively controlled by the chalcone synthase (CHS) and the hydroxycinnamoyl transferase (HCT). The low flavonoid content of *P. oceanica* and *C. nodosa* suggests a poor expression of the gene encoding the chalcone synthase. This flavonoid deficiency contrasts with the two Mediterranean *Zostera* species. Also noticeable is the lack of sulphated phenolics in *P. oceanica* and *C. nodosa*, while they are abundant in *Z. noltei* and *Z. marina*.

Our results show that the *P. oceanica* and *C. nodosa* phenolic metabolism is dominated by caffeic tartrates, along with low amounts of coumaric and ferulic tartrates. Chicoric acid was found to be the major phenolic product, at all the sites studied. The large accumulation of chicoric acid in *P. oceanica* and *C. nodosa* and the mixture of rosmarinic acid and sulphated flavonoids in *Zostera* most likely play an ecological role. The factors controlling the phenolic chemistry and its functions in seagrasses remain poorly understood. The understanding of the enzymes of the phenylpropanoid pathway in seagrasses in general and of the ecological roles of the resulting secondary phenolic metabolites will be of great help in the understanding of the chemically mediated interactions in seagrass beds, and also the management and preservation of these crucial ecosystems.

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## **CHEMICAL WARFARE IN COASTAL ECOSYSTEMS: *ZOSTERA VERSUS ALEXANDRIUM***

### **Abstract**

*Allelopathic effects of Zostera marina and Z. noltei on the growth of the toxic red tide dinoflagellate Alexandrium catenella were studied in laboratory co-cultures. For the first time, exudates from Zostera leaves were successfully extracted from seawater, purified, and their effect on the biology of A. catenella investigated. Results showed a strong inhibition of photosynthetic activity and a significant reduction of vegetative cells growth. Both, exudates and pure phenolics isolated from Zostera induced severe structural anomalies in Alexandrium cells. The photosynthetic and growth inhibitions as well as cellular degradations were dose and time-dependent. The present study shows that Zostera species exhibit negative allelopathic effects on harmful bloom-forming microalgae.*

**Key-words:** *Zostera* species, *Alexandrium catenella*, allelopathy, inhibition growth, phenolic exudation

### **Introduction**

Plants are provided with a large arsenal of defensive secondary metabolites. Most of these chemical defences belong to the huge family of phenolics. They are well known as allelopathic agents in terrestrial plants, and similar ecological functions have been proposed for seagrasses. There is now strong evidence that chemical interactions play an important role in marine ecosystems regulating algal biodiversity, structure and seasonal variation. However, the effects of marine plant secondary metabolites on co-occurring microorganisms remain largely undocumented.

*Z. marina* and *Z. noltei* are important seagrasses occurring along the European and North African coasts. *Alexandrium catenella* is a widespread PSP toxin-producing dinoflagellate species. In recent decades, seagrass beds have declined across Europe, while the frequency of HAB events has increased. This is the case of Crique de l'Angle (Thau Lagoon, French Mediterranean coast), where *Zostera* bed virtually disappeared and recurrent *Alexandrium* outbreaks began and developed. This led us to hypothesize that *Zostera* bed regression could facilitate the settlement of *Alexandrium* blooms. In support of this hypothesis, we have demonstrated that extracts of *Zostera* leaves significantly inhibit the growth of *A. catenella* (Laabir et al., 2013). The highest concentrations of phenolics were found to correspond with the lowest EC50 values, suggesting that these secondary metabolites might be responsible for the observed algicidal activity. In continuation of this work, we proceeded to the purification of the crude extracts in order to identify the causative bioactive substances in the cocktail of phenolics produced by *Z. marina* and *Z. noltei* (Achamlale et al., 2009a,b). Their ability to diffuse in the water column was also studied.

### **Materials and methods**

*Z. noltei* and *Z. marina* were collected at Bouzigues (Thau Lagoon). After collection, the plants were gently rinsed in fresh water, then air dried at room temperature. Methanolic and aqueous extracts were prepared from the plant material. Exudates were obtained by

maceration of *Zostera* tissue in seawater. The identity and quantity of phenolics present in these extracts were determined using NMR, HPLC and LC/MS. Purifications were conducted on C18 silica gel.

The monoclonal *A. catenella* strain (ACT03) was cultivated in Enriched Natural Sea Water (ENSW medium). The *Alexandrium* cells used in the bioassays were in the exponential phase of growth (Laabir et al. 2013). The inhibitory effects were tested at different concentrations on ACT03 strains in batch cultures. Effects on the photosynthetic activity of *A. catenella* were studied with a Phyto-PAM Fluorometer. Analysis of cell damage was performed microscopically, using a digital video camera connected to an optical microscope. All the experiments were conducted in triplicate.

## Results

Exudation of phenolics in the seawater from both fresh leaves was demonstrated. Phenolic acids and flavonoids were successfully isolated from *Zostera* extracts and *Zostera* exudates. All of them inhibited *A. catenella* growth. Exposition to these substances and exudates led to severe structural anomalies of *Alexandrium* cells, which became incapable to divide. In acute cases, intracellular organelles disappeared, the membranes were lytic, and the cells became necrotic. The amplitude of the inhibition on the growth and photosynthetic activity was time and dose-dependent.

## Discussion and conclusion

Allelopathy is a phenomenon by which an organism produces biochemicals that influence the growth, survival, and reproduction of other organisms. They can have beneficial or detrimental effects on the target organisms. Negative allelopathic effects are an important part of plant defences. To demonstrate allelopathy, production, and identification of allelochemicals must be established as well as their persistence in the environment over time in concentrations sufficient to affect plant species. The present study demonstrates the inhibitory effects of phenolics secreted by *Z. marina* and *Z. noltei* on the neurotoxic dinoflagellate *A. catenella*. The photosynthetic activity was targeted by phenolics. They stopped cell division and provoked cell damage, which led to the disruption of cellular processes. The results confirm the negative allelopathic effects of *Zostera* versus *A. catenella* and throw new light on the role that *Zostera* secondary chemistry could play in regulating HAB dynamics.

## Acknowledgments

This work was supported by the SUDOE program (ECO-LAGUNES project), the Region of Aquitaine (OSQUAR project) and the CNRS (EC2CO - Zost-Alex project)

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## **ETUDE DU MILIEU MARIN EN PALMES MASQUE TUBA GUIDE METHODOLOGIQUE**

### **Résumé**

*Le suivi des petits fonds côtiers peut être réalisé à l'aide d'un matériel simple et peu encombrant : les Palmes, le Masque et le Tuba (PMT). Afin de réaliser un guide méthodologique à destination des gestionnaires d'Aires Marines Protégées (AMP) sur l'étude du milieu marin en PMT, six protocoles ont été sélectionnés en concertation avec scientifiques et gestionnaires. En premier lieu, le guide rappelle le rôle du suivi naturaliste dans le cadre de la gestion d'une AMP puis les spécificités de la plongée libre concernant le matériel, les mécanismes de l'apnée et les principes de sécurité. Dans un second temps, les protocoles sélectionnés font l'objet d'une fiche méthodologique illustrant le suivi d'espèces et d'habitats marins du médiolittoral et de l'infralittoral supérieur méditerranéen notamment sur *Diplodus spp.*, *Pinna nobilis* et *Lithophyllum byssoides*. Chaque fiche détaille le protocole de terrain ; des pistes pour l'exploitation des données sont également proposées.*

**Mots-clés :** AMP, Protocole, PMT, Infralittoral, Guide.

### **Introduction**

Les suivis scientifiques mis en œuvre à long terme par le gestionnaire d'une Aire Marine Protégée (AMP) se doivent d'être simples et facilement mobilisables. Concernant les espèces et les habitats marins du médiolittoral et de l'infralittoral supérieur, les suivis peuvent être réalisés à l'aide d'un matériel peu encombrant : les palmes, le masque et le tuba (PMT). Un guide méthodologique du suivi du milieu marin en PMT est proposé à partir d'une première sélection de protocoles.

### **Matériel et méthodes**

Une démarche de concertation a été conduite initialement auprès de dix gestionnaires d'AMP de méditerranée nord-occidentale et de treize organismes scientifiques pour inventorier les protocoles réalisables en PMT. Six protocoles ont été sélectionnés et ont fait l'objet de tests sur le terrain pour identifier d'éventuelles difficultés pour leur mise en œuvre.

### **Résultats**

Ce guide méthodologique à destination des gestionnaires d'AMP méditerranéennes comprend quatre parties. La première partie, rappelle le rôle d'aide à la décision que constitue le suivi scientifique dans le cadre de la gestion d'une AMP. Un accompagnement scientifique est préconisé pour la mise en œuvre d'un suivi afin de définir précisément les objectifs et la stratégie d'échantillonnage : choix du matériel biologique, moyens matériels et humains, période et fréquence d'acquisition des données et répartition des stations d'échantillonnage.

Dans une seconde partie, les spécificités de la plongée libre sont présentées en particulier le matériel, les mécanismes de l'apnée et les principes de sécurité. Précisons que nombre de suivis peuvent être réalisés depuis la surface sans réaliser d'apnée.



La troisième partie est consacrée au matériel de suivi avec entre autres la fabrication d'une plaque immergeable pour la prise de notes, d'un quadra ou encore le choix d'un appareil photo, d'un GPS.

Dans la dernière partie les six fiches méthodologiques sont présentées : (1) comptage visuel des poissons : le suivi d'espèces cibles, (2) identification et cartographie des nourriceries du genre *Diplodus spp.* (Chordata, Metazoa) sur les côtes rocheuses, (3) cartographie et évaluation de la vitalité des encorbellements à *Lithophyllum byssoides* (Lamarck) Foslie, 1900 (Florideophyceae, Stramenopiles), (4) inventaire et suivi de *Pinna nobilis* (Linnaeus, 1758) (Mollusca, Metazoa), (5) cartographie de la limite supérieure de l'herbier à *Posidonia oceanica* (Linnaeus) Delile seagrass meadow, (6) suivi des populations de *Paracentrotus lividus* (Lamarck, 1816) (Echinodermata, Metazoa) sur fonds rocheux.

En introduction de chaque fiche les enjeux de conservation des espèces ou des habitats considérés sont présentés. Le protocole de terrain est ensuite détaillé : matériel requis, méthode d'acquisition des données, organisation de la collecte des données, conseils concernant la répartition des stations d'échantillonnage. Puis des pistes sont données quant à la saisie et à la valorisation des données. En exemple une évaluation de la vitalité d'un encorbellement à *Lithophyllum byssoides* est présentée selon les travaux conduits par Verlaque M. (2010) (Fig.1). La vitalité est évaluée au sein d'un quadra de 20 cm x 20 cm subdivisé en carrés de 2 cm (Fig.1, a). A chaque carré est identifiée une catégorie de recouvrement : V (colonies vivantes), R (recrutement annuel), M (colonies mortes), A (recouvrement algal épiphytique), T (trou) (Fig. 1, b). Le pourcentage de recouvrement de chaque catégorie est ensuite calculé : colonies vivantes (31%), recrutement annuel (7%), colonies mortes (11%), recouvrement algal épiphytique (28%), trou (23%).

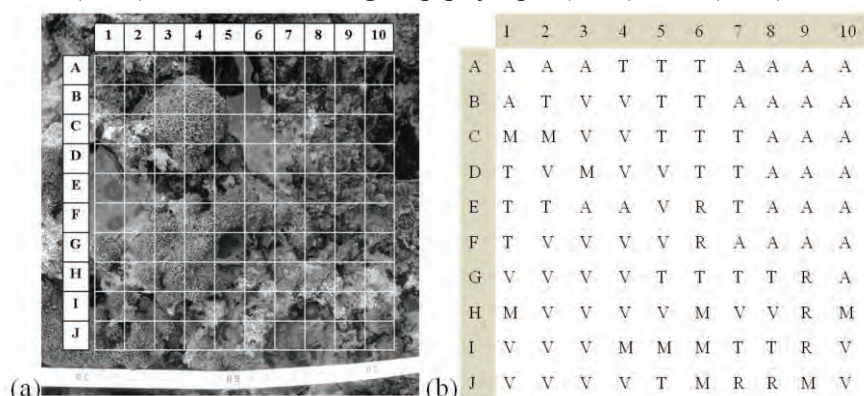


Fig. 1: Evaluation de la vitalité d'un encorbellement à *Lithophyllum byssoides* au sein d'un quadra de 20 cm x 20 cm (a), identification et répartition des différentes catégories (b)

## Discussion

Chaque protocole devra toutefois être adapté aux spécificités (conditions environnementales et réglementaires) de chaque AMP. Ce guide se veut être un outil pragmatique apportant des solutions techniques facilement mobilisables. Il constitue une première étape dans la réalisation d'un recueil méthodologique et n'est pas exhaustif tant les espèces, les habitats et les problématiques de gestion diffèrent d'un coin à l'autre de la méditerranée. L'idée est d'initier une démarche collaborative au sein des réseaux de gestionnaires (MedPAN et AAMP) et que ce travail soit complété à terme par de nouveaux protocoles.

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## **MAPPING OF MARINE HABITATS IN PORTO PALERMO BAY TO SUPPORT ESTABLISHMENT AND ZONING OF FUTURE MPA**

### **Abstract**

*Porto Palermo is a protected bay and had a very limited access in the former time (until 1991), as it was a military area. Due to this, marine biodiversity is presumed to have been rich in that period, although the surveys were very limited. Several studies, including the 2013 Marine and Coastal Protected Areas Strategic Action Plan, have proposed the area to be established as a MCPA. The study has covered the area inside the Porto Palermo and the intermediate surroundings towards north and south of the bay. Particular attention has been paid to the vulnerable/sensitive Mediterranean habitats and species of conservation interest. Among the most sensitive habitats of the Mediterranean should be mentioned *Lithophylum byssoides* for the mediolittoral zone, the *Cystoseira* communities as an index of hydrodynamism at the upper littoral zone, and the *Posidonia oceanica* meadows in the infralittoral zone. A total of twelve species of green algae, fourteen species of brown algae and twenty five species of red algae were recorded during the survey. The study also has listed some of the main threats to biodiversity values in the Porto Palermo area. The final result of this study is a preliminary map of boundaries and zoning of the Porto Palermo MPA.*

**Key-words:** marine biodiversity, Mediterranean habitats, *Posidonia oceanica*, species of conservation interests, zoning.

### **Introduction**

The main objective of this survey is to identify the important habitats and species in the area and the intermediate surroundings. The scope is to map and assess the seagrass meadows and other habitats, in order to provide a spatial description of the seabed environment within and around the Porto Palermo bay, including the identification of important/sensitive habitats and species. Main threats to biodiversity are identified in order to provide for a preliminary zoning of the future marine protected area

### **Material and methods**

The study has covered an area of inside the Porto Palermo and the intermediate surroundings towards north and south of the bay. The investigation has been organized in surveying transects starting from the coastline and mid-littoral, following with infra-littoral, until the upper circumlittoral. A total of 24 surveying transects were covered. An underwater video-recorder (model Camera controller CBX GA, Inspecam LX&Z) was used for the survey in the infra-littoral, starting from 3–4 m depth, until the upper circumlittoral. Mapping of habitats has been carried out using ARCGIS software and combining data from the underwater surveys with the underwater video recorder and experts notes from spot diving.

### **Results of the study**

The results are focused on highlighting most important marine habitats, species and associations. Particular attention has been paid to the vulnerable/sensitive Mediterranean habitats and species of national and international conservation interest (Bern Convention; EU Habitat Directive; Albania Red List of Species). Marine communities and habitats in Porto

Palermo Bay (RAC/SPA 2014) include:

- Littoral organogenic concretions (*Lithophyllum byssoides* cushions) present only in fragments (isolated cushions) along the coast of north zone of Porto Palermo area.
- Biocenosis of medio-littoral caves are present within the bay and along the coasts of north zone of the studied area, where species like *Hildenbrandia prototypus*, *Phymatolithon lenormandii*, *Peyssonelia* spp., etc. are grown.
- Biocenosis of infralittoral algae. (Association with *Cystoseira amentacea* var. *spicata*) located in the first meter of the infralittoral and creates belts mainly in exposed coasts of the north zone of Porto Palermo area. A total of twelve species of green algae, fourteen brown algae and twenty five red algae were recorded during the survey.
- Three species of Magnoliophytes are present in the area: *Posidonia oceanica*, *Cymodocea nodosa* and *Halophila stipulacea*. *Posidonia oceanica* is the most abundant species, which form large meadows in shallow and medium depths at the study area. The upper limit of *Posidonia oceanica* meadows in the studied area was identified at 3.5 meters depth and the lower limit at 28 m. In shallow waters (2-3 m) within the bay there are identified small areas covered with *Cymodocea nodosa* patches, partly accompanied with *Halophila stipulacea*.
- Associations of the Coralligenous biocenosis have often been observed in different sites between 20 m to 60 m depths during this survey.

The main sources of threat to biodiversity values in Porto Palermo bay area include fishing activities, aquaculture, destructive harvesting of date mussel (*Lithophaga lithophaga*), boat anchoring and coastal constructions causing permanent destruction and alteration of natural habitats, or decreases of habitat size and its fragmentation. *Invasive Species Caulerpa racemosa* var. *cylindracea* is widely dispersed in Porto Palermo area. Two other alien species, the seagrass *Halophila stipulacea* and the Nimble spray crab *Percnon gibbesi* are also present in Porto Palermo area.

### Discussion and/or conclusions

The ecological study has identified biodiversity rich areas as well as areas under pressure from human activities. The survey shows that future MPA should include not only areas within the Porto Palermo bay but also extending north and south to its immediate surroundings. The analysis of threats suggest that it is necessary to establish the marine protected area as soon as possible and develop a sound system of management zones and rules for development/controlling of activities within the MPA in order to provide for the conservation of important biodiversity values.

### Acknowledgements

This poster is prepared in the frame of the project “Support planning, zoning and development of a Marine Protected Area in the Porto Palermo Bay (Albania), based on sound scientific and socio-economic knowledge” supported by RAC/SPA.

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## **MAPPING *POSIDONIA OCEANICA* LOWER LIMIT COMBINING HIGH RESOLUTION INSTRUMENTS (SSS AND MBS)**

### **Abstract**

*High resolution Side Scan Sonar (SSS) combined with MultiBeam echo Sounder (MBS) was used for mapping Posidonia oceanica lower limit. For this purpose an acoustic survey was performed in Mondello Bay (Palermo, Italy) where a progressive lower limit of the seagrass occurred. To improve the positioning precision of the SSS sonograms, data were joined by GIS platform and a repositioning process was performed. In particular, each sonogram was joined with the Digital Terrain Model (DTM) obtained by MBS, in which recognizable shapes of the sea-bottom were used as control points. After the repositioning process of the SSS data, the limit investigated during different periods showed a negligible error in absolute position, with an high level of spatial concordance compatible with the intrinsic error of the system.*

**Key-words :** *Posidonia oceanica*, seagrass lower limit, Sicily, Side Scan Sonar, MultiBeam echo Sounder.

### **Introduction**

The assessment of the status of *Posidonia oceanica* meadows is based on the monitoring of a set of variables among which the progressive lower limit (Pergent-Martini *et al.*, 2005). It represents a good indicator because any changing in its position highlights a variation of the environmental condition (Boudouresque *et al.*, 2009). Traditionally, the progressive lower limit is identified and monitored by the “*balisage* method” (Meinesz and Laurent, 1978). However, this technique shows many drawbacks as the positioning and the regular maintenance of the *balise*, labor intensive and time consuming of scuba divers (Montefalcone *et al.*, 2014). Currently, it is possible to map *P. oceanica* meadows by using high resolution sonar systems, able to provide morphometric and bathymetric information on large surface areas, such as those occupied by *P. oceanica* along the Mediterranean coasts (Di Maida *et al.*, 2011 and reference therein). The aim of this research was to apply a powerful and innovative methodology for a fast and accurate mapping and monitoring of *P. oceanica* progressive lower limit.

### **Material and methods**

The investigations were carried out on seabed of the northwest coast of Sicily (Bay of Mondello), characterized mainly by a sandy substratum colonized by an extensive *P. oceanica* meadow (Calvo *et al.*, 1993). On June 2010, a preliminary survey was carried out by using a Remote Operated Vehicle (ROV) to find progressive lower limit of *P. oceanica* meadow. Then, on July 2010 and on February 2011, two acoustic surveys were simultaneously carried out by SSS (Klein 3900) and MBS (RESON SEABAT 8125) for acquiring morphometric and bathymetric data. Sonograms obtained by SSS were corrected for speed, direction and TVG (Time Varying Gain). After correction, the sonograms were imported on GIS platform (Esri ArcGIS 9.3) for mosaicing and vectorizing seabed morphologies and *P. oceanica* lower limits. Bathymetric data, acquired

by MBS, were manually processed to remove each bathymetric spike or errors. Therefore, a DTM with a pixel size of 0.5 m<sup>2</sup> was constructed using GIS platform. Both SSS and MBS data for each survey were joined on GIS platform with the primary goal to remove the positioning errors of sonograms by a re-georeferencing process. In particular, each sonogram was matched with the corresponding DTM by the recognizable shapes of the sea-bottom used as control points. Afterwards, ArcGis Spatial Analyst tool was used to calculate the distances between *P. oceanica* lower limits detected during two different surveys. This procedure allowed to extrapolate information about possible position errors. In particular, minimum, maximum and mean error and Standard Deviation were calculated.

### Result and discussion

The analysis of the sonograms obtained by SSS, comparing the relative backscatter responses, allowed to easily discriminate by different shades of grey *P. oceanica* meadow and its limit from sandy substratum. The SSS data showed that the meadow exhibited a gradual coverage decrement close to its lower limit according to a progressive limit (Meinesz and Laurent, 1978). Moreover, according to Kamman and Huston (1999), the lower limit positions, acquired in the different periods, showed a wide difference (up to 5 m) in the absolute position. The MBS data allowed to give the bathymetric range of meadow lower limit (28.98 m and 40.17 m) and morphological features of the sea-bottom. Moreover, the comparison of the different DTMs, created by MBS data acquired in the two periods, showed that MBS was able to locate the correct position of every pixel with a negligible error. After the re-georeferencing process of the SSS data with the relative DTMs, the overlapping of the lower limits investigated during the different periods showed a negligible error in absolute position, with a high level of spatial concordance. Indeed, it was observed a range between 0 m and 0.70 m with an average error value of 0.1 m and a Standard Deviation of 0.0966 m. These results showed that the combining of SSS and MBS data represents a powerful and innovative methodology for a fast and accurate mapping and monitoring of *P. oceanica* progressive lower limit.

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## ***POSIDONIA OCEANICA* AND *CAULERPA RACEMOSA* VAR. *CYLINDRACEA* CO-EFFECTS ON PHENOLOGICAL PARAMETERS**

### **Abstract**

Researches on interaction between invasive *Caulerpa racemosa* var. *cylindracea* and the seagrass *Posidonia oceanica* were carried out along the northern Tunisian coasts at two different sites (Cape Zebib & Sidi Raïs) from April 2009 to April 2010. The study showed that phenological parameters of *P. oceanica* seem to be not affected by the presence of *C. racemosa* excepting the leaves width.

**Key-words:** *C. racemosavar. cylindracea*, *P. oceanica*, phenological parameters, co-effects.

### **Introduction**

The introduced algae *Caulerpa racemosa* (Forsskal) J. Agardh var. *cylindracea* (Sonder) Verlaque, Huisman and Boudouresque settle actually the entire Mediterranean basin (Klein&Verlaque, 2008) where it behaves as an invasive species (Boudouresque and Verlaque, 2002). To assess the impacts of this species on the Seagrass *Posidonia oceanica* we studied the effects of each species on the vegetative production of the other one.

### **Materials and methods**

The study was conducted in Cape Zebib and Sidi Raïs (Northern Tunisian coasts). Three situations were considered in sampling: non-invaded *Posidonia* meadow, *Posidonia* meadow invaded by *C. racemosa* and *C. racemosa* free settlements. Samples were collected every two months between April 2009 and April 2010. Samples of *C. racemosa* were triplicate into randomly positioned plots of 20x20 cm. We quantified the number of fronds and their mean length. Regarding *P. oceanica*, twenty orthotropic rhizomes were randomly collected. We estimated the mean number of leaves per shoot, the mean length and width of adult and intermediates leaves, the mean length of petioles and the coefficient "A" (percentage of the leaves having lost their apex)(Giraud,1977).

### **Results**

As is generally found, the phenological parameters of *C. racemosa* exhibited maximum values in autumn and minimum values in winter (Tab.1) regardless of the interaction. However, no significant differences due to interaction with *P. oceanica* were recorded. A similar trend seems to exist in *P. oceanica* phenological parameters which also varied according to the seasons. In terms of interaction we recorded a significant decrease of the leaves width, when interacting (Tab. 1).



**Tab.1:** Leaf biometric variations of *C. racemosa* var.*cylindracea* and of *P. oceanica*. **NI** : Non-Invade; **I**: Invaded; **Min**: minimum value; **Max**: maximum value; **Av**: Annual average; **A**: Adults leaves; **I**:intermediates leaves.

	Parameters		Cape Zebib		Sidi Raïs	
			NI	I	NI	I
<i>C. racemosa</i> var. <i>cylindracea</i>	Density of fronds (F.m <sup>-2</sup> )	Min	0	0	825±42	2525±90
		Max	4550±801	4083.3±440	15091±1203	16700±2236
		Av	1450.6	1986.9	4705.6	5645.2
	Length of fronds (cm)	Min	0	0	0.8±0.3	0.9±0.2
		Max	1.1±0.3	1.3±0.2	1.5±0.3	1.8±0.1
		Av	0.7	0.9	1.1	1.2
<i>P. oceanica</i>	Number of A and I leaves	Min	4.7±0.6	4.8±0.4	4.1±1	4.1±0.9
		Max	6.5±1	6.7±0.6	5.9±0.6	5.8±0.9
	length of leaves (A and I) (mm)	Min	298.2±86.3	250.2±121.5	181.9±56.7	195±52.5
		Max	901.2±220.1	620.3±139.4	467.8±98.2	421.2±78.8
	Length of petiole (mm)	Min	34.3±12.5	24.8±7.2	34.3±6.9	30.2±6.6
		Max	44.9±10.3	46.7±11.8	45.9±11.8	47.8±11.7
	width of adult Leaves (mm)	Min	-	-	-	-
		Max	9.5±0.1	9.0±0.1	9.9±0.1	9.4±0.2
	Coefficient A (%)	Min	32	31	35	20
		Max	90	86	80	85

### Discussion & Conclusion

*C. racemosa* phenological maximum values recorded in Tunisia does not differ from values recorded in other Mediterranean sites and reported by Klein & Verlaque (2008). On another hand, results from Tunisia show that, excepting the leaves width, phenological parameters of *P. oceanica* does not seem to be affected by the presence of the invasive species *C. racemosa*. This is obviously different from what is recorded in the northern Mediterranean where particularly Dumay *et al.*, (2002) reported that in front of another Caulerpa *i.e. Caulerpa taxifolia* (M.Vahl) C. Agardh, the mean leaf length of *P. oceanica* decreases and the number of leaves produced annually increases.

### Acknowledgment:

The authors would like to thank M. Imed ESSETI for revising the English text.

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## **NEW BIONOMIC MAP OF PROTECTED MARINE AREA "TAVOLARA - PUNTA CODA CAVALLO", SARDINIA, ITALY, WEST MEDITERRANEAN**

### **Abstract**

*Bionomic mapping is one of the main tool for the analysis, knowledge and management of marine ecosystems. Between 2011 and 2012 the sea bottom of the Italian Marine Protected Area "Tavolara - Punta Coda Cavallo" (hereafter MPA) has been surveyed with multi-beam echo sounder, side scan sonar and scientific scuba divers: high-quality geodatasets has been produced in raster and feature formats describing marine habitats and biocenoses according to the typology of benthic biological communities of Mediterranean National Museum of Natural History. In 2013 MPA has reordered these feature datasets according to Natura 2000 and in early 2014 published a new bionomic map. In this poster we illustrate results, technologies and procedures that led to the publishing of this new bionomic map. The poster also integrates a diachronic comparison of the *P. oceanica* meadows in MPA between 1992 and 2012.*

**Key-words:** bionomic mapping, MPAs, GIS, Natura 2000, benchmarking

### **Introduction**

The European Union defines Marine Protected Areas (MPAs) as territories where conservation and management measures for coastal marine environments must be implemented and new approaches tested (Francour *et al.*, 2001; Guidetti *et al.*, 2008). Inside Marine protected areas, cartography is an essential tool to represent the spatial aspects of natural environments (Tricart & Kilian, 1985). One of the main activities of the Italian Marine Protected Area "Tavolara - Punta Coda Cavallo" (hereafter simply MPA - [www.amptavolara.it](http://www.amptavolara.it)) has been the mapping of the seafloor using more refined techniques as they became available. In the first years from its institution, a marine geomorphological map was redacted (Orrù, 2006) to corredate a pre-existing map of marine habitats (Navone *et al.*, 1992) that was obtained mostly with single beam echo-sounder, side scan sonar and SCUBA scientific dives. These early maps were redacted at synoptic scale (1:25,000) but were essential in developing maps of habitat vulnerability, potential quality and susceptibility to use of the marine territory inside the MPA (Rovere *et al.*, 2013).

### **Materials and methods**

Between 2011 and 2012 the sea bottom of MPA has been surveyed within a PIM-funded project (Initiative pour les Petites Iles de Méditerranée - [www.initiative-pim.org](http://www.initiative-pim.org)). The mapping was carried out by Andromède Oceanologie with the support of the MPA. Main activities consisted of: 1) mapping of the seafloor with side scan sonar Klein 3900 with dGPS and SonarPro software; 2) mapping of the seafloor with multi-beam echo sounder GeoSwath with 2 transducers 250 kHz (100m range) together with dGPS and GeoSwath+ software; 3) SCUBA divers made with rebreather equipment, and audio recordings of

the bottom features as dictated by the diver; 4) interpretation of the dataset acquired in GIS environment according to the typology of benthic biological communities of Mediterranean National Museum of Natural History (Michez *et al.*, 2011) and map production. For management purposes in 2013 MPA has reordered in GIS environment a wide series of geodatasets coming from old and recent surveys and available in different formats. This activity included the transposition of these last benthic biological feature datasets according to classes and attributes of Natura 2000. In 2013 MPA has also started a collaboration with SeaMap for a first comparison of homogeneous geodatasets dated 1992 and 2012 and some other management analysis.

## Results

During the 2012, final production led to 2 final raster datasets with the resolution respectively of 30cm/pixel (sonar images) and 1m/pixel final (bathymetry) and to 1 bionomic feature layer of 1:25,000 scale, arising from interpretation of all the data acquired. In 2013, focusing on *Posidonia oceanica* meadows, a diachronic map was produced showing areas where *P. oceanica* has disappeared, due for example to anchoring, professional fishing or exceptional coastal floods; in other areas *P. oceanica* registered an advancement while in most areas it is confirmed that no change happened since 1992. In early 2014 MPA has published a new bionomic map in 1:25,000 scale coming from the transposition of the 2012 bionomic feature layers to Natura 2000, with the integration of the land habitats of coast and islands inside MPA.

## Conclusions

Mapping and GIS analysis are strategically actions and tools for MPAs: higher results coming from continuous monitoring and different times series' analysis, although realized with different methods and at different scales, give the opportunity to benchmark managing policies and to better plan conservation efforts.

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## **WHICH CONTAMINATION OF THE SEAGRASS *POSIDONIA OCEANICA* ALONG THE PROVENCE COAST (FRANCE, NW MEDITERRANEAN SEA)?**

### **Abstract**

*Posidonia oceanica* seagrass meadows are exposed to natural and anthropogenic pressures. Sewage effluents are a major recurrent problem due to the ever-increasing population along the coasts, inducing nutrient, particulate organic matter and contaminant enrichment of marine coastal waters. We studied contamination by organic (PCBs) and inorganic (As, Ba, Cd, Cr, Cu, Hg, Ni, Pb and Zn) contaminants in *P. oceanica* adult leaves and rhizomes collected at 4 sites exhibiting different levels of human pressure along the coasts of Provence (France, Mediterranean Sea) near the city of Marseilles (3 sites), and Hyères (the reference site). Higher values were recorded at Marseilles, due to the vicinity of major urban, harbour and industrial centres, but only for trace elements. In addition, more contaminants were accumulated in adult leaves than in rhizomes. Thus, *P. oceanica* leaves could be used as a tracer of spatial contamination and as a tool for water quality evaluation to assess the contamination level in food webs.

**Key-words:** trace elements, PCBs, *Posidonia oceanica*, Mediterranean Sea

### **Introduction**

Coastal marine habitats are exposed to intensive human activities. The environmental occurrence of polychlorinated biphenyls (PCBs) and Trace Elements (TEs) is a matter of concern for shallow coastal ecosystems. The Gulf of Marseilles is subject to major inputs of contaminants from different sources: sewage, industrial effluents, large rivers and atmospheric depositions. But it is important to consider also sporadic inputs from small rivers and diffuse runoff from coastal urban areas. The endemic seagrass *Posidonia oceanica* (Linnaeus) Delile was used as a contaminant bioindicator to evaluate the bioavailable levels of contaminants (Warnau *et al.*, 1995; Lafabrie *et al.*, 2008) for living organisms in seagrass meadows. As part of a study of food webs and contaminant bioaccumulation in *P. oceanica* meadows, we focused here on contaminant levels in this plant to test their relation to human pressure by comparing different sites with a contrasting degree of inputs and by analysing different parts of the seagrass.

### **Material and methods**

Shoots of *P. oceanica* were collected in winter 2012 in seagrass meadows located at 3 sites near Marseilles (Plateau des Chèvres: PC, Prado Bay: PB, Côte Bleue: CB; from the highest to the lowest impacted), one of the largest cities of the Mediterranean, sites frequently exposed to the urban sewage of Marseilles, and in the Gulf of Hyères (HY), a less urbanized area, considered here as the reference site. At each site, 5 to 8 shoots were sampled between 8 and 12 m depth. Adult leaves (i.e. the oldest ones) were cleaned of epibionts and sediment with a scalpel and rinsed with ultrapure water. For rhizomes, only the 3 last years were retained using lepidochronology; rhizome samples were pooled per site. Samples were frozen (-20°C),

lyophilized and reduced to a coarse powder with a ball mill. Contaminants were measured in adult leaves and rhizomes. Total Hg was analysed using an Advanced Mercury Analyser. As, Ba, Cd, Cr, Cu, Ni, Pb and Zn analyses were carried out with Varian Vista-Pro ICP-OES and a Thermo Fisher Scientific X Series 2 ICP-MS. PCBs (sum of 32 individual congeners) were analysed by gas chromatography with an Agilent Technologies HP 6890 equipped with a  $^{63}\text{Ni}$  electron capture detector. Statistics were performed by Kruskal-Wallis one-way analysis of variance to look for differences between stations and Friedman's two-way ANOVA by ranks (leaves vs rhizomes).

### Results and discussion

The concentration of most contaminants varied considerably in leaves, depending on location (Tab. 1). TE concentrations decreased following the order:  $\text{Zn} > \text{Ni} > \text{Cu} > \text{Cd}$ ,  $\text{Pb}$ ,  $\text{Ba}$ ,  $\text{As}$ ,  $\text{Cr} > \text{Hg}$ , from 192.1 for Zn to  $0.1 \mu\text{g g}^{-1}$  dry mass for Hg, which was in agreement with previous studies (Lafabrie *et al.* 2008). Values of As, Ba, Cr and Hg in leaves were lower at Hyères than at the Marseilles sites. However, no significant spatial difference was observed for PCBs (mean  $14.4 \pm 1.8 \mu\text{g kg}^{-1}$  dry mass). Higher concentrations of As, Ba, Cd, Cr, Pb, and Zn were detected in leaves than rhizomes, which reflect their preferential uptake from the water by photosynthetic tissues. This partition suggests that physiological factors are the main drivers of contaminant distribution. PCBs and Hg were more heavily accumulated in rhizomes than leaves. Higher Hg levels can be explained by the existence of bacteria in the rhizosphere, which enhance Hg accumulation in aquatic plants (De Sousa *et al.*, 1999). *P. oceanica* is located at the base of the food web in shallow Mediterranean ecosystems and is probably a main source of contaminants for primary consumers. Therefore, the assessment of contaminants in *P. oceanica* leaves may provide useful indications on the spatial contamination and the potential transfer of these elements to higher consumers in food webs. In contrast to leaves, no difference was observed in rhizomes between study sites (results not shown).

**Tab.1: Mean concentrations of PCBs ( $\mu\text{g kg}^{-1}$  dry mass  $\pm$  SD) and TEs ( $\mu\text{g g}^{-1}$  dry mass  $\pm$  SD) measured in *P. oceanica* leaves from PC (Plateau des Chèvres), PB (Prado Bay), CB (Côte Bleue) and HY (Hyères) from the highest to the lowest human pressure. ns = non-significant.**

Adult leaves	As	Ba	Cd	Cr	Cu	Ni	Pb	Zn	Hg tot	PCBs
PC	$2.5 \pm 0.3$	$6.1 \pm 0.6$	$11.1 \pm 4.4$	$4.3 \pm 0.5$	$9.8 \pm 0.2$	$209.9 \pm 95.8$	$1.7 \pm 0.2$	$458.5 \pm 190.5$	$0.1 \pm 0.002$	$17.1 \pm 0.8$
PB	$2.2 \pm 0.3$	$1.9 \pm 0.2$	$1.9 \pm 0.1$	$1.8 \pm 0.2$	$10.7 \pm 0.8$	$24.8 \pm 1.2$	$8.5 \pm 1.9$	$127.4 \pm 9.7$	$0.1 \pm 0.01$	12.7 -
CB	$4.0 \pm 0.5$	$4.0 \pm 1.3$	$2.1 \pm 0.3$	$2.4 \pm 0.1$	$14.6 \pm 2.7$	$21.5 \pm 1.8$	$2.3 \pm 1.0$	$93.9 \pm 19.8$	$0.1 \pm 0.01$	$13.7 \pm 1.0$
HY	$1.1 \pm 0.1$	$1.1 \pm 0.2$	$1.9 \pm 0.3$	$0.8 \pm 0.2$	$8.6 \pm 1.8$	$23.8 \pm 3.7$	$1.8 \pm 0.3$	$88.8 \pm 13.9$	$0.02 \pm 0.0002$	14.3 -
CB > HY	PC > HY	ns	PC > HY	ns	ns	ns	ns	ns	PC > HY	ns

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## **DEVELOPMENT OF A MEDITERRANEAN MPAS NETWORK THROUGH THE BOOSTING OF MPAS CREATION AND MANAGEMENT (MedMPAnet) - PILOT PROJECT CROATIA**

### **Abstract**

*The MedMPAnet Project has been developed with the objective of 'enhancing the effective conservation of regionally important coastal and marine biodiversity features, through the creation of an ecologically coherent MPA network in the Mediterranean region', as required by the Barcelona Convention's Protocol concerning Specially Protected Areas and Biological Diversity in the Mediterranean (SPA/BD Protocol).*

*The main objectives of the Pilot Project Croatia are to improve MPA management at local level through filling gaps in ecological and fisheries knowledge and better enforcement and monitoring and to assist the Croatian Government in implementing SPA/BD Protocol and developing marine part of Natura 2000 network.*

*The project has brought Mediterranean best practice to the Adriatic through the development of national monitoring protocols for Posidonia oceanica meadows and coralligenous to meet monitoring and reporting requirements of the Habitats Directive, building on UNEP/MAP-RAC/SPA guidelines and standard methods. Habitat map and marine species inventory of one MPA and three Natura 2000 sites were prepared. Assessment of coastal fisheries resources was conducted in the same study areas as the mapping and socio-economic study of local fisheries was conducted through interviews with fishermen. Capacity of regional MPA authority with regard to inventory, mapping and monitoring of marine biodiversity has been built.*

*Direct beneficiaries and partners of the project are the Public Institution Priroda, the Ministry of Environmental and Nature Protection (MENP) and the State Institute for Nature Protection (SINP). Regional Project is coordinated by the Regional Activity Center for Specially Protected Areas (RAC/SPA) and Pilot Project Croatia by the MENP. The project is financed by the European Commission (EC), the Spanish Agency for International Cooperation to Development (AECID) and the French Global Environment Facility (FFEM) and its implementation period in Croatia is 2013-2014.*

**Key-words:** MPA, MedMPAnet, mapping, monitoring, protocol.

### **Introduction**

MedMPAnet Pilot Project Croatia (2013-2014) is part of the Regional MedMPAnet Project (2010-2014), which includes 12 Mediterranean riparian countries. Project partners agreed to develop common actions in support of the implementation of the SPA/BD Protocol.

### **Materials and methods**

Project consists of ecological studies and fisheries studies, all of which were implemented within Primorje-Gorski Kotar County (Northern Adriatic). Marine and coastal habitats and species were mapped using scuba-diving in the area of 7 islands. Presence and depth distribution of habitats, as well as presence of 170 predetermined species, including the invasive green algae *Caulerpa racemosa*, were recorded (Jakl & Prvan, 2014). Monitoring protocols were developed following the unified templates from the Guidelines for preparation of monitoring programs (VV.AA., 2012). Testing of the coralligene protocol included photosampling and visual census to collect data for the characterization of the coralligenous structure and dynamics as well as on

potential disturbances such as mass mortalities, mucilaginous algal aggregates, sedimentation and fishing impacts (Garrabou & Kipson, 2014). Testing of the *Posidonia* protocol included *Posidonia* meadows conservation status assessment, i.e. assessment of coverage and density, presence of other seagrasses, *Pinna nobilis*, alien species, impacts and damages (Guala *et al.*, 2014). Coastal fisheries resources were assessed through analysis of the professional fishermen landings, using logbooks. Socio-economic study of local fisheries focused on interviews with local fishermen on the actual spatial and temporal dynamics of fishing activities and landings (Matić-Skoko & Stagličić, 2013).

## Results

Habitat and species mapping of one MPA and 3 Natura 2000 sites was conducted by Association Sunce. GIS map of habitats and marine species distribution was prepared in GIS shapefile format and habitats were determined in accordance with the National Habitat Classification (Jakl & Prvan, 2014). Monitoring protocols for coralligenous community and *Posidonia oceanica* meadows were developed by the Working Groups for the development of the National Monitoring Protocols. A total of 101 macrobenthic taxa were identified from photographs of coralligenous community (Garrabou & Kipson, 2014). Details on field methods and the equipment used during *Posidonia oceanica* fieldwork are reported in the Field Manual (Puvan *et al.*, 2014). Fisheries resources include 119 fish species. Project conducted first ever study on socio-economic condition of the local coastal fisheries in the area (Matić-Skoko & Stagličić, 2013).

## Discussion / Conclusions

Results of the Project are intended to be used to define necessary measures to regulate the fishing effort in proposed Natura 2000 sites. Data collected will be used as baseline data for future reference and monitoring protocols in fulfillment of Habitats Directive and Marine Strategy Framework Directive requirements, as well as implementation of the SPA/BD Protocol.

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## **BIODEGRADABLE ANCHOR MODULAR SYSTEM FOR TRANSPLANTING *POSIDONIA OCEANICA* CUTTINGS**

### **Abstract**

*Seagrass vegetative transplanting methods involve fixing to the seabed by anchoring the vegetative shoots. Several seagrass restoration projects have been attempted using this method, and most of transplants mortality, particularly in high water motion environments, is precisely attributable to dislodgement. For this purpose, a new and innovative *Posidonia oceanica* (Alismatales, Tracheophyta) transplant system, composed of an artificial underwater support constructed of starch-based biodegradable materials (Mater-Bi), was developed. To evaluate the efficacy of this system, in May 2012 a *P. oceanica* transplant was implemented in an area of high environmental quality (Mondello Bay, Palermo - Italy). In particular, after twenty months from transplanting cuttings survival was 94% and shoot density per unit showed a slight increment than the starting one. These results can be particularly important when large scale transplanting projects are carried out because this anchor system increases success probability and reduces costs.*

**Key-words:** *Posidonia oceanica*, biodegradable support, Sicily, seagrass transplanting.

### **Introduction**

Seagrass meadows are the most widespread and productive coastal ecosystems worldwide and provide a high-value ecosystem service (Vassallo *et al.*, 2013). Recently, a seagrass decline rate of 2-5% yr<sup>-1</sup> was globally estimated (Waycott *et al.*, 2009); in particular, for the Mediterranean basin, *Posidonia oceanica* areal extent appears to be lost (between 13% and 50%), with a decrease of shoot density of remaining meadows by 50% for the last 20 years (Marbà *et al.*, 2014). Thus, recovery planning are required to arrest and reverse the decline of this species. For mitigating the continuous loss of this seagrass habitat, restorations through transplanting projects have been suggested (Duarte *et al.*, 2013). Since the 1970s, reforestations with *P. oceanica* have been carried out to assess various transplanting techniques and most of the failures of *P. oceanica* transplants (80%) has been ascribed to anchor system (Park & Lee, 2010). Here we show a new innovative technique developed for the restoration of *P. oceanica* meadows based on the use of an underwater anchor modular system constructed of starch-based biodegradable materials (Mater-Bi), in order to: i) speed up the natural growth and colonization of an area by the plant, ii) increase transplant success probability limiting costs, and iii) reduce the impact on marine environment.

### **Materials and methods**

Study area is located in the Mondello Bay (Palermo, Sicily, Italy). The seabed is characterized by sand, colonized by a *P. oceanica* meadow and *Cymodocea nodosa*, and *P. oceanica* dead matte (Calvo *et al.*, 1993). In order to evaluate the feasibility of a newly

transplanting technique, in May 2012 *P. oceanica* shoots have been transplanted by using an innovative product (patent pending by Biosurvey Srl, Spin-off University of Palermo) realized with biodegradable plastic (bioplastic Mater-Bi). This system consists of a radial structure fixable on the seabed by a picket (Fig.1). The structure is modular with six arms on which a variable number of clips for optimal fixation of *P. oceanica* cuttings occurs. Ten modular systems with 30 cuttings and about 130 shoots per unit have been positioned at a depth of 6 m on two substratum typologies: sand with *C. nodosa* and *P. oceanica* dead *matte*. Transplant has been monitored monthly up to December 2013 by evaluating survival, detachment and mortality of cuttings, and shoot density.

### Results and Discussion

Transplant monitoring showed no significant differences between substrata. In particular, both on sand with *C. nodosa* and *P. oceanica* dead *matte*, after twenty months from transplanting, cuttings survival was 94%, both mortality and detachment were about 3%, and shoot density per unit showed a slight increment than the starting one. Therefore, *P. oceanica* anchor modular system allows the rapid attachment and expansion as evidenced by high survival rates and density increasing. This new developed *P. oceanica* system was successful and effective for our transplanting. Indeed, although

*P. oceanica* is slow-growing seagrass, after 1 year from transplanting an increase of shoot density has been detected. Moreover, anchor system, made with a new generation of bioplastic, is totally biodegradable maintaining the same physical characteristics of plastics. This new material, unlike that of other transplant systems, does not release harmful residues in the environment. In addition, biodegradation of the system is compatible with cuttings rooting and their stabilization into the substratum. Therefore, transplanting by using this artificial underwater anchor system may be an effective technique to successfully restore *P. oceanica* habitat.

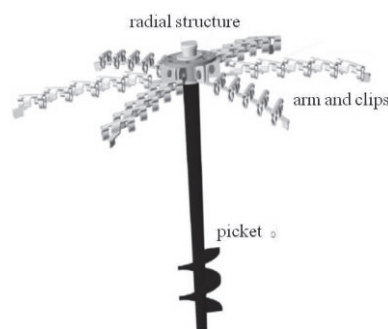


Fig. 1: Anchor modular system.

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# LES MACROALGUES COMME INDICE DE QUALITE POUR L'EVALUATION DE L'ETAT ECOLOGIQUE DE LA COTE ALGERIENNE

## Résumé

*L'étude qualitative et quantitative des communautés phytobenthiques de la côte algérienne est réalisée dans le but de mettre en évidence l'état écologique de la côte. Ainsi, la qualité de l'eau a été estimée par les espèces présentes (indicatrices de pollution et milieu perturbé) et par les indices écologiques (indice de Shannon :  $H'=2,5$ bits ; Equitabilité :  $E=0,4$  ; Indice de Sorensen :  $Is=0,4$ ). Une analyse multivariée distingue les différents groupements d'espèces déterminant une qualité de l'eau en relation avec les activités anthropiques et les conditions environnementales. La zone littorale côtière semble être une zone de haute sensibilité aux agressions anthropiques. Ce constat repose sur la faible richesse floristique, ( $Q=25$  espèce en moyenne), qui révèle un état environnemental de qualité médiocre, liée à l'impact des activités anthropiques.*

**Mots-clés** : Macroalgues, phytobenthos, richesse spécifique, activités anthropiques, impact, Algérie.

## Introduction

Les écosystèmes benthiques sont le lieu d'accumulation des perturbations d'origine anthropiques. Pour les espèces sensibles à la pollution, la présence de ces végétaux démontre la qualité de l'eau. La prolifération d'espèces dites nitrophiles démontre également la mauvaise qualité de l'eau, d'un milieu perturbé, ou d'un milieu eutrophe. L'utilisation d'indicateur écologique, pour déterminer la qualité du milieu marin, apparaît comme une méthode pratique et performante. L'objectif est d'estimer et caractériser les macroalgues afin d'en apprécier la qualité de l'eau. Les macroalgues constituent des peuplements différents selon l'exposition à l'hydrodynamisme, où se distinguent les peuplements de mode abrité et ceux de mode battu.

## Matériel et méthodes

L'étude porte sur des sites répartis sur la côte algérienne. Ces sites sont soumis à l'influence du courant qui draine les produits des activités anthropiques. L'échantillonnage est de type saisonnier et réalisé à une profondeur variant de -0.5m à -2m. Les paramètres pris en considération, sont essentiellement : la diversité spécifique et les indices de diversité. Une analyse multivariée est utilisée pour décrire la qualité de l'eau et les communautés des macroalgues benthiques.

## Résultats et discussion

L'analyse des différents prélèvements a permis d'identifier 42 espèces réparties en 29 Rhodophyta (69%), 7 Ochrophyta (17%) et 6 Chlorophyta (14%). Les Rhodophyta restent dominants quelque soit la nature et le mode du site. Les Ceramiales sont l'ordre dominant (35.7%), les espèces les plus souvent rencontrées sont : *Laurencia papillosa*, et *Ceramium* ssp suivi des Corallinales (14.3%): *Corallina elongata*, *Jania rubens* et *Lithohyllum*



*incrustans*. Celles-ci recouvrent des surfaces appréciables ; d'où, une bonne vitalité et adaptation aux conditions environnementales. Aussi, l'apparition de *Corallina elongata* indique la présence de mauvaise qualité des eaux. Pour les Ochrophyta ce sont les Fucales qui dominent (7%), avec *Cystoseira compressa* et *Sargassum vulgare*. Pour les Chlorophyta, ce sont les Cladophorales (9.52%) et les Ulvales (2.38%) avec *Ulva rigida*. Cette richesse spécifique évolue en fonction des apports terrigènes, véhiculées par les eaux usées. La présence d'un peuplement à *Corallina elongata* ou à *Cystoseira compressa* montre la sensibilité des espèces à la qualité de leur environnement et à la qualité du sédiment qui piège les éléments polluants. La densité du peuplement diminue par la disparition des espèces sensibles à la pollution. Les indices de diversité utilisée ( $H'$  et  $E$ ) ont confirmés l'état du peuplement étudié. Ces indices sont relativement faible ( $H'=0.5$  à  $2.5$  bits et  $E=0.1$  à  $0.4$ ). Ce qui traduit l'instabilité du peuplement, il est considéré comme pollué si l'on se réfère à la classification de Simboura et Zenetos (2002). L'indice de Sorensen ( $Is=0.4$ ) montre l'homogénéité du peuplement algal. Il est admis que le long d'un gradient d'eutrophisation le nombre d'espèces diminue mais la biomasse des espèces tolérantes augmente (Pearson et Rosenberg, 1978). La classification hiérarchique ascendante (Fig.1) montre deux catégories : une catégorie constituée par : le groupes I rassemblant les espèces tolérants à la pollution, (*Ceramium tenerimum*, *Gigartina acicularis*, *Sphacelaria tribuloides*). Le groupe II formé par les espèces indicatrices de pollution (*Pterocladie capillecea*, *Cystosira compressa*, *Corallina elongata*). La deuxième catégorie formée par : le groupe III constitués par les espèces tolérantes à la pollution (*Jania rubens*, *Ceramium ssp*, *Cladophora albida*, *Bryopsis muscosa*).

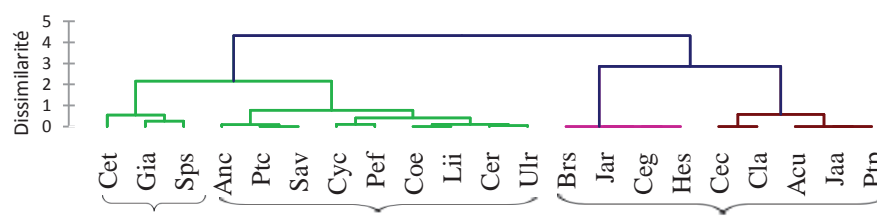


Fig.1 : Classification Ascendante Hiérarchique du peuplement algal

## Conclusion

Les données obtenues reflètent les conditions environnementales des différents sites. Cependant, la turbidité de l'eau qui est liée à l'apport de matières en suspension et au sédiment n'a pas été évaluée et pourrait jouer un rôle dans la répartition des macroalgues, voir l'état écologique des écosystèmes.

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## **CYMODOCEA NODOSA DISTRIBUTION ALONG THE LEBANESE COAST**

### **Abstract**

Two marine surveys of Magnoliophyta along the Lebanese coast (about 160 km long) as a component of the biological monitoring in Enfeh Peninsula, Ras Chekaa cliffs, Raoucheh cliffs, Sidon, Tyre and Naqoura were conducted in 2012 and 2013. They were undertaken within the framework of the Regional Project for the Development of a Mediterranean Marine and Coastal Protected Areas (MPAs) Network through the boosting of MPA Creation and Management (MedMPAnet Project), led by RAC/SPA. During these surveys along the six sites of the Lebanese coast, *Cymodocea nodosa* was encountered in 33.7% (16/48) of the stations and was recorded only in three localities: Enfeh Peninsula and Ras Chekaa cliffs in the northern coast and Tyre in the northern coast. *C. nodosa* was common around Enfeh area but less common in Ras Chekaa. On the contrary, it has been rare in the southern sector of Lebanon, where it has been observed only in front of the south beach of Tyre. *C. nodosa* colonises the sandy and muddy sand bottoms and their meadows are developed in shallow waters (1-4m depth). It is noteworthy the abundance of germinated seeds in June of 2012, that colonised deeper sediments (as far as 31m depth).

**Key-words:** Lebanon coast, biological monitoring, Magnoliophyta, *Cymodocea nodosa*.

### **Introduction**

Lebanon is a Middle Eastern country located on the eastern coast of the Mediterranean Sea. This part of the Mediterranean Sea is characterized by a high level of salinity and temperature (Marullo et al., 2006). Three seagrasses were recorded in Lebanon coast *Zostera noltii*, *Cymodocea nodosa* and *Halophila stipulacea* (Bitar and Kouli-Bitar (2001). The ecology of *C. nodosa* populations is relatively well known in the Atlantic Ocean, the Western Mediterranean Sea and the Adriatic Sea. However, the knowledge of *C. nodosa* in the eastern Mediterranean Sea is limited. Furthermore, the distribution, the plant size shoot density and the biomass of *C. nodosa* meadows in the Lebanese coast Sea has not been studied so far.

### **Materials and methods**

Shoot density was measured using 20x20cm plots (10 replicates). The plant biomass (above and belowground) was collected using a 20x20cm plots (5 random replicates). Twenty shoots were collected randomly at the sampling station to measure plant morphometrics.

## Results

During these surveys along the six sites of the Lebanese coast, *Cymodocea nodosa* was encountered in 33.7% (16/48) of the stations and was recorded only in the three localities: Enfeh Peninsula (in a small port) and Ras Chekaa cliffs (in front of Beny beach and Florida hotel), in the northern coast and Tyre, in the southern coast. Furthermore, sparse shoots of *Cymodocea* were also observed in depth area (11m) between Enfeh Peninsula and Ras Chekaa. On the other hand, *Cymodocea* has been rare in the southern sector of Lebanon, where some shoots had been observed only in front of the south beach of Tyre colonises sandy and muddy sand bottoms. At Enfeh Peninsula, the areas colonised by *C. nodosa* were very small, covering respectively 15 m<sup>2</sup> (3×5m) and 2.25m<sup>2</sup> (1.5x1.5m). At Ras Chekaa, *C. nodosa* was more present with a very low density less than 5 shoot m<sup>-2</sup>. No other meadows were found during the survey. Biometric data for the prospected meadows in Enfeh Peninsula and Ras Chekaa were provided in Tab. 1.

**Tab. 1: Biometric data (± SD) for Enfeh Peninsula and Ras Chekaa**

	Enfeh Peninsula	Ras Chekaa	
		Beny beach	Florida hotel
Density (Shoot m <sup>-2</sup> )	1567±80	5	1
Number of leaves per shoot	4,25±0,35	-	-
Differentiated leaves per shoot	2,35±0,20	-	-
Intermediate leaves per shoot	1,9±0,2	-	-
Mean adult leaf length (mm)	57,05±12,95	-	-
Mean adult leaf wide (mm)	1,2±0,15	-	-
Mean intermediate leaf length (mm)	44,8±14,35	-	-
Leaf area index (m <sup>2</sup> m <sup>-2</sup> )	0,15±0,01	-	-
Shoot biomass (g DW m <sup>-2</sup> )	41,6±14,4	-	-
Below-ground biomass (g DW m <sup>-2</sup> )	74,86±3,74	-	-

## Conclusion

*C. nodosa* at the present study site exhibited a lower value in morphology, shoot density and biomass of summer period. This could be related to the extreme environmental condition in this part of the Mediterranean which is the limit of the distribution of the *Cymodocea nodosa*.

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## **UNDERSTANDING RESIDENTS' PERCEPTION OF MARINE BIODIVERSITY AND THEIR ATTITUDES TOWARDS BIODIVERSITY CONSERVATION INSTRUMENTS: THE CASE OF THE VENETIAN TEGNÙE**

### **Abstract**

*Understanding the importance of marine biodiversity and its value are fundamental for designing appropriate conservation and protection policies. Also, the success of these policies is closely related to how people perceive marine biodiversity. This study aims to understand what local residents know and think of a particular marine ecosystem in the North Adriatic Sea, locally named Tegnùe.*

**Key-words:** Marine biodiversity; Perception and attitude, Marine conservation, focus groups

### **Introduction**

Coastal and marine ecosystems provide environmental goods and services that are essential for our well-being (Beaumont *et al.*, 2007). These important ecosystems hold high rate of biodiversity but they are also highly vulnerable and under the pressure caused by human activities and natural factors. Understanding the importance of marine biodiversity and its value are fundamental for designing appropriate conservation and protection policies. On the other hand, the implementation and the success of these policies are closely related to how people perceive marine biodiversity. Understanding people's attitudes is important to researchers because attitudes determine how people perceive and implement actions. Researchers have shown particular interest in understanding public attitudes towards environmental problems as demonstrated by numerous studies published by social scientists. The economic literature shows that biodiversity perception is not uniform across the population, and such diversity is likely to impact upon methods to catalyse societal engagement with marine conservation. The aims of this contribution are to present the results of several focus groups organized with local residents to better understand people's knowledge of a particular marine ecosystem localized in the North Adriatic Sea, the *Tegnùe*, and to investigate their perception regarding biodiversity conservation policies and measures.

### **Materials and methods**

The study was conducted in the North Adriatic Sea characterized by shallow waters with sandy seabed and some rocky habitats, locally named *Tegnùe*, in which there is high biodiversity richness. However, these areas are nowadays experiencing a loss of biodiversity as a consequence of human activities such as over-fishing and environmental contamination. In particular, among the negative impact on the conservation of local biodiversity there is the loss of fishing equipment (e.g. nets, traps, metal tools), a phenomenon known as Abandoned, Lost or Discarded Fishing Gears (ALDFG). The ALDFG causes considerable damages to the natural environment, among which the covering nesting places, and accidental entrapment of marine protected species.

Our work presents the results of four focus groups organized in February and May 2014 with the residents of Venice. In total, we selected 32 participants among the population with the help of a professional marketing research firm. The purpose of these meetings was to reveal the extent to which the residents were aware of the existence of Tegnùe ecosystem and its biodiversity. People's perceptions of marine biodiversity were generated by asking them several questions about the knowing of the areas, the main benefits it provides and the threats suffered by the ecosystem, and, finally, which interventions they will support or not for protecting this ecosystem.

### **Results**

We found that only few people had some personal and direct experienced with the *Tegnùe*. But almost all participants were surprised of the great variety of species hosted by *tegnùe*, and they were even more surprised by the fact this big variety of biodiversity could be found in the Italian Adriatic Sea, and so closed to them. All participants agreed that this beautiful ecosystem has also a lot of vital functions and provides important environmental goods. On the other hand, the Tegnùe is a delicate and vulnerable environment that is threatened by different causes that, in the opinion of our respondents, are mainly pollution, waste abandoned, excessive over extraction of natural resources, and destructive fishing practices. The instruments and policies people preferred for reducing the decline of biodiversity and the environmental damages in these habitats are the reduction of pollutant discharges, to put some restrictions on fishing activities, to design a marine reserve, and to educate and provide more information to the residents. People strongly agreed to the potential establishment of a marine park, and they are firmly convinced that the best managing authority is the environmental organizations or the research institutions but not the central or regional governments.

### **Conclusions**

In the scientific literature there is a general consensus that the lack of knowledge on biodiversity is considered one of the main causes of biodiversity loss (de Groot et al., 2012). But the valuation and understanding of biodiversity are complicated, especially for common people, thus it is necessary to investigate them in a different way. Several studies demonstrated that environmental governance of biodiversity is affected by people's perception (Vodouhê 2010) that is influenced by many factors such as the education level, the community involvement in management strategies, the degree of awareness of marine biodiversity, and many others. This research aimed to understanding the factors which influence people's connections with marine biodiversity, and their attitudes towards biodiversity conservation and protection.

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## ON SOME PECULIAR MACROALGAL COMMUNITIES FROM GREECE

### Abstract

*In pristine areas of the Mediterranean Sea typical macroalgal populations form dense communities in the shallow infralittoral zone, mainly dominated by the canopy-forming *Cystoseira* species. During an extensive macroalgal survey along the Greek coasts 3 rare macroalgal communities were detected. 1) *Sargassum acinarium* forest: a population of extremely large individuals of this brown alga, up to 2 m in height, formed a dense and luxuriant "forest" in 3-6 m depth, hosting abundant and diverse epiphyte flora and fauna. 2) *Tenarea tortuosa* "trottoire": this red calcified alga formed extensive, broad and continuous formations, similar with that of the genus *Lithophyllum*, up to 30 m long, along an exposed rocky upper infralittoral zone (0-1 m depth). 3) *Penicillus capitatus* beds: massive abundances of this green alga were found to monopolize the sea bottom of a protected bay, growing on dead and mostly buried *Posidonia oceanica* meadow between 5-30 m depth.*

**Key-words:** *Sargassum acinarium*, *Tenarea tortuosa*, *Penicillus capitatus*, Eastern Mediterranean

### Introduction

In pristine areas of the Mediterranean Sea typical macroalgal populations form dense communities in the shallow infralittoral zone, mainly dominated by the canopy-forming *Cystoseira* species (Pérès & Picard, 1964). However, in more rare cases other species can form characteristic macroalgal communities. In the current study we present three peculiar macroalgal communities from Greece (Eastern Mediterranean Sea).

### Materials and methods

The macroalgal communities were detected during an extensive macroalgal survey along the Greek coasts. Sampling was performed in the infralittoral zone, from surface down to 30 m depth through scuba-diving, both on rocky and sandy substratum, during spring time.

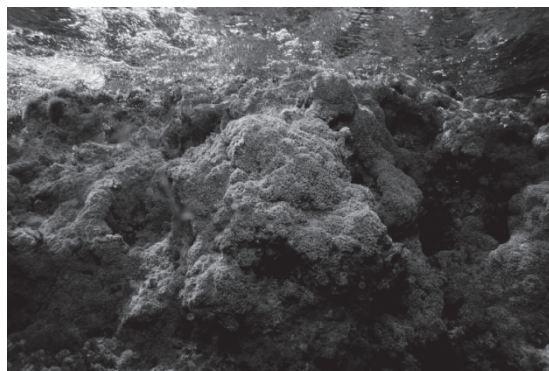
### Results

1) *Sargassum acinarium* forest (Fig. 1): a population of extremely large individuals of this rare brown alga, up to 2 m in height, formed a dense and luxuriant "forest" in 3-6 m depth, hosting abundant and diverse epiphyte flora and fauna. *S. acinarium* thalli were growing on rocks along with *Cystoseira foeniculacea* and *C. elegans*, in a protected bay in Elafonissos Island, South Peloponnesus (36.491243°N, 22.940955°E). 2) *Tenarea tortuosa* "trottoire" (Fig. 2): this red calcified alga is restricted in the Eastern Mediterranean Sea. It formed extensive and broad formations, up to 30 m long, along the rocky upper infralittoral zone (0-1 m depth), at an exposed coast of Lindos, Rhodes Island (36.068624°N, 28.078766°E). These formations were continuous, forming a "trottoire" habit, similar with that of the genus *Lithophyllum*. *Jania* spp. and *Laurencia* spp. were growing as epiphytes on *T. tortuosa* thalli. 3) *Penicillus capitatus* beds (Fig. 3): massive

abundances of this rare green alga were found to monopolize the sea bottom of a protected bay in Aliveri, Evvoia Island (38.372780°N, 24.053043°). The species was growing on dead and mostly buried *Posidonia oceanica* meadow between 5-30 m depth.



**Fig. 1: *Sargassum acinarium* forest in Elafonissos Island, South Peloponnesus.**



**Fig. 2: *Tenarea tortuosa* "trottoire" in Lindos, Rhodes Island.**



**Fig. 3: *Penicillus capitatus* beds in Aliveri, Evvoia Island.**

### **Discussion**

Both *Sargassum acinarium* and *Tenarea tortuosa* are included in the list of endangered / threatened species of the Annex II of the Barcelona Convention. Although individuals of *S. acinarium* and *T. tortuosa* can be found scattered along the Greek coasts, this is the first time that dense and extensive formations of these species are recorded from Greece. Thus, taking into account the rarity of the above species we state that these communities should be protected. Regarding *Penicillus capitatus*, we claim that extensive communities of this green alga should be more common than already recorded, but remain overlooked due to its sandy and kind of deep habitat.

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## **ECOLOGICAL QUALITY CLASSIFICATION of AEGEAN AND MEDITERRANEAN SEAS IN TERMS OF INVASIVE FISH SPECIES**

### **Abstract**

*The aim of this study was to investigate ecological quality classification and demersal fish fauna of the Aegean and Mediterranean coastal waters of Turkey. Fish samples were collected by bottom trawl. Among 106 fish species which was sampled, 81 fish species were recorded in the Aegean Sea, 68 in the Mediterranean Sea and 43 species were shared in these two. 19 of 68 fish species in the Mediterranean Sea was Indo-Pacific migrant species. It can be evaluated that the effects of Red Sea seems to dominate when it is compared with the anthropogenic factors in Mediterranean Sea.*

*In conclusion, ecological status of Mediterranean and Aegean Seas based on fish parameters are evaluated according to Shannon Weiner index, population parameters and expert decision. Evaluation is based on Water Framework Directive criteria.*

**Key-words:** Fish fauna, ecological quality, Aegean Sea, Mediterranean Sea, Turkey.

### **Introduction**

Fisheries is one of the important economic sources for Turkey. Monitoring of the ichthyofauna, with immigrant of Red Sea cannot be evaluated without taking into account these immigrant fishes. The aim of this study was to investigate ecological quality classification and demersal fish fauna of the Aegean and Mediterranean coastal waters of Turkey.

### **Materials and methods**

Sampling of the demersal fish fauna was conducted in coastal waters of Mediterranean and Aegean Sea at 20 meters depth. Sampling had been conducted with research vessel at 14 stations from Mediterranean coast and 11 stations from Aegean coast which were suitable for trawling with 30 minutes. Identification of the fish was done according to Whitehead et al., 1984-1986; Fischer et al., 1987. Shannon Weiner biodiversity index analysis at PAST software (Hammer et al., 2001).

### **Results**

Demersal fish fauna of Aegean Sea comprised of 81 species belonging to Osteichthyes (68 species) and Chondrichthyes (13 species) classes. A total of 12718 specimens were collected in sampling localities at Aegean Sea. Mediterranean Sea samples were made up of 6889 specimens from 68 species belonging to Osteichthyes (61 species) and Chondrichthyes (7 species) classes. Shannon Weiner biodiversity index scores of Aegean and Mediterranean Seas were summarized at Figure 1.

Ecological states of Aegean and Mediterranean Seas by the summarizing of Shannon Weiner biodiversity index, population parameters of demersal fish fauna and expert opinion were done by transformation of '5 classed Water Framework Directive criteria' to a '3 class criteria' with classes as 'good (5)', 'fair (3)' and 'bad (1)'.

### **Discussion and Conclusion**

Aegean Sea ichthyofauna seems to exhibit higher biodiversity than Mediterranean with more established populations. According to results of this research no lessepsian fish was sampled



from Aegean waters although there are some reports (Keskin et al.; Bilecenoglu et al., 2002). This better establishment of the fauna seem to prevent an easy colonization of the area. Aegean Sea faunal composition was mainly affected by the outflow of cold, less saline and highly productive water currents from the Black Sea.



**Fig. 1: Sampling stations from Aegean and Mediterranean Seas colored according to criteria.**

In Mediterranean Sea 19 lessepsian fish comprise 28% of the species. These immigrant fishes follow a northern path in Mediterranean Sea, in coastal waters of Turkey avoiding cyclonic coastal currents and hydrologic barrier formed by discharge of Nile River. (Golani, 1998). According to these reports from literatures and this research Red Sea immigrants seem to dominate eastern Mediterranean ichthyofauna and widening their distribution through the western part. This study aims to demonstrate the recent status of Mediterranean and Aegean Seas by means of demersal fish fauna.

### Acknowledgments

Authors thanks to Turkish Ministry of Environment and Urbanization and Alka Environmental Laboratories for their financial supports.

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## **COMARATIF STUDY OF TRACE METAL DISTRIBUTION IN *POSIDONIA OCEANICA*, *CYMODOCEA NODOSA* AND SEDIMENTS FROM EL KANTAOUI (EAST OF TUNISIA)**

### **Abstract**

*Accumulation of trace metals (Pb, Cu, Cd, Ni and Zn) was measured in sediments, leaves of Cymodocea nodosa and Posidonia oceanica collected from El Kantaoui (Eastern coast of Tunisia). Our results show that metal concentrations in sediments are very low compared with seagrass. However, Posidonia oceanica leaves accumulate more trace metals than leaves of Cymodocea nodosa. Levels of metals in the studied seagrass were similar to those reported by other authors in uncontaminated areas of the Mediterranean Sea. This study reinforces the usefulness and the relevance of Posidonia oceanica as an indicator of spatial metal contamination and an interesting tool for environmental quality evaluation.*

**Key-words:** Trace element; Seagrass; Mediterranean Sea; ICP-OES.

### **Introduction**

Magnoliophytes are important contributors to primary production in marine ecosystem and the meadows constitute the basis of the richness of coastal waters in the Mediterranean Sea maintaining the equilibrium of coastal waters and their concomitant economic activities. Furthermore, they are exceptionally fragile and more vulnerable to disturbance than other benthic populations. Seagrass may serve as heavy metal reservoirs taking up metals both from water through leaf surfaces and their roots. Seagrass leaves especially play an important role in the cycling of metals, and are probably the main source of metals for seagrass consumers (Boudouresque *et al.*, 2006). The aim of this study is to determine and to compare trace metals concentrations in sediment, leaves of *P. oceanica* and *C. nodosa*.

### **Materials and methods**

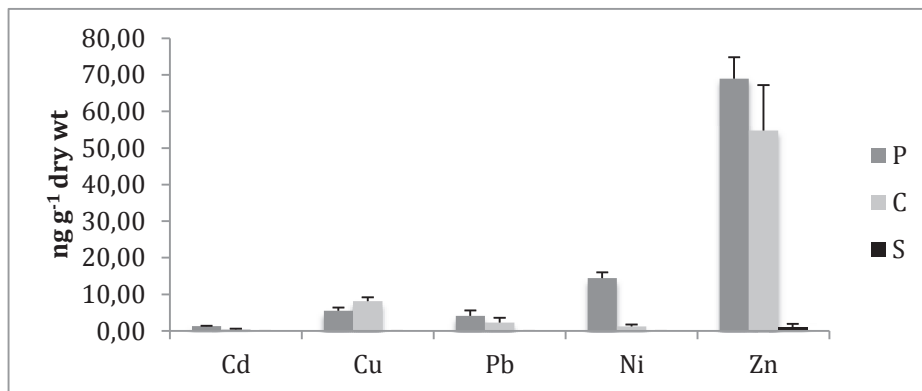
Samplings were collected in 4 stations along the coast of port El Kantaoui (N35.896°E10.609°) by scuba diving in March 2012 between 3 and 10 m of depth. In each station, 6 samples of superficial sediment were collected and the fractions of sediment inferior to 2mm were analysed. Three pools of 15 *P. oceanica* were collected in each station. Only leaves were analysed, epiphytes were removed using a glass strip. Leaves samples were lyophilized, reduced to powder then analysed by atomic absorption spectrometry. The same treatment is done for *C. nodosa* leaves samples. Five trace metals were tested in this study Pb, Cu, Cd, Ni and Zn.

### **Results**

Seagrass leaves collected around the port El Kantaoui at the four sampling sites contained the five studied metals. However, in the sediment samples only Zn was detected. Mean metals concentrations in the leaves of *P. oceanica* whatever the sampling station



decreased in the following order: Zn > Ni > Cu > Pb > Cd (Fig. 1) according to the sequence commonly observed by Conti *et al.* (2010). *P. oceanica* leaves accumulated more metal than leaves of *C. nodosa*, except for Cu. However, this difference is statistically significant only for Ni.



**Fig. 1: Mean metal concentration in sediment, *P. oceanica* and *C. nodosa* leaves around El Kantaoui port**

### Discussion and conclusions

Seagrass is often considered as a useful metal bioindicator. This observation is confirmed by the present study. Indeed, comparison between concentrations of trace metals in two seagrass species and sediment show that *P. oceanica* may have a greater bioaccumulation capacity than sediment or *C. nodosa* (except for Cu).

### Acknowledgments

The present study was funded by MAPMED project (I-B/2.1/395).

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# **RECOMMENDATIONS**

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# **RECOMMANDATIONS**



## RECOMMENDATIONS OF THE 5<sup>th</sup> MEDITERRANEAN SYMPOSIUM ON MARINE VEGETATION

### Scientific interest in the Action Plan

Generally speaking, there has been increased interest in the knowledge of marine plant species and growing scientific attendance, plus greater geographical diversity, at the 'Marine Vegetation' Symposium.

Most of the papers (81%) concern marine magnoliophytes, particularly *Posidonia* meadows (59%), though there is growing interest in other marine magnoliophytes (19%), particularly the Fucales, notably the *Cystoseira* genus.

In the papers, the major role played by Marine Protected Areas in conserving marine plants was highlighted, but the continuation of illegal trawling practices constitutes a significant danger to plant formations, especially *Posidonia* meadows.

### Recommendations

1 – Much has been done regarding the knowledge and monitoring of plant formations, and new data is now available even though there is still a lack of balance between the countries of the north-western Mediterranean and those of the southern shore and the eastern basin. Efforts here must be kept up; particularly the need to integrate this data to adapt the existing protocols to the specific features of these sub-regions (defining reference conditions, oligotrophy of water, etc.).

→ Pursue and amplify the procedure initiated by RAC/SPA within the framework of the Med*Posidonia* and MedKeyHabitats programmes, with particular attention paid to the eastern basin and the southern shore of the Mediterranean.

2 – The scientific community highlighted the general regression of Fucales (e.g. *Cystoseira* forests, gulfweeds) even though putting the *Cystoseira* genus on Annex II of the SPA/BD Protocol has been a significant advance. As well as taking into account such regulatory aspects, it is necessary to enhance national capacities to get a Mediterranean-scale overall vision.

→ Mobilise all the actors as to the interest of these formations (distribution, dynamics) and the need to identify the origin of the regressions observed, in order to suggest concrete steps likely to solve this problem (e.g. being taken into account in the context of impact studies).

3 – Regulations that are indispensable for conserving vegetation exist in almost all the countries. However, the difficulties encountered in enforcing these persist, particularly because of illegal fishing activities. A special effort must be made to make the Focal Points aware of this, for this lacuna is responsible for significant regressions.



→ Draw up an updated assessment of the direct and indirect regulatory measures (regulating human-origin activities that affect these formations) that aim at protecting and conserving the species in the Action Plan on the Conservation of Mediterranean Marine Vegetation, working alongside the General Fisheries Commission for the Mediterranean.

4 – Guidelines for mapping and monitoring are an interesting element but there is still a need for simpler, more robust and non-destructive tools to assess the conservation state of marine plants. Similarly, synergy of actors (scientists, managers) and the widest possible provision of the available data would certainly enable the means to be optimized. The need to have long-term monitoring (chronological series) was recalled; it remains an indispensable precondition for better grasping the changes that have been observed.

→ Make exchange of data possible, especially under the aegis of RAC/SPA, by improving the functionality of the current internet site.

## RECOMMANDATIONS DU 5<sup>ème</sup> SYMPOSIUM MEDITERRANEEN SUR LA VEGETATION MARINE

### Intérêt scientifique pour le Plan d'Action

De manière générale, une augmentation de l'intérêt porté à la connaissance des espèces végétales marines et une fréquentation croissante, allié à une plus grande diversité géographique, des scientifiques participant au symposium « végétation marine » sont enregistrées.

Le plus grand nombre de communications concerne les magnoliophytes marines (81%), et plus particulièrement l'herbier à *Posidonia oceanica* (59%), même si un intérêt croissant est noté pour les autres macrophytes marines (19%) et notamment les fuciales dont le genre *Cystoseira*.

Au cours des communications le rôle majeur joué par les Aires Marines Protégées dans la conservation de la végétation marine a été souligné mais la poursuite de pratiques de chalutage illégal constitue un danger significatif pour les formations végétales et notamment les herbiers de Posidonies.

### Recommandations

1 – D'importants efforts ont été fait en matière de connaissance et de suivi des formations végétales et de nouvelles données sont d'ores et déjà disponibles même si un déséquilibre persiste entre les pays de la rive Nord occidentale de la Méditerranée et ceux de la rive Sud et du bassin oriental. Les efforts en ce sens doivent être maintenus, avec en particulier la nécessité d'intégrer ces données pour adapter les protocoles existants aux particularités de ces sous-régions (définition des conditions de références, oligotrophie des eaux,...).

→ Poursuivre et amplifier la démarche initiée par le CAR/ASP dans le cadre des programmes Med*Posidonia* et MedKeyHabitats, avec un intérêt particulier pour le bassin oriental et la rive sud de la Méditerranée.

2 – La communauté scientifique a souligné la régression généralisée des fuciales (ex forêts de *Cystoseires*, sargasses) et ce, même si l'inscription du genre *Cystoseira* sur les annexes II du protocole ASP/DB constitue une avancée significative. Au delà de la prise en compte de ces aspects réglementaires, un renforcement des capacités nationales est nécessaire pour disposer d'une vision globale à l'échelle de la Méditerranée

→ Mobiliser l'ensemble des acteurs sur l'intérêt de ces formations (distribution, dynamique) et sur la nécessité d'identifier l'origine des régressions observées afin de proposer des mesures concrètes susceptibles d'y remédier (ex. prise en compte dans le cadre des études d'impacts).

**3** – La réglementation indispensable à la conservation de la végétation existe dans la quasi-totalité des pays. Néanmoins des difficultés dans son application persistent, en particulier du fait d'activités illégales de pêche. Un effort particulier doit être fait pour sensibiliser les points focaux car cette lacune est responsable de régressions significatives.

→ Etablir un bilan actualisé des mesures réglementaires directe et indirecte (réglementations des activités anthropiques impactant ces formations) qui visent à la protection et à la conservation des espèces du Plan d'Action relatif à la conservation de la végétation marine de Méditerranée, en concertation avec la Commission Générale des Pêches de Méditerranée.

**4** – Les lignes directrices pour la cartographie et la surveillance constituent un élément intéressant mais le besoin d'outils plus simples, plus robustes et non destructifs persiste pour évaluer l'état de conservation de la végétation marine. De même, la mise en synergie des acteurs (scientifiques, gestionnaires) et la mise à disposition la plus large possible des données disponibles permettraient sans doute d'optimiser les moyens. La nécessité de disposer de suivis à long terme (séries chronologiques) a été rappelée et reste un préalable indispensable pour mieux appréhender les changements observés.

→ Rendre possible l'échange des données notamment sous l'égide du CAR/ASP en améliorant les fonctionnalités du site internet actuel.

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