

## MARINE MACROPHYTES AND THE WATER FRAMEWORK DIRECTIVE: PRELIMINARY STUDIES IN SOUTHERN SPAIN

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### Abstract

The Water Framework Directive (WFD) includes angiosperms and macroalgae as biological quality elements (BQE) for coastal and transitional waters. In southern Spain (Andalusia), studies to implement the WFD using angiosperms (seagrasses) started in June 2009. Different approaches have been considered; the first was the variation of this BQE along an environmental gradient; the second consisted on an extensive sampling campaign in the coastal waters from the western province of Huelva to the eastern province of Almería. The surveys showed marked differences among the sites and species, indicating a broad variation in the ecological quality values for the water bodies. Additionally, these results will be compared with a similar research conducted on Ria Formosa (Portugal) to provide preliminary classifications of the ecological status of coastal and transitional waters of the southern Iberian Peninsula.

With respect to the BQE "macroalgae", two indices; the CFR (quality of rocky shores) and RSL (reduced species list) have been estimated. The ecological differences between coastal zones in the North Atlantic ecoregion where these methods of assessment have been developed (Northern Spain and the United Kingdom, respectively) and the Atlantic coast of Cadiz, suggested the need for an adaptation of both indices in southern Spain. These extensive surveys have allowed the elaboration of a reduced species list for the rocky shores on the southwestern Spanish Atlantic coasts.

The present study shows the preliminary efforts and results of the assessment of BQEs based on marine macrophytes in Southern Spain. Some concerns about the need of reassessment of the proposed tools from the North Atlantic ecoregion when applied to Andalusian water bodies are also discussed.

**Keywords:** *Andalusia, Biological Quality Elements, macroalgae, seagrasses*

### Resumen

La Directiva Marco de Agua (DMA) incluye como elementos de calidad biológica (BQE) de las aguas de transición y costeras a las angiospermas y macroalgas. En el sur de España (Andalucía), los trabajos para implementar la DMA utilizando angiospermas marinas comenzaron en Junio de 2009. Se han considerado distintas aproximaciones; la primera fue el estudio de los cambios de este BQE a lo largo de un gradiente medioambiental; la segunda consistió en una intensa campaña de muestreo en las aguas costeras desde la provincia más occidental (Huelva) hasta la más oriental (Almería). Las campañas mostraron diferencias notables entre los sitios y las especies, lo que implicó una amplia variación en los valores de calidad ecológica de las masas de agua. Adicionalmente, estos resultados se compararán con un trabajo similar desarrollado en Ria Formosa (Portugal) de forma que se pueda proporcionar una clasificación preliminar del estado

ecológico de las aguas costeras y de transición del sur de la Península Ibérica. Con respecto al BOE "macroalgas" se han estimado dos índices: el CFR (calidad de los fondos rocosos) y el RSL (lista reducida de especies). Las diferencias ecológicas entre las zonas costeras de la ecorregión Atlántico Norte donde se han desarrollado estos métodos de evaluación (norte de España y Reino Unido respectivamente) y la costa atlántica de Cádiz, sugirieron la necesidad de una adaptación de ambos índices para el sur de España. Los muestreos intensivos han permitido la elaboración de una lista reducida de especies para las costas rocosas sudoccidentales de España.

El presente trabajo muestra los esfuerzos y resultados preliminares de la evaluación de BQE basados en macrófitos marinos en el sur de España. Se discuten también algunos interrogantes sobre la necesidad de una reevaluación de las herramientas propuestas para la ecorregión Atlántico Norte cuando se aplican a las masas de agua de Andalucía.

**Palabras clave:** Andalucía, angiospermas marinas, Elementos de Calidad Biológica, macroalgas

## 1. THE ANDALUSIAN CONTEXT

In Spain, the management of coastal and transitional waters of the river basin districts has been transferred to different territories (administrative divisions called Autonomic Communities). Andalusia (Fig. 1) is the only territory in Spain where the coastal and transitional waters lay within two different Ecoregions: the (North) Atlantic Ocean (region 1) and the Mediterranean Sea (region 6). In addition, in the Strait of Gibraltar, which connects both Ecoregions, there is an important water exchange between the inflowing Atlantic waters and the outflowing Mediterranean waters (Farmer and Armi 1988). This exchange markedly affects the characteristics of the coastal waters and their algal communities. Andalusia also has the largest length of coast in Spain (almost 1000 km).

Therefore, unlike the rest of the administrative divisions of the country, to implement the European Water Framework Directive (WFD) double the effort is required to classify the ecological status of the water bodies. This implies more economic resources, more

personal and increased time consuming sampling and data analysis.

The goal of this article for the special issue celebrating 10 years since the publication of the European WFD is to present the state of the art of the results obtained with the biological quality elements (BQE) macroalgae and angiosperms in both coastal and transitional waters of Andalusia. Additionally, future prospects on the use of these biological elements to assess the ecological status of both surface waters in Andalusia are discussed.

## 2. MACROALGAE AND ANGIOSPERMS AS BQE FOR THE ATLANTIC ECOREGION

Macroalgae (Rhodophyta, Chlorophyta and Heterokontophyta) are an heterogeneous group of macroscopic photosynthetizers that are not properly a taxonomic group. In fact they are placed into Plantae and Chromista kingdoms in the tree of life (Brodie and Lewis 2007). Marine angiosperms are rooted flowering plants that are successfully adapted to grow in the marine environment. These two groups of photosynthetic organisms have been selected as BQE to estimate

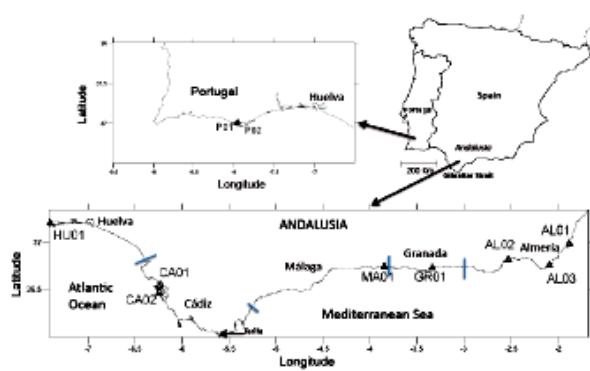
the ecological quality of coastal and transitional waters.

Regarding macroalgae, two main indices have been proposed to estimate the ecological status of coastal waters: the CFR (Quality of Rocky Bottoms; Juanes et al. 2008, Guinda et al. 2008) and the RSL (Reduced Species List; Wells et al. 2007). These two indexes have been tested on the coasts of Andalusia after a large field sampling programme carried out since 2006. The CFR index estimates the ecological status of the water mass taking into account the macroalgal species richness, the presence of opportunistic species and the cover of hard substrates. The RSL index has a more complex metric scoring system and estimates the species richness, the percentages of Chlorophyta, Rhodophyta and opportunistic algae, the ESG ratio (ESG I/ESG II) (Ecological Status Group; Orfanidis et al. 2001) and a shore description. The ESG concept (see also Orfanidis 2007) is based on the functional groups of Littler and Littler (1984) and assembles macroalgae within two groups: EGS I (late successional or perennials) and ESG II (opportunists or annual species).

The main difference between both methods is that RSL is based on species number and composition whereas CFR depends on the relative abundance of species. This becomes very important when results are intercalibrated, as the ecological sensitivity as well as the temporal and spatial scales of both indices, depend on this subtle differentiation in data treatment. For example, the cover of the opportunistic macroalgae *Ulva spp.* changes seasonally on many shores. That would be very significant for the CFR index, which considers variations in percentage cover but it might be irrelevant for the

SRL index, which only considers the presence or absence of the species.

In addition, the ecological differences between the coastal zones where these two methods were developed (Northern Spain; CFR and the United Kingdom; RSL) and the Atlantic coasts of the province of Cadiz (Fig. 1), suggest a need for reassessment of both indices within Andalusia. In order to compare each method, following Wells et al. (2007), the subjective ecological quality status of shores was preliminary classified by expert judgment as "high", "good" and "moderate" (no shores were classified subjectively as "poor" or "bad"). The calculation of the scores for the CFR index as well as the characteristics of the shores and algal communities along the Andalusian coast suggested that the specific richness was the only significant factor to discriminate among sites and therefore this index may not be the best choice to be recommended. Nonetheless, the limits given by Guinda (2008) were recalculated according to Wells et al. (2008) if the CFR index is used in the southern Atlantic coasts (table I). The CFR has been also estimated in sublittoral zones near Tarifa Island, Cádiz (Hernández et al. 2010). The ecological status classification of the body of water according to all sites was "high", which coincided with the experts perception. Therefore, this index seems to be suitable to assess the ecological status in subtidal rocky substrates.



*Figure 1: Map of sampling sites of macroalgae and marine angiosperms in southern Iberian Peninsula. Provinces of Andalusia are separated by bars. P01 and P02: Ria Formosa; HU01: Huelva (Guadiana river mouth); CA01 and CA02: Cádiz (Río San Pedro and Guadalete river estuary in Cadiz Bay Natural Park); MA01: Maro (Málaga); GR01: Granada (Cambriles); AL01, AL02 and AL03: Almeria (Carboneras, El Palmer y San José).*

The RSL index was also adjusted to the particularities of the southern Atlantic coast of Andalusia (Bermejo 2009; Bermejo et al. in prep.). This reassessment involved adjusting the number of species included on the reduced species list and recalculating boundaries between classes, following the recommendations given by Wells et al. (2007). Three main results were derived from intense field sampling along the Atlantic coast of Cadiz: a reduced species list (table II), which is composed of 10 chlorophytes, 15 phaeophyceans and 39 rhodophytes, a relationship between the species richness (SR) and the score of the shore description (SD) ( $SR = e^{(5,14 - 23,9 \cdot SD)}; R^2 = 0,77$ ) and the existence of two ecological regions defined

according to the macroalgal intertidal flora via hierarchical-cluster analysis (Bermejo 2009):

Atlantic Gibraltar Strait and Western Atlantic zone. Interestingly, these regions coincided with the coastal boundaries between the Algarve and Gadiano littoral districts of the biogeographical classification of Andalusia (Rivas-Martínez 1997). Overall, the SRL index, as adapted for the southern Atlantic coasts of Spain, demonstrated a better capacity to detect differences in the ecological status of the Atlantic coastal waters than the CFR.

*Table 1. Mean (X), standard deviation (S), quality thresholds and proposed score system with classification status ranges for the CFR index according to the field data from the Andalusian Atlantic coast (Bermejo 2009).*

Element	Status	X	S	Range	CFR
Coverage (%) of characteristic macroalgae populations	Moderate	62.5	33.04	55 - 75	20
	Good	74.9	18.29	75 - 80	35
	Very good	88.0	16.43	>80	45
Relative coverage (%) of opportunistic species	Moderate	22.2	19.36	40 - 20	35
	Good	9.5	6.43	20 - 10	25
	Very good	6.8	4.02	<10	15
Richness of characteristic macroalgae populations	Moderate	5.75	0.96	5 - 6	20
	Good	6.8	1.32	7 - 8	15
	Very good	8.2	1.30	>8	10

Table II. Reduced species list for the Atlantic coasts of Spain. The ecological strategy of algae is classified based on Grime (1977). T: Stress tolerator; R: ruderal, C: competitor. \* Species considered as opportunistic.

Species	Estrategy	ESG	Reference
<i>Bryopsis spp.*</i>	T	II	Orfanidis <i>et al.</i> 2003
<i>Chaetomorpha linum</i> (O.F. Müller) Kützing*	T	II	Orfanidis <i>et al.</i> 2003
<i>Cladophora spp.*</i>	R	II	Orfanidis <i>et al.</i> 2003
<i>Codium spp</i> like-tree	T	II	Orfanidis <i>et al.</i> 2003
<i>Codium spp</i> encrusting	T	II	Bermejo 2009
<i>C. bursa</i> (Olivi) C. Agardh	T	II	Orfanidis <i>et al.</i> 2003
<i>Flabellia petiolata</i> (Turra) Nizamuddin	C	I	Orfanidis <i>et al.</i> 2003
<i>Pedobesia simplex</i> (Meneghini ex Kützing) M.J. Wynne & Leliaert	T	II	Hernández <i>et al.</i> 2009
<i>Ulva spp.*</i>	R	II	Orfanidis <i>et al.</i> 2003
<i>Valonia utricularis</i> (Roth) C. Agardh	T	II	Orfanidis <i>et al.</i> 2003
<i>Cladostephus spongiosus</i> (Hudson) C. Agardh	T	II	Orlando-Bonaca <i>et al.</i> 2008
<i>Colpomenia sinuosa</i> (Mertens ex Roth) Derbès & Solier	R	II	Panayotidis <i>et al.</i> 2004
<i>Cystoseira compressa</i> (Esper) Gerloff & Nizamuddin	T	I	Orfanidis <i>et al.</i> 2003
<i>C. mauritanica</i> Sauvageau	C	I	Orfanidis <i>et al.</i> 2003
<i>C. tamariscifolia</i> (Hudson) Papenfuss	C	I	Orlando-Bonaca <i>et al.</i> 2008
<i>C. usneoides</i> (Linnaeus) M. Roberts	T	I	Orlando-Bonaca <i>et al.</i> 2008
<i>Dictyota dichotoma</i> (Hudson) J.V. Lamouroux	R	II	Orfanidis <i>et al.</i> 2003
<i>Dictyopteris polypodioides</i> (A.P. De Candolle) J.V. Lamouroux	R	II	Orfanidis <i>et al.</i> 2003
<i>Fucus spiralis</i> Linnaeus	C	I	Guinda <i>et al.</i> 2008
<i>Halopteris filicina</i> (Grateloup) Kützing	T	II	Orfanidis <i>et al.</i> 2003
<i>Laminaria ochroleuca</i> Bachelot de la Pylaie	C	I	Guinda <i>et al.</i> 2008
<i>Padina pavonica</i> (Linnaeus) Thivy in W.R. Taylor	T	I	Orfanidis <i>et al.</i> 2003
<i>Saccorhiza polyschides</i> (Lightfoot) Batters	C	I	Guinda <i>et al.</i> 2008
<i>Sargassum vulgare</i> C. Agardh	C	I	Orfanidis <i>et al.</i> 2003
<i>Stylocaulon scoparium</i> (Linnaeus) Kützing	T	II	Hernández <i>et al.</i> 2009
<i>Zanardinia typus</i> (Nardo) P.C. Silva in W. Greuter	T	I	Orlando-Bonaca <i>et al.</i> 2008
Delesseriaceae ( <i>Acrosorium</i> , <i>Cryptopleura</i> , <i>Haraldiphyllum spp.</i> )	C	I	Guinda <i>et al.</i> 2008
<i>Asparagopsis armata</i> Harvey	T	II	Hernández <i>et al.</i> 2009
<i>Bonnemaisonia asparagoides</i> (Woodward) C. Agardh	T	II	Bermejo 2009
<i>Botryocladia botryoides</i> (Wulfen) Feldmann	C	I	Hernández <i>et al.</i> 2009
<i>Caulacanthus ustulatus</i> (Mertens ex Turner) Kützing	T	I	Hernández <i>et al.</i> 2009
<i>Ceramium spp.</i>	R	II	Orfanidis <i>et al.</i> 2003
<i>Chondracanthus acicularis</i> (Roth) Fredericq	R	I	Hernández <i>et al.</i> 2009
<i>Corallina spp.</i>	R	I	Orfanidis <i>et al.</i> 2003
<i>Cryptonemia seminervis</i> (C. Agardh) J. Agardh	C	I	Bermejo 2009
<i>Gastroclonium ovatum</i> (Hudson) Papenfuss	T	II	Guinda <i>et al.</i> 2008
<i>G. clavatum</i> (Roth) Ardissono	T	II	Guinda <i>et al.</i> 2008

Tabla II (cont.).

Species	Estrategy	ESG	Reference
<i>Gelidium spinulosum</i> (C. Agardh) J. Agardh	C	I	Bermejo 2009
<i>G. spinosum</i> (S.G. Gmelin) P.C. Silva in Silva, Basson & Moe	C	I	Hernández <i>et al.</i> 2009
<i>G. corneum</i> (Hudson) J.V. Lamouroux	C	I	Bermejo 2009
<i>G. pusillum</i> (Stackhouse) Le Jolis	T	II	Orfanidis <i>et al.</i> 2003
<i>Gigartina pistillata</i> (S.G. Gmelin) Stackhouse	C	II	Orlando-Bonaca <i>et al.</i> 2008
<i>Gymnogongrus/Ahnfeltiopsis</i> sp.	C	I	Orlando-Bonaca <i>et al.</i> 2008
<i>Halopithys incurva</i> (Hudson) Batters	T	I	Orlando-Bonaca <i>et al.</i> 2008
<i>Halurus equisetifolius</i> (Lightfoot) Kützing	T	I	Juanes <i>et al.</i> 2008
<i>Hildenbrandia rubra</i> (Sommerfelt) Meneghini	T	I	Hernández <i>et al.</i> 2009
<i>Hypoglossum</i> & <i>Apoglossum</i> spp	C	I	Guinda <i>et al.</i> 2008
<i>Jania rubens</i> (Linnaeus) J.V. Lamouroux	T	I	Orfanidis <i>et al.</i> 2003
<i>Laurencia obtusa</i> (Hudson) J.V. Lamouroux	T	II	Orfanidis <i>et al.</i> 2003
<i>Lithophyllum byssoides</i> (Lamarck) Foslie	C	I	Guinda <i>et al.</i> 2008
<i>L. dentatum</i> (Kützing) Foslie	T	I	Orlando-Bonaca <i>et al.</i> 2008
<i>L. incrustans</i> Philippi	T	I	Orlando-Bonaca <i>et al.</i> 2008
<i>Nemalion helminthoides</i> (Velley) Batters	T	I	Juanes <i>et al.</i> 2008
<i>Lomentaria articulata</i> (Hudson) Lyngbye	T	II	Hernández <i>et al.</i> 2009
<i>L. catenata</i> Harvey	C	II	Guinda <i>et al.</i> 2008
<i>Osmundea pinnatifida</i> (Hudson) Stackhouse	C	I	Hernández <i>et al.</i> 2009
<i>O. hybrida</i> (A.P. de Candolle) K.W. Nam	T	II	Orfanidis <i>et al.</i> 2003
<i>Peyssonnelia</i> spp.	C	I	Orfanidis <i>et al.</i> 2003
<i>Plocamium cartilagineum</i> (Linnaeus) P.S. Dixon	T	II	Hernández <i>et al.</i> 2009
<i>Pterocladiella capillacea</i> (S.G. Gmelin) Santelices & Hommersand	R	II	Orlando-Bonaca <i>et al.</i> 2008
<i>Pterosiphonia complanata</i> (Clemente) Falkenberg	C	II	Hernández <i>et al.</i> 2009
<i>Rhodymenia</i> & <i>Schottera</i> spp.	T	I	Orlando-Bonaca <i>et al.</i> 2008
<i>Sphaerococcus coronopifolius</i> Stackhouse	C	II	Guinda <i>et al.</i> 2008

Regarding angiosperms, we are developing an index for marine angiosperms (*Zostera marina*, *Zostera noltii* and *Cymodocea nodosa*) based on the multivariate index proposed for *Posidonia oceanica* (Romero *et al.* 2007). The index is called SISIP (Seagrass Indicator for South Iberian Peninsula) and it is being tested in Cadiz Bay and Ria Formosa (Portugal). This index follows recommendations given by other authors (Romero *et al.* 2007, Fernandez-Torquemada *et al.* 2008) and includes not only the estimation of

shoot density, percentage cover and biomass in quadrats but also plant tissue variables (non structural carbohydrates, tissue N, C, P, tissue stable isotope ratios) and variables at the individual, population and community level (biometry, foliar necrosis, rhizome burial, etc). Preliminary data for *Z. noltii* has classified the Río San Pedro, Cádiz (an arm of the sea or semi enclosed tidal inlet) as "good" (García-Marín *et al.* 2009). However, further research is still necessary to establish a more accurate and integrated ecological quality ratio considering other water bodies within the same biogeographical region.

We are sampling meadows of *Cymodocea nodosa* in several sites along the Atlantic coast so that future intercalibration with results obtained by the Portuguese research groups may be possible.

There have been relatively few attempts to determine the biological quality of Atlantic transitional waters using macroalgae as BQEs (e.g. Borja et al. 2004). There are methods proposed in the UK and Portugal based on opportunistic macroalgal blooms (Scanlan et al. 2007, Patrício et al. 2007, WFD-UKTAG 2009) and above all, the extent of upstream penetration of perennial fucoid algae (Wilkinson et al. 2007). The methods based on opportunistic algae may be difficult to apply, as coverage or algal density of opportunistic Ulvaceans is highly variable both spatially and seasonally among estuaries (Hernández et al. 1997). In addition, in some cases, opportunistic algal cover responds to environmental variables not necessarily related to pollution, as water clarity and salinity (Krause-Jensen et al. 2007). These methods could be used during peak bloom periods but still needs an assessment in Andalusian transitional waters. The method based on fucoids cannot be applied in the Spanish south Atlantic estuaries as there are few records of fucoids in transitional waters.

The ecological status of transitional waters in Andalusian estuaries can be evaluated with the angiosperm quality index (AQI) developed by García et al. (2009). A previous version of this multi-metric method was applied in the Guadalete estuary (García de Lomas et al. 2007). Based on the composition, abundance, coverage and loss of spatial extent of the estuarine habitat, the assessed section of this estuary was classified as "moderate". In addition, the SISIP index can be tested in meadows of *Cymodocea nodosa* thriving in some Iberian Atlantic transitional waters (e.g.

Guadiana estuary) where we have samples that still have to be analysed.

### 3. MACROALGAE AND ANGIOSPERMS AS BQEs FOR THE MEDITERRANEAN ECOREGION

As for the Atlantic coasts, the ecological status of the different bodies of Mediterranean coastal waters in Andalusia has to be assessed using macroalgae and angiosperms as BQEs. It is expected to use macroalgae to apply the methods proposed in other Spanish Mediterranean territories with the already available CARLIT-BENTHOS index (Ballesteros et al. 2007, Pinedo et al. 2007), although possibly some adjustments for the Andalusian Mediterranean coast may be needed. Alternatively, the RSL index should be developed for the Mediterranean.

Regarding seagrasses, an extensive field campaign along the Mediterranean coastal waters from Málaga to Almería has been carried out (Fig. 1, Table III). As a result, the environmental status of 8 sampling points was established by expert judgement, considering the anthropogenic pressures in each zone. Data from the meadows have been collected in order to implement the POMI (*Posidonia Oceanica* Multivariate Index; Romero et al. 2007) and SISIP multifactorial indexes. We are at present processing all this information.

*Table 3. Seagrass species and environmental status in selected sampling stations surveyed along the Mediterranean coast of Andalusia. AL: Almería, GR: Granada, MA: Málaga.*

Sampling site	Species	Environmental status
AL - Carboneras	<i>P. oceanica</i>	Healthy
AL - El Palmer	<i>P. oceanica</i>	Unhealthy
AL - El Palmer	<i>C. nodosa</i>	Unhealthy
AL - El Palmer	<i>Z. noltii</i>	Unhealthy
AL - San José	<i>P. oceanica</i>	Healthy
AL - San José	<i>C. nodosa</i>	Healthy
GR - Cambriles	<i>P. oceanica</i>	Intermediate
MA - Maro	<i>P. oceanica</i>	Healthy

There are seven water bodies classified as transitional waters on the Andalusian coast. However no information is available either for macroalgae or seagrasses. The SISIP index could be tested in the scarce meadows of *Zostera noltii* growing in some estuaries, as in Palmones river estuary (Peralta et al. 2000), and perhaps metrics based on the growth of opportunistic green algae could be tested, but again with the same limitations imposed for the Atlantic waters. Recently, a macrophyte quality index (MAQI) has been proposed to assess the ecological status of Italian marine transitional environments (Sfriso et al. 2009). The dichotomical key for the assessment of the ecological status developed in this method may be also applied in the Andalusian bodies of transitional Mediterranean waters.

#### 4. FUTURE PROSPECTS

As it can be deduced from the information described above, despite the vast extension of the Andalusian coastal and transitional waters, the

information available for macroalgae and seagrasses as BOEs is still rather scarce compared with other Spanish Autonomic Communities. Investment in field data acquisition and monitoring is therefore needed, especially for the Mediterranean. There is, however, information for both the CFR and SRL indexes in the Atlantic. Each one has their own strengths and limitations (Hernández 2008) but data suggest that the RSL is more convenient to assess the ecological status of the rocky shores in Andalusia. Both the AQI (atlantic transitional waters) and the SISIP (coastal and transitional waters) indexes are promising and should be developed and intercalibrated with data from other water bodies. We are at present in contact with groups developing and/or assessing methods for macroalgae in Spain and Portugal (PmarMAT, CYMOX; Marta Pérez com. per., POSIMED network) and the Andalusian environmental management company EGMASA, which develops specific environmental programs for seagrass monitoring. The perspectives are therefore that reliable information for BOEs will be available within the next couple of years.

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