European Commission

Technical Guidelines for assessing and monitoring the condition of Annex I habitat types of the Directive 92/43/EEC

Posidonia beds (*Posidonion oceanicae*) (1120*)

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Technical Guidelines for assessing and monitoring the condition of Annex I habitat types of the Directive 92/32/EEC

Posidonia beds *(Posidonion oceanicae)* (1120*)

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Contents

Acknowledgements 1
Glossary and definitions 2
Abbreviations 3
Executive summary 4
1 Definition and ecological characterisation 6
1.1 Definition and interpretation of habitats covered 6
1.2 Environmental and ecological characterization and selection of variables to measure habitat condition 7
1.3 Selection of typical species for condition assessment 13
2 Analysis of existing methodologies for the assessment and monitoring of habitat condition ...15
2.1 Variables used, metrics and measurement methods, existing data sources 15
2.2 Definition of ranges and thresholds to obtain condition indicators 24
2.3 Aggregation methods at the local scale 27
2.4 Aggregation at biogeographical scale 28
2.5 Selection of localities 28
2.6 General monitoring and sampling methods 29
2.7 Other relevant methodologies 31
2.8 Conclusions 32
3 Guidance for the harmonisation of methodologies for assessment and monitoring of habitat condition 34
3.1 Selection of condition variables, metrics and measurement methods 34
3.2 Guidelines for the establishment of reference and threshold values, and obtaining condition indicators for the variables measured 41
3.3 Guidelines for the aggregation of variables at the local level 46
3.4 Guidelines for aggregation at the biogeographical region scale 49
3.5 Guidelines on general sampling methods and protocols 49
3.6 Selecting monitoring localities and sampling design 53
3.7 Use of available data sources, open data bases, new technologies and modelling 56
4 Guidelines to assess fragmentation at appropriate scales 57
5 Next steps to address future needs 59
6 References 60

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Glossary and definitions

Habitats

Natural habitats: are terrestrial or aquatic areas distinguished by geographic, abiotic and biotic features, whether entirely natural or semi-natural (Habitats Directive).

Habitat condition: is the quality of a natural or semi natural habitat in terms of its abiotic and biotic characteristics. Condition is assessed with respect to the habitat composition, structure and function. In the framework of conservation status assessment, condition corresponds to the parameter "structure and function". The condition of a habitat asset is interpreted as the ensemble of multiple relevant characteristics, which are measured by sets of variables and indicators that in turn are used to compile the assessments.

Habitat characteristics: are the attributes of the habitat and its major abiotic and biotic components, including structure, processes, and functionality. They can be classified as abiotic (physical, chemical), biotic (compositional structural, functional) and landscape characteristics (based on the Ecosystems Condition Typology defined in the SEEA-EA; United Nations et al., 2021).

Species

Characteristic species: are species that characterise the habitat type, are used to define the habitat, and can include dominant and accompanying species.

Typical species: are species that indicate good condition of the habitat type concerned. Their conservation status is evaluated under the structure and function parameter. Usually, typical species are selected as indicators of good condition and provide complementary information to that provided by other variables that are used to measure compositional, structural and functional characteristics.

Variables

Condition variables: are quantitative metrics describing individual characteristics of a habitat asset. They are related to key characteristics of the habitat that can be measured, must have clear and unambiguous definition, measurement instructions and well-defined measurement units that indicate the quantity or quality they measure. In these guidelines, the following types of condition variables are included:

- **Essential variables:** describe essential characteristics of the habitat that reflect the habitat quality or condition. These variables are selected on the basis of their relevance, validity and reliability and should be assessed in all MSs following equivalent measurement procedures.
- **Recommended variables:** are optional, additional condition variables that may be measured when relevant and possible to gain further insight into the habitat condition, e.g. according to contextual factors; these are complementary to the essential variables, can improve the assessment and help understand or interpret the overall results.
- **Specific variables:** are condition variables that should be measured in some specific habitat types or habitat sub-groups; can thus be considered essential for those habitats, which need to be specified (e.g. salinity for saline grasslands, groundwater level for bog woodlands, etc.).

Descriptive or contextual variables: define environmental characteristics (e.g. climate, topography, lithology) that relate to the ecological requirements of the habitat, are useful to characterise the habitat in a specific location, for defining the relevant thresholds for the condition variables and for interpreting the results of the assessment. These variables, however, are not included in the aggregation of the measured variables to determine the condition of the habitat.

Reference levels and thresholds: are defined for the values of the variables (or ranges) that determine whether the habitat is in good condition or not. They are set considering the distance from the reference condition (good). The value of the reference level is used to re-scale a variable to derive an individual condition indicator.

Condition indicators: are rescaled versions of condition variables. Usually, they are rescaled between a lower level that corresponds to high habitat degradation and an upper level that corresponds to the state of a reference habitat in good condition.

Aggregation: is defined in this document as a rule to integrate and summarise the information obtained from the measured variables at different spatial scales, primarily at the local scale (sampling plot, monitoring station or site).

Abbreviations

EU: European Union

HD: Habitats Directive

IAS: Invasive Alien Species

MS: Member State

EU Member States acronyms:

Austria	(AT)	Estonia	(EE)	Italy	(IT)	Portugal	(PT)
Belgium	(BE)	Finland	(FI)	Latvia	(LV)	Romania	(RO)
Bulgaria	(BG)	France	(FR)	Lithuania	(LT)	Slovakia	(SK)
Croatia	(HR)	Germany	(DE)	Luxembourg	(LU)	Slovenia	(SI)
Cyprus	(CY)	Greece	(EL)	Malta	(MT)	Spain	(ES)
Czechia	(CZ)	Hungary	(HU)	Netherlands	(NL)	Sweden	(SE)
Denmark	(DK)	Ireland	(IE)	Poland	(PL)		

MSFD: Marine Strategy Framework Directive

SEEA EA: System of Environmental Economic Accounting- Ecosystem Accounting

WFD: Water Framework Directive

Executive summary

Posidonia beds (habitat type 1120*) are included in Annex I of the EU Habitats Directive as a priority habitat. *Posidonia oceanica is* a seagrass endemic to the Mediterranean Sea that forms extensive underwater meadows from shallow waters up to 30-40m deep. It is present in eight EU Member States (Croatia, Cyprus, France, Greece, Italy, Malta, Slovenia and Spain) playing a crucial ecological role in the Mediterranean Sea. These guidelines aim to facilitate consistent condition assessment for habitat 1120* across EU Member States, to support Article 17 reporting under the Habitats Directive, and contribute to setting conservation objectives for *Posidonia* beds.

This document presents a structured approach to support a harmonised assessment and monitoring of *Posidonia* beds across Member States, addressing the following three main sections:

1) A definition and ecological characterisation of the habitat identify the **key ecological characteristics** and the corresponding **variables** related to the habitat condition**,** under the six classes (physical, chemical, compositional, structural, functional and landscape/seascape), defined in the Ecosystems Condition Typology of the United Nations' System of Environmental Economic Accounting - Ecosystem Accounting (SEEA-EA).

2) A review of **existing methodologies** for assessing and monitoring the habitat from six Member States (Croatia, France, Italy, Malta, Slovenia and Spain) was carried out to identify best practices and gaps in the use of variables and indicators, survey methods and monitoring protocols. The analysis reveals some differences but also some interesting commonalities that deserve particular attention.

Almost all the Member States (5/6) monitor abiotic variables including physical and chemical parameters to assess the condition of the water and the sediments in the areas where *Posidonia* beds are found. All the MSs monitor some biotic characteristics of the habitat with different level of detail.

The associated fauna, including characteristic and indicator species, fish species, but also invasive species, are included among the main biotic variables. Structural variables include the percentage of cover of living rhizome, and the density of shoots. The latter is monitored by all the MSs, being a variable that is easy to measure and is useful to detect changes in the habitat condition due to changing environmental conditions or the presence of pressures or impacts. Most of the biotic characteristics analysed are monitored through visual observation and in situ measurements (diving). Functional characteristics of the habitat are mainly assessed by measuring shoot and rhizome growth and mortality, flowering events and the effects of pressures and disturbances. Three MSs address seascape characteristics, including the area covered by the habitat and the degree of fragmentation, in order to assess the evolution and potential regression and loss of the meadow. The assessment and monitoring of these characteristics involve remote sensing or, in the case of small areas/patches, in situ evaluation.

The use of reference values and thresholds for determining good condition vary significantly among the MSs: some MSs have well-defined and specific thresholds for the different variables; others use existing indices under other European Directives (the Water Framework Directive and the Marine Strategy Framework Directive) to assess the ecological status of the habitat; and some rely mostly on the expert judgement for this assessment.

3) Guidelines for harmonising the assessment and monitoring of the habitat condition include a proposal for a set of **essential and recommended variables,** together with their **measurement procedures** and **general monitoring and sampling protocols**. In this section, emphasis is placed on the selection of variables ecologically meaningful, cost-effective, and feasible for long-term monitoring. The variables that are most frequently used in the existing national methodologies are considered to promote this harmonisation. Among the physical variables parameters as water temperature and transparency/turbidity are proposed as essential. Among the chemical variables, the total organic carbon, nutrient concentration (phosphates, nitrates), pH and salinity are considered essential parameters to measure in the water. Among the biotic variables, the presence of certain species (other seagrass species, endangered species as *Pinna nobilis* and some indicator and characteristic species) as well as non-indigenous species as *Caulerpa* spp., are considered essential to assess the compositional characteristics. The percentage covered by living rhizomes, the shoot density, the characterisation and monitoring of the lower limit of the meadow, the size of patches (specifically in patchy meadows) and the burial degree of the rhizomes are proposed to be measured as essential structural variables. The evaluation of the presence and effects caused by different pressures and impacts are proposed as an essential variable to assess the functional characteristics of the meadow. Finally, the area covered by the meadow and the evaluation of its fragmentation are considered essential parameters to assess the condition the habitat under seascape characteristics.

These guidelines also define the main criteria and provide recommendations on the establishment of reference values and thresholds that would help determining whether the habitat is in good condition or not. Additionally, recommendations for the aggregation of results at various scales and for the selection and the number of monitoring localities needed to ensure the representativity of the results obtained at the biogeographical scale are also discussed.

1 Definition and ecological characterisation

1.1 Definition and interpretation of habitats covered

The Interpretation Manual (European Commission, 2013) gives the following definition for *Posidonia* beds (*Posidonion oceanicae*) (habitat type 1120*):

"Beds of *Posidonia oceanica* (Linnaeus) Delile are characteristic of the infralittoral zone of the Mediterranean (depth: ranging from a few dozen centimetres to 30-40 metres). On hard or soft substrate, these beds constitute one of the main climax communities. They can withstand relatively large variations in temperature and water movement, but are sensitive to desalination, generally requiring a salinity of between 36 and 39 parts per 1000."

This habitat, included in the Annex I of the Habitats Directive as a priority habitat, is characterised by the presence of Neptune grass (*Posidonia oceanica*), a seagrass species that is endemic to the Mediterranean Sea. It forms extensive underwater meadows that can be found growing either between **rocky substrates** or in **sandy areas** (Pagès et al., 2014), providing crucial services and supporting diverse and complex ecological communities (Campagne et al., 2015). Thus, *Posidonia oceanica* beds are the habitat of a great variety of species that find shelter and food in this underwater forest playing a **fundamental role as a nursery** (Francour, 1997).

Structurally, *P. oceanica* meadows are characterised by two distinctive strata: a **higher stratum, formed by a dense leaf canopy** that can exceed 1,000 bundles/m^2 in shallow areas (Díaz & Marbá, 2009) and reach up to 1 m in length during summer; and a **lower stratum, formed by a thick structure of rhizome, roots and detritus** called "matte" (Boudouresque & Jeudy de Grissac, 1983) or mat, that is buried in the sediment, growing vertically throughout the years and slowly building a reef. This buried part of the meadow promotes sediment stability (Gacia & Duarte, 2001) and is a major carbon sink, sequestrating carbon dioxide (CO_2) and storing it in the sediment. This is mainly why *P. oceanica* is considered a key species for mitigating the effects of climate change (Pergent-Martini et al., 2021), as well as being an excellent oxygen (O_2) producer and having the ability to protect the coast from erosion by slowing down hydrodynamics (Pergent et al., 2016) with its long leaves.

Neptune grass meadows lose their older leaves in autumn (Ros et al., 1985), which accumulate on the beaches due to the action of waves and storms. These leaf accumulations, known as *banquettes*, cover the shoreline and are a typical sight on the Mediterranean coast in autumn. *Banquettes* also play an important role in buffering the impact of waves, preventing coastal erosion (Mateo et al., 2002) and the consequent loss of sand from the beaches.

In marine systems, seagrasses are important structural habitats in sandy bottoms (Hemminga & Duarte, 2000). Specifically, *P. oceanica* meadows can have different configurations, with large continuous meadows, patchy meadows in sedimentary bottoms and patchy meadows in rocky bottoms being three of the most frequent (Ricart et al., 2015). This heterogeneity plays a major role in the functioning of seagrass meadows and their associated fauna (Boström et al., 2006).

The meadow morphology of Neptune grass depends on its **degree of fragmentation**, which can occur due to natural processes (storms, hydrodynamic action) or human activities (Boudouresque et al., 2009). Anthropogenic impacts such as **nutrient and sediment pollution** in coastal waters (Colomer et al., 2017), coastal constructions (Ruiz & Romero,

2003) or boat mooring and anchoring (Francour et al., 1999) may cause extensive fragmentation and a general regression of *P. oceanica* beds. This regression has been estimated to be around 34% in the last 50 years (Telesca et al., 2015) and is a generalised phenomenon in the Mediterranean Sea.

'*Posidonia* beds' (*Posidonia oceanica*) (code 1120*) is listed in the Habitats Directive under Coastal and Halophytic habitats and in the subcategory 'Open Sea and Tidal Areas'. This habitat may be found in association with 'Sandbanks which are slightly covered by sea water all the time' (code 1110). We do not suggest clustering both habitats because, even though the underlying sediment needs to be examined for both, there is a much stronger emphasis on biological monitoring, particularly with regard to the condition, extent and associated species for this habitat (1120*).

Distribution across the regions

'*Posidonia* beds (*Posidonion oceanicae*)' are present in the following Member States: Croatia (HR), Cyprus (CY), France (FR), Greece (GR), Italy (IT), Malta (MT), Slovenia (SI) and Spain (ES).

Posidonia oceanica meadows have been estimated to cover about 1.5% of the total Mediterranean Sea surface (Pasqualini et al., 1998), representing an area of 8,014.51 km^2 as estimated in the latest Art. 17 reporting period (2013-2018). Local abundance is influenced by the presence of suitable substrates and by water conditions, not being able to survive in areas with low salinity, turbidity, high levels of organic matter (Telesca et al., 2015) or pollutants. *P. oceanica* is therefore considered a good bioindicator for assessing water quality and is used as an indicator in the Water Framework Directive (Romero et al., 2007), and a good environmental descriptor (Pergent-Martini et al., 2005).

Distribution-wise (Vacchi et al., 2017), Neptune grass meadows are less abundant in the Levantine Sea and very limited in the Marmara Sea (Boudouresque et al., 2012). *P. oceanica* beds are widespread in the western Mediterranean basin, but rare or absent in the northern Adriatic Sea (Gamulin-Brida, 1974) and along the Languedoc coast between the Rhone delta and Port la Nouvelle (Boudouresque & Meinesz, 1982). The westernmost distribution limit of *P. oceanica* is considered to be in Punta Chullera (Málaga, Spain) in the Alboran Sea, as meadows start to become more scattered and isolated moving west from Cabo de Gata (Almería, Spain) as a result of the influence of the colder, more turbid and nutrient-rich Atlantic waters (Ruiz et al., 2015).

1.2 Environmental and ecological characterization and selection of variables to measure habitat condition

Posidonia beds (*Posidonion oceanicae*) are strongly influenced by different physical and chemical factors that determine their distribution. *Posidonia oceanica* is very sensitive to poor water conditions, low salinity (Ruiz et al., 2009), settling on the right substrate (Pérez et al., 2015) and, as a vegetal species, light irradiation, which is directly related to depth and turbidity (Pérez et al., 2015). Consequently, the habitat '*Posidonia* beds' is restricted to areas where this seagrass has the capacity to survive and form a meadow. The slow growth rate (Hemminga & Duarte, 2000) makes it more difficult for meadows to survive an impact, and it is therefore urgent to prevent further pressures and to stop the generalised regression of this habitat.

The main characteristics that determine the structure and function of *Posidonia* beds are:

- **Depth**: this determines the distribution of *Posidonia oceanica*, as it affects light availability, essential for the plant to grow (Pérez et al., 2015). Greater depth makes it more difficult for the plant to grow.
- **Turbidity**: as with depth, turbidity also determines the amount of light that can reach the sea bottom, thus conditioning its growth (Pérez et al., 2015). Greater turbidity makes it more difficult for the plant to grow.
- **Nutrient concentration**: this refers to the amount of organic matter present in the water and is directly linked to its quality (Telesca et al., 2015) and turbidity, affecting the growth and distribution of the plant. *P. oceanica* requires water with a low concentration of nutrients (Díaz & Marbà, 2009).
- **Substrate type**: the substrate type affects the growth, morphology and distribution of *P. oceanica* meadows. For example, the availability of rocky substrate affects the vertical distribution of *Posidonia* beds (Pérez et al., 2015) and the type of substrate influences the root development of the seedlings (Guerrero-Meseguer et al., 2017).
- **Hydrodynamism**: the water movement plays a role in the vertical distribution of this seagrass species (Pérez et al., 2015).
- **Sedimentation/erosion rate**: the slow growth rate makes it difficult for *Posidonia* beds to survive in areas with intense sedimentation (Pérez et al., 2015).
- **Water quality**: poor water quality affects the growth and survival of *Posidonia* beds, being very sensitive to environmental changes. For this reason, *P. oceanica* is used as a bioindicator species for water quality (e.g., Romero et al., 2007).
- **Sediment quality:** *P. oceanica* requires stable sediment with a low concentration of organic matter to grow and survive (Díaz & Marbà, 2009).
- **Associated species**: *Posidonia* beds provide food and refuge to a great number of species. There are multiple ecological links between this complex habitat and the species that inhabit it (e.g. herbivory pressure affecting the biomass of epiphytic species (Alcoverro et al., 1997) or root-fungus symbiosis that allows the plant to take up nutrients through roots (Borovec & Vohník, 2018)). In addition to naturally associated species, invasive fast-growing algal species affect the distribution and recovery of *P. oceanica,* owing to their great capacity to colonise dead areas of the meadow (Díaz & Marbà, 2009).

Examples of variables used for measuring these characteristics when reporting on the condition of habitat type 1120* are summarised in Table 1. The classification of the ecological characteristics and associated variables follows the UN-SEEA Ecosystem Condition Typology (ECT), which consists of six classes: abiotic physical, abiotic chemical, biotic compositional, biotic structural, biotic functional and landscape/seascape characteristics (United Nations et al., 2021). A summary description of these main characteristics is presented below.

1.2.1 Abiotic characteristics

Abiotic physical characteristics

- **Water transparency**: light availability is a limiting factor for the growth and distribution of *Posidonia* beds. Turbidity plays a major role in attenuating light and is therefore an important factor to evaluate.
- **Substrate type**: the type of substrate modifies the morphology of the meadow and its distribution and is therefore a factor that should be considered when evaluating the

results for different variables, such as shoot density, which is higher in hard bottoms (Mutlu et al., 2023).
- **Water temperature**: *Posidonia oceanica* meadows are highly vulnerable to warming, with climate change being an important threat to the habitat (Marbà & Duarte, 2010).

Abiotic chemical characteristics

- **Water quality**: *P. oceanica* is very sensitive to environmental conditions, requiring good water quality with low nutrient concentration to grow.
- **Sediment quality**: the nutrient concentration in the sediment is a crucial factor for the growth and survival of *P. oceanica*, requiring a low concentration. Also, the stability of the sediment is very important for the settlement of meadows.

1.2.2 Biotic characteristics

Biotic compositional characteristics

- **Presence of characteristic species**: a high diversity of species can be found in *Posidonia* beds. One of the most characteristic species is the noble pen shell (*Pinna nobilis*) which is also endemic in the Mediterranean Sea. Different monitoring protocols for this habitat included the study of abundance and density of *P. nobilis* populations (Díaz & Marbà, 2009). Now that this species is critically endangered, it is important to continue looking for surviving individuals when studying *P. oceanica* meadows.
- **Sea urchin abundance**: density fluctuations in the population of species such as *Paracentrotus lividus* can become an issue as they can cause massive defoliation in the meadow, especially when leaves and epiphytes are enriched with N and P because of anthropogenic pollution (Ruiz et al., 2001).
- ***Holothuria* spp. abundance**: holothurians are a good indicator species for evaluating the habitat quality (Díaz & Marbà, 2009), being sensitive to increases in salinity levels (Gacia et al., 2007) and chemical pollution (Harmelin et al., 1981). As detritivores, they feed on the organic matter present in the sediment, and their abundance is limited in oligotrophic environments (Roberts et al., 2000) but also in highly eutrophic sediment.
- **Associated fish**: there is a high diversity of fish species in this habitat (Templado et al., 2004), which is important for assessing habitat condition. Also, *Sarpa salpa* species plays an important role as an herbivore in the meadow (Alcoverro et al., 1997), as it is a good indicator of herbivory pressure.
- **Presence of invasive algal species**: a warmer environment facilitates the establishment of invasive, fast-growing algal species (Díaz & Marbà, 2009), affecting the distribution and recovery of *P. oceanica* meadows.
- **Presence of other seagrass species**: seagrass species such as *Zostera noltii* and especially *Cymodocea nodosa* can cohabit with *Posidonia oceanica* in coastal shallow waters (Pérez et al., 2015). All seagrass species are good bioindicators and provide multiple benefits, playing a crucial ecological role (Mateo, 2015).

Biotic structural characteristics

- **Meadow configuration:** heterogeneity in the configuration of the meadow plays a major role in the functioning of seagrass meadows and their associated fauna (Boström et al., 2006).
- **Patch size:** monitoring the size of different patches (only in patchy meadows) provides information about their evolution, allowing growth or regression to be detected.

- **Lower limit of the meadow**: lower limits are diffuse and usually not well represented in the cartography, thus making it important to define the limit in situ. This area also presents very challenging conditions for the growth of *Posidonia oceanica*, which struggles to survive at depth and with poor light availability, so its evolution should be monitored.
- **Living rhizome coverage**: interannual variations in the coverage can show changes in the meadow status (Díaz & Marbà, 2009). As with shoot density, living rhizome coverage is a good local descriptor of the meadow structure and an indicator of its ecological status.
- **Shoot (or beam) density**: this descriptor is highly sensitive to changes in the water conditions (Díaz & Marbà, 2009). As with living rhizome coverage, shoot density is a good local descriptor of the meadow structure and an indicator of its ecological status.
- **Burial degree of the rhizome**: this measurement monitors both the sedimentation rate and the hydrodynamics of the area. It can also be associated with sedimentation events that could be affecting the meadow. When burial depth is more than 10 cm it results in 50% shoot mortality and when more than 14-15 cm, 100% mortality (Cabaço et al., 2008). However, if sediment inputs are drastically reduced or local changes in the hydrodynamics occur, they can cause erosion of the matte (Blanc & Jeudy de Grissac, 1989; Medina et al., 2001).
- **Shoot sampling**: this enables different aspects of the plant to be analysed at the laboratory, where they cannot be measured in the field. Using these samples, it is possible to measure different morphological descriptors of the plant (epiphyte load, number of leaves, length and width of leaves) and to analyse the biochemical composition of the shoots.

Biotic functional characteristics

- **Flowering events:** massive flowering events of *Posidonia oceanica* could occur as a result of the plant's response to thermal stress (Díaz-Almela et al., 2007). It is important to detect these events as they are an early warning of climate change and water warming.
- **Evidence of mechanical pressures or signs of impact**: detecting potential pressures and impacts on the meadow is extremely important for assessing the habitat condition. This will also allow MSs to reduce those pressures or impacts in order to improve the condition of the habitat.
- **Sedimentation**: as explained in the section on of burial depth of the rhizome, *Posidonia oceanica* meadows are highly sensitive to changes in the sedimentation, so the sedimentation rate should be measured to evaluate possible changes. Besides this, and regarding functionality, *Posidonia oceanica* meadows reduce resuspension, contributing to increase the sediment retention and reducing erosion in the coastal zone (Gacia & Duarte, 2001).

1.2.3 Landscape/seascape characteristics

- **Habitat distribution/fragmentation**: mapping the habitat distribution is essential for evaluating its evolution and possible regression and fragmentation over time.

Table 1. Ecological characterisation and selection of variables used to measure habitat condition of habitat 1120* (*Posidonia* beds)

Ecological characteristics	Types	Description	Examples of associated variables
Abiotic characteristics	**Physical state characteristics**	Water transparency	Light attenuation in relation to depth.
		Substrate type	Morphology of the seafloor (sandy bottom, rocky bottom, dead *P. oceanica* matte). Visual observation.
		Water temperature	Temperature variation. Measured with a thermometer.
	Chemical state characteristics	Water quality	Multiple parameters to be analysed, including organic matter (especially carbon), nutrients (phosphates and nitrates) and pollutants.
		Sediment quality	Composition and particle size (granulometry) using sediment samples taken with a hand core.
Biotic characteristics	**Compositional state characteristics**	Presence of characteristic species	Density of *Pinna nobilis* individuals in the meadow. This species is currently facing extinction so its current conservation status makes it difficult to consider it as a characteristic species, or to use previous thresholds to evaluate its populations. There are different methods for evaluating its presence. One method is to establish a circular census, with a radius of 5-15 m (normally 10 m), around the central point of the monitoring station, to evaluate its presence/absence and mark living individuals.
		Sea urchin abundance	Density of individuals per square metre. Herbivory indicator.
		Holothuria spp. abundance	Density of individuals per square metre. Indicator of the environmental status.
		Associated fish	Species and abundance, through visual census using a linear transect. Abundance of *Sarpa salpa* used as an herbivory indicator.
		Presence of invasive algal species	Species, distribution and abundance (% present in the quadrats used to measure shoot density and living rhizome coverage). Also, visual exploration within a 20-metre radius of the central point of the monitoring station to record its presence or absence.
		Presence and cover of other seagrass species	% coverage of substrate colonised by other seagrass species in four random 10 m linear transects in the monitoring station. The % is calculated by adding together the length of each section found and then dividing this number by the total length of the transect.

Ecological characteristics	Types	Description	Examples of associated variables
Biotic characteristics	**Structural state characteristics**	Meadow configuration	Meadow typology (continuous or patchy/fragmented). Presence of vertical reefs of matte. Visual observation.
		Patch size (patchy meadows)	In patchy meadows, measurement and monitoring of the size of different control patches to measure its growth or regression.
		Lower limit of the meadow	Limit depth of the meadow, strongly associated with turbidity and poor light availability. Mark the limit with stakes to monitor its evolution over time.
		Living rhizome coverage	% coverage of living rhizome. Multiple replicates in a linear transect.
		Shoot (or beam) density	Two methods: number of *P. oceanica* shoots measured in the part of the studied meadow with the greatest coverage (selected density); and number of *P. oceanica* shoots in different replicates within a study transect (random density).
		Burial degree of the rhizome	Distance (cm) between the sediment surface and the ligule of an external leaf of a shoot. The distance is greater in the case of uprooting and less in the case of sedimentation events.
		Shoot sampling	Morphological and biometric parameters (number of leaves, length and width of leaves, etc.), biomass parameters (epiphytes load) and biochemical composition of the sampled shoots.
	Functional state characteristics	Flowering events	Massive flowering events associated with high temperatures. Visual observation.
		Evidence of mechanical pressures or signs of impact	Presence of mechanical pressures or signs of impact (mooring blocks, anchoring effects, etc.). Visual observation.
		Sedimentation	Sedimentation rate, using benthic sediment traps.
Landscape/ seascape characteristics		Habitat distribution (area) and fragmentation	Habitat distribution (area) and degree of habitat fragmentation. On a large scale, by mapping and using existing (and updated) cartography. On a smaller scale (monitoring station) it could be done by visually evaluating the meadow to define a ratio between living shoots and dead matte/substrate.

1.3 Selection of typical species for condition assessment

Posidonia oceanica is the main species that should be monitored to assess the condition of this habitat. However, there are many species present in this habitat, some of which are indicators of the environmental conditions and other processes that may affect the habitat, such as herbivory pressure.

Table 2 shows frequently present groups from which species for monitoring may be selected, and the types of changes in quality they could indicate.

Table 2. Selecting typical species for monitoring habitat 1120* (*Posidonia* beds)

Species group	Ecological notes	Sensitive to changes in quality
Angiosperms	Habitat builders (*P. oceanica* and other seagrass species).	Turbidity, physical disturbance, nutrients, productivity.
Macroalgae	Primary producers. Epiflora growing on leaves/rhizome or algae growing in the meadows. Also, invasive species colonising the habitat.	Turbidity, water quality and temperature, organic matter, nutrients, productivity, invasive species.
Bryozoa	Fragile organisms, very sensitive to physical disturbance. Epifauna living in the meadows.	Physical disturbance, contacts due to frequentation or anchoring.
Molluscs	Both infauna and epifauna.	Physical disturbance (especially P. nobilis, if its populations recover from their current conservation status), nutrients, productivity.
Nematodes	Both infauna and epifauna.	Nutrients, productivity, sediment quality.
Polychaetes	Both infauna and epifauna.	Nutrients, productivity, meadow typology.
Echinoderms	Both infauna and epifauna, also living among the meadows.	Sediment quality, nutrients, organic matter (detritivore species), herbivory pressure, productivity.
Fish	Meadows are nursery and spawning areas and offer rest and shelter. Species diversity as a habitat quality indicator and some species (*S. salpa*) are important herbivores.	Physical disturbance (habitat fragmentation), productivity, indicator of herbivory pressure.

Angiosperms are habitat builders that are highly sensitive to changes in environmental conditions. Seagrass species play a crucial ecological role in the marine environment (Mateo, 2015). It is therefore important to also monitor the presence of other species apart from *Posidonia oceanica*.

Macroalgae species are sensitive to changes in water conditions (nutrient inputs) and temperature, and the blooms of some species are an early warning of change. Also, invasive, fast-growing algae affect the distribution and recovery of *P. oceanica* meadows, colonising areas of dead matte (Díaz & Marbà, 2009).

Bryozoans are fragile organisms that are highly sensitive to mechanical impacts. They are therefore used, for example, as indicators for evaluating diving pressure in frequented areas (e.g., in the environmental monitoring conducted in the Medes Islands in Catalonia, Spain). Specifically, in *P. oceanica* meadows there are more than 90 species of bryozoans described as epifauna (Kocak et al., 2002), so this group should be considered, given its sensitivity and presence.

Molluscs are one of the most present groups in *Posidonia oceanica* meadows. For example, in Almería (Spain) more than 200 mollusc species have been described in this habitat (Templado et al., 2004). The species *Pinna nobilis*, which is endemic to the Mediterranean Sea, was once a typical species found in *Posidonia oceanica* meadows and was used as an indicator species to evaluate water quality and the conservation status of the meadow (Díaz & Marbà, 2009). However, *P. nobilis* is now critically endangered, with its populations on the brink of extinction, so it is essential to find and monitor healthy individuals.

Nematodes are mostly found in the rhizomes of *Posidonia oceanica* meadows and are one of the most important groups among their meiofauna. This group is considered a good indicator for evaluating environmental quality in the meadows, since it shows early responses to environmental disturbance in their composition and diversity (Losi et al., 2012). These compositional changes in nematode communities are noticeable after a decrease in oxygen availability, which could potentially have major implications for the benthic food web and the functioning of the seagrass systems (Gambi et al., 2009).

Polychaetes are mostly present in the lower stratum of the meadow, with around 80% of the individuals of this group found in the rhizome (Templado et al., 2004) and the matte (Wilsie, 1983), although some species can also be found on the leaves. The functional diversity of benthic polychaete communities has been observed to change between different types of *P. oceanica* habitats (continuous meadow, patchy meadow and dead matte), showing how specific traits relate to the different habitat types in terms of functional composition, diversity and the resultant ecosystem functions involved (Katsiaras et al., 2022).

In the **echinoderm** group, some species of holothurians and herbivore (or omnivore, feeding on epiphytes) sea urchins are very abundant and play an important role in the meadow dynamics (Díaz & Marbà, 2009). Holothurians, like detritivores, ingest sediment to feed on the organic matter present, and therefore play a key part in recycling the organic detritus produced by the meadow. Also, the species diversity depends on the meadow morphology, with some species having more affinity for particular meadow types (e.g., *Holothuria tubulosa* is frequent in dense meadows). In the case of sea urchins, *Paracentrotus lividus* is the most abundant and studied species, playing a small role in the herbivory pressure on healthy meadows (Tomas et al., 2005a) and an important role in feeding on epiphytes present on the leaves (Tomas et al., 2005b). As previously explained, density fluctuations of sea urchin populations can also affect meadows, causing massive defoliation (Ruiz et al., 2001).

Lastly, **fish** biodiversity is high in this habitat (Templado et al., 2004) and should therefore be evaluated in order to fully assess habitat condition. Also, as previously mentioned, *Sarpa salpa* plays an important role as an herbivore in the meadow (Alcoverro et al., 1997) and is a good species for evaluating herbivory pressure.

2 Analysis of existing methodologies for the assessment and monitoring of habitat condition

This analysis has considered the methodologies used for assessing and monitoring the condition of habitat 1120* in six out of eight Member States where *Posidonia oceanica* beds are present: Spain (ES), Croatia (HR), France (FR), Italy (IT), Malta (MT) and Slovenia (SI). These methodologies have been implemented to fulfil the reporting requirements of the **Habitats Directive** (Díaz & Marbá, 2009 for Spain; Bakran-Petricioli, 2007 and Guala et al., 2014 for Croatia; Lipej et al., 2022 for Slovenia; Lepareur, 2011 for France) and the **Marine Strategy Framework Directive** (MSFD) (Ministero dell'Ambiente e della Tutela del Territorio e del Mare, 2017 for Italy; Environment and Resources Authority, 2023 for Malta).

Greece does not present a common methodology for monitoring habitat 1120* under the Habitats Directive. However, a comparison test of four indexes used to assess the ecological status of coastal waters following the **Water Framework Directive** (WFD) has been implemented in the Saronikos Gulf (Gerakaris et al., 2017).

Cyprus monitors *P. oceanica* as one of the four Biological Quality Elements (BQEs) of the coastal waters as defined in the WFD (Law 13(I)/2004). Since 2019, Cyprus has been mapping habitat 1120* along the entire national coast using the latest technology. Furthermore, a monitoring programme was initiated in the six marine areas of the Natura 2000 network and an assessment of the ecological status was carried out by analysing an array of metrics and applying several biotic indexes[1].

2.1 Variables used, metrics and measurement methods, existing data sources

A summary of the main ecological characteristics and the associated variables useful for measuring the condition of *Posidonia* beds (*Posidonion oceanicae*) is presented in Table 1 (Section 1). An overview of variables used by Member States to monitor and evaluate the status of this habitat is presented in the following table (Table 3). Table 4 shows the commonalities and differences in the variables used in the six Member States considered in this analysis.

[1] Mapping and evaluation of *Posidonia* meadows and other important marine habitats under the European Habitats Directive (92/43/EEC), in the coastal waters of Cyprus: https://www.merresearch.com/portfolio/mapping-and-evaluation-of-posidonia-meadows-and-other-important-marine-habitats-under-the-european-habitats-directive-92-43-eec-in-the-coastal-waters-of-cyprus/

Table 3. Examples of variables used by Member States to assess condition of habitat 1120* (*Posidonia* beds)

Description	Examples of variables used by Member States	Notes
1. Abiotic characteristics		
1.1 Physical state characteristics		
Substrate	Three Member States (MSs) analyse the type of substrate where the meadows grow: • **HR** - substrate type. • **MT** - substrate type. • **FR** - substrate type (rock, matte or sand).	The type of substrate determines the morphology of the seagrass bed and is sometimes considered a structural descriptor of the habitat. All cases where the substrate is specifically monitored have been included here to simplify the analysis.
Sediment type	Two MSs analyse specifically the particle size of the sediment conducting a granulometric analysis: • **IT** - granulometric analysis (particle size). • **FR** - volume and granulometric analysis specifically of the sediment present in the matte.	One of the MSs evaluates the granulometry of the sediment as part of the characterisation of the matte (focusing only on the sediment present). It has also been included in the 'Structural state characteristics' section of this table as part of the matte analysis. Some MSs do not analyse sediment size but do analyse the sedimentation rate of a meadow as a functional variable.
Water (physical parameters)	Three MSs analyse different physical aspects of water condition, with temperature and transparency/turbidity being two of the main aspects measured: • **ES** - temperature, transparency. • **MT** - different physical parameters of water. • **IT** - different physical parameters of water.	Two MSs specify the parameters measured in their methodology while the third MS specifies the use of a multiparameter probe to analyse water condition. Commonly measured physical parameters using a multiparameter probe are temperature, depth and turbidity.
1.2 Chemical state characteristics		
Water (chemical parameters)	One MS indicates to use a multiparameter probe to analyse water quality without specifying which parameters to measure and one MS analyses different chemical parameters of water through other monitoring procedures: • **MT** - different chemical parameters of water. • **IT** - different chemical parameters of water.	Some MSs do not specify a water quality analysis in their methodology for monitoring the habitat, but they show the importance of water quality as a limiting factor for *Posidonia oceanica*'s growth and distribution.
Sediment quality	Three MSs analyse the concentration of different elements present in the sediment: • **ES** - hydrogen sulphide concentration in calcareous-type sediment. • **IT** - total organic carbon in sediment. • **FR** - organic matter rate in the sediment of the matte; thickness of the oxidised sediment layer, colour and odour.	One of the MSs evaluates the organic matter present in the matte sediment. It is included as part of the characterisation of the matte and has also been included in the 'Structural state characteristics' section of this table as part of the matte analysis. Some MSs do not analyse sediment quality but do analyse the sedimentation rate of the meadow for different nutrients and compounds as a functional variable for the habitat.

Description	Examples of variables used by Member States	Notes
2. Biotic characteristics		
2.1 Compositional state characteristics		
Presence of characteristic/ endangered species	Four MSs monitor the density of individuals of the various characteristic species of *Posidonia* beds, commonly using the noble pen shell (*Pinna nobilis*): • **ES** - density of noble pen shell (*Pinna nobilis*) individuals. • **HR** - density of noble pen shell (*Pinna nobilis*) individuals. • **FR** - presence of protected, sedentary or territorial threatened species (i.e., *Pinna nobilis*). • **SI** - density of stony coral (*Cladocora caespitosa*) and sponge (*Geodia cydonium*).	Three MSs monitor *Pinna nobilis* as a characteristic and protected species of this habitat (which could be also used as an indicator for mechanical impacts on the meadow) but its current conservation status means it is no longer a common species found in habitat 1120*. One MS monitors specifically the presence of endangered and protected sessile species (*Cladocora caespitosa* and *Geodia cydonium*) but these may not be the most representative species of Neptune grass meadows.
Indicator species	Three MSs analyse the presence and/or density of different species that can be used as indicators: ES monitors the density of species that indicate changes in nutrient and organic matter concentrations (sea urchins and sea cucumbers), FR monitors the density of herbivore species to evaluate herbivory pressure, and HR monitors the presence of other seagrass species. • **ES** - density of sea urchins (*Paracentrotus lividus, Sphaerechinus granularis* and *Psammechinus microtuberculatus*) and *Holothuria* spp. (*H. tubulosa, H. polii, H. foskalii* and *H. xantorii*). • **FR** - density of herbivore (or mainly herbivore) species (*Sarpa salpa* and *Paracentrotus lividus*). • **HR** - presence of other seagrasses (*Cymodocea nodosa* and *Zostera noltii*).	Seagrasses have been included as indicator species as they can be used for evaluating environmental conditions and some mechanical pressures/impacts in the area. However, *Posidonia oceanica* itself is highly sensitive to changes and is already a great indicator species.
Presence of invasive species	Five MSs monitor the presence and abundance of invasive algal species, mainly *Caulerpa* spp: • **ES** - presence and abundance of *Caulerpa taxifolia, Caulerpa cylindracea* or *Lophocladia trichoclados*. • **FR** - presence and estimation of the % coverage of invasive species (*Caulerpa taxifolia, Caulerpa cylindracea* and red algae). • **HR** - presence/abundance of invasive or alien species (i.e., *Caulerpa taxifolia* and *Caulerpa cylindracea* or *Womersleyella setacea*). • **IT** - presence of alien algae. • **MT** - % coverage of invasive algae.	Only one MS (SI) does not specifically address invasive species. The monitoring carried out in all MSs focuses on the presence and coverage of this type of species.

Description	Examples of variables used by Member States	Notes
Associated species	Four MSs address different species (fish, associated species, etc.) that make up the community present in this habitat: • **SI** - fish community inventory. • **HR** - species composition. • **IT** - associated species. • **FR** - associated fauna and flora (only herbivore and protected or endangered species).	One MS (SI) evaluates the fish community, one MS (IT) lists the associated invertebrate species that can be found in this habitat (infauna, epifauna and endofauna) but does not suggest a monitoring methodology nor consider it part of the habitat monitoring. One MS (FR) evaluates only the presence of herbivore species (which is also included in the 'Indicator species' section of this table) and protected or endangered species (which is also included in the 'Presence of characteristic/endangered species' section of this table). One MS (HR) suggests evaluating the condition of typical species and communities and the species composition but do not specify which species should be monitored and how.
2.2 Structural state characteristics		
Meadow limits	Five MSs monitor the lower limit of the meadow, which is strongly associated with depth and light attenuation. The variables measured are its position using a GPS, its depth, and the type of limit, that is visually characterised: • **HR** - position and depth of the lower limit and type of limit (progressive, sharp cover >25%, sharp cover <25%, sparse and regressive). • **IT** - depth of the lower limit and type of limit (regressive, progressive or stable). • **ES** - depth of the lower limit and type of limit (progressive or regressive, sharp or continuous). • **MT** - depth of the lower limit, type of limit (regressive, progressive or stable) and also evaluates different aspects of the plant specifically at the lower limit of the meadow (rhizome baring and % of plagiotropic rhizomes). • **FR** - depth of the lower limit, type of limit (progressive, clear with high overlap, clear with low overlap, sparse and regressive) and also evaluates different aspects of the plant, specifically at the lower limit of the meadow (% coverage of shoots and % of plagiotropic rhizomes). Two MSs also evaluate the depth and the position of the upper limit of the meadow: • **ES** - depth of the shallow limit of the meadow. • **HR** - position and depth of the upper limit of the meadow.	The lower limit of the meadow could be also considered a 'physical state characteristic' as it is strongly influenced by a physical factor (depth). It has been included in this section because the limits of the meadow are also a structural aspect of the habitat. Five MSs evaluate the lower limit of the meadow and two MSs also evaluate its upper/shallow limit.
Meadow morphology	Four MSs (HR, FR, IT, MT) address the morphology of meadows and their continuity. One MS address the size of patches in patchy meadows. • **HR** - distribution of the meadow according to the sea bottom. • **FR** - type of seagrass bed, morphology. • **IT** - meadow continuity. • **MT** - meadow continuity. • **ES** – patch size in patchy meadows.	This section includes different descriptive aspects of a meadow analysed by the MSs (though some MSs have not provided much information on the methodology followed) and the methodology used by one MS to monitor the evolution of patches specifically in patchy meadows. Note that the continuity of a meadow could be also considered a meso-coverage analysis but it has been included here as there was no specific information on the methodology used to evaluate it.

Description	Examples of variables used by Member States	Notes
Coverage of living rhizomes/dead matte	Five MSs evaluate the coverage of living rhizomes or dead matte: • **ES** - % of surface covered by living rhizomes (2 different methodologies can be applied depending on the type of transects used). Also calculates a Conservation Index based on the % coverage of living rhizome and the % coverage of dead matte. • **FR** - coverage rate at the lower limit of the meadow. Also estimates the % of meadow covered by dead matte. • **HR** - % coverage in a 10 m linear transect. • **IT** - coverage (% of living rhizome and % of dead matte). • **MT** - % cover of dead matte.	Only some MSs have a detailed methodology on how to measure % coverage. One MS focuses on coverage only in the lower limit (included in the 'Meadow limits' section).
Shoot/beam density	All six MSs measure the shoot density of the meadow: • **ES** - shoot density measured in defined monitoring stations. • **HR** - shoot density measured in defined monitoring stations. • **FR** - density of shoots at -15 m. • **IT** - density of shoots at -15 m. • **MT** - density of shoots at -15 m and at the lower limit of the meadow. • **SI** - density of shoots at different depths.	Shoot density is one of the most used descriptors for assessing the environmental status of *Posidonia oceanica* meadows and evaluating their evolution. All 6 MSs use this descriptor but not all of them specify the methodology used to calculate the density.
Rhizome burial and matte characterisation	Four MSs monitor different variables of the lower stratum of the meadow (rhizome/matte): • **ES** - burial degree of the rhizome. • **FR** - distance between the sediment and the lower part of the rhizomes; compactness of the matte; structure of the matte (volume of sediment, size of particles and organic matter rate). • **IT** - uprooting of rhizomes. • **MT** – rhizome baring and % of plagiotropic rhizomes.	Most of the MSs do not evaluate specifically the lower stratum of the meadow. One MS carries out an in-depth analysis and the other three MSs focus on the sedimentation rate, by evaluating the exposure of the rhizomes and monitoring their evolution. One of them, additionally, evaluates the % of plagiotropic rhizomes.
Shoot/beam analysis	Three MSs use shoot sampling for studying different variables in the lab: • **FR** - morphometric, biometric, lepidochronological and biomass parameters (epiphytes) analysed. • **IT** - morphometric, lepidochronological and biomass parameters. • **MT** - leaf surface area, epiphytes/leaf biomass.	Shoot sampling, with the consequent lab analysis, shows aspects of the state of the plant that cannot be shown in the field. The methodologies applied and the variables studied are not equally explained between the different MSs.

Description	Examples of variables used by Member States	Notes
2.3 Functional state characteristics		
Sedimentation rate	One MS evaluates different variables of the sedimentation: • **ES** - total sedimentation and organic matter, phosphorus and iron.	Sedimentation rate was analysed separately from sediment quality and size sections because it shows a functional aspect of the habitat. The concentration of each compound analysed can also be a useful variable for analysing sediment quality.
Shoot and rhizome dynamics	One MS analyses different variables in the field to evaluate the dynamics of the rhizomes and shoots of a meadow: • **ES** - vertical rhizome growth and shoot dynamics (birth, growth, mortality).	The methodology used by this MS is well defined but it is not commonly evaluated by other MSs.
Flowering events	Four MSs evaluate flowering events. FR, IT and MT evaluate the sexual reproduction events (flowering) of the plant. HR gathers this information only as additional information during the monitoring. • **FR** - presence of flowering. • **IT** - presence of flowering. • **MT** – presence of flowering. • **HR** - included only as an additional note during the monitoring.	Flowering events are not equally common across the entire Mediterranean region. However, this variable is highly recommended as there is evidence of *Posidonia oceanica* massive flowering events caused by rising temperatures and climate change.
Impacts and disturbances	Five MSs evaluate different chemical and mechanical pressures/disturbances affecting the habitat: • **HR** - presence of pressures and impacts in the area (mechanical, chemical, etc.). • **IT** - disturbance sources. • **MT** – presence of pressures for each coastal water body and presence of pollutant sources and physical disturbances at monitoring stations. • **FR** - presence of physical disturbances and pollutant source. • **ES** - obtained from the Water Framework Directive in the analysis of the water bodies surrounding the meadow.	Two MSs evaluate both chemical and mechanical pressures that could affect habitat 1120*. One MS does not specify the methodology or the variables to be monitored as disturbance sources. One MS assesses the pressures for each coastal water body pursuant to the WFD, which then informs the selection of monitoring locations for operational monitoring stations, which consider the presence of pollutant sources and physical disturbances. One MS does not specifically monitor the pressures/disturbances in the meadow but instead uses information from the WFD for this evaluation, excluding potential local mechanical impacts such as the effect of mooring on the meadow.
3. Landscape/seascape characteristics		
Habitat distribution/ fragmentation	Three MSs analyse the area covered by the habitat, one of them suggests analysing the degree of habitat fragmentation: • **ES** –area covered by the habitat • **FR** - area covered between the upper and lower limit of the meadow. Degree of habitat fragmentation. • **MT** - area covered by the habitat.	The MSs describe how to calculate the area covered by the habitat using remote (satellite, side-scan sonar, etc.) and in situ (diving) measurements depending on the area to cover. However, they do not provide a specific methodology to follow, only the methods that could be used. One MS suggests monitoring habitat fragmentation, but no protocol has been included

Table 4. Main ecological characteristics and associated variables monitored in the assessment of structure and function of habitat 1120* (*Posidonia* beds) by EU Member States

Ecological characteristics	Variables	Metrics	ES	HR	FR	IT	MT	SI
1. Abiotic characteristics								
1.1 Physical state characteristics								
Substrate	-Substrate type	-Type (rock, matte or sand).						
Sediment type	-Granulometric analysis of the sediment present in the matte.	-Particle size.						
Water (physical parameters)	-Different physical parameters of water.	-Temperature, turbidity (Secchi depths, NTU).						
1.2 Chemical state characteristics								
Water (chemical parameters)	-Different chemical parameters of water.	-Dissolved Oxygen (mg / l, % saturation), pH, salinity (psu).						
Sediment quality	-Hydrogen sulphide concentration in calcareous-type sediment. -Total organic carbon in sediment. -Organic matter rate in the sediment of the matte; thickness of the oxidised sediment layer, colour and odour.	-µM / l -% -N/A						
2. Biotic characteristics								
2.1 Compositional state characteristics								
Presence of characteristic/ endangered species	-Density or presence of noble pen shell (*Pinna nobilis*) -Density of stony coral (*Cladocora caespitosa*) and sponge (*Geodia cydonium*).	-Nº individuals / m^2 or 100 m^2, presence in the meadow. -Nº individuals / 100 m^2 (*Cladocora caespitosa* and *Geodia cydonium*).						
Indicator species	-Density of sea urchins, *Holothuria* spp. or *Sarpa salpa.*	-Individuals / m^2 -Presence / absence, % of coverage in transects.						

Ecological characteristics	Variables	Metrics	ES	HR	FR	IT	MT	SI
	-Presence of other seagrasses (*Cymodocea nodosa* and *Zostera noltii*).							
Presence of invasive species	-Presence and coverage of invasive species (*Caulerpa taxifolia, Caulerpa cylindracea, Lophocladia trichoclados, Womersleyella setacea* or red algae).	-Presence/absence, % of coverage, biomass (g / m^2).						
Associated species	-Fish community, species composition, associated fauna and flora.	-Fish inventory, presence, abundance.						
2.2 Structural state characteristics								
Meadow limits	-Position and depth of lower limit. -Position and depth of upper limit.	-Lower limit: • Position and depth. • Type of limit: ○ Progressive, sharp cover >25%, sharp cover <25%, sparse and regressive. ○ Progressive, regressive or stable. ○ Progressive or regressive, sharp or continuous. ○ Progressive, clear with high overlap, clear with low overlap, sparse and regressive. -Upper limit: • Position and depth.						
Meadow morphology	-Size of patches. -Distribution of the meadow according to sea bottom. -Type of seagrass bed according to its morphology. -Meadow continuity.	-Enlargement / regression of patches in patchy meadows. -Flat / step, continuous / discontinuous, a series of characteristics from a standardised list. -Type according to a standardised list. -N/A						
Coverage of living rhizomes/dead matte	-Coverage of living rhizome -Coverage of dead matte.	-% coverage of living rhizome: • In a linear transect (10 or 25 m). • In a 40x40 cm^2 quadrat. • Conservation Index. • At the lower limit. -% coverage of dead matte.						
Shoot/beam density	-Shoot density.	-Number of shoots / m^2 at different depths or at a fixed depth (usually 15 m depth).						

Ecological characteristics	Variables	Metrics	ES	HR	FR	IT	MT	SI
Rhizome burial and matte characterisation	-Burial degree of the rhizome. -Plagiotropic rhizomes.	-cm of burial of rhizomes / distance from the sediment to the lower part of rhizomes. -% of plagiotropic rhizomes.						
Shoot/beam analysis	-Morphometric, lepidochronological and biomass (epiphytes) parameters.	-Longest leaf per shoot / annual leaves production / annual rhizome production / total rhizome weight / annual rhizome elongation, number of leaves per rhizome per year / rhizome length and age / Epiphyte/leaf biomass ratio (epiphyte load).						
2.3 Functional state characteristics								
Sedimentation rate	-Total and organic sedimentation rate. -Phosphorous and iron sedimentation rate.	-g/m^2 day. -mg/m^2 day.						
Shoot and rhizome dynamics	-Vertical rhizome growth. -Presence of new shoots. -Net population growth. -Shoot mortality	-Millimetres per internode. -New shoots per m^2 per year. -Growth per m^2 per year. -Dead shoots per m^2 per year/mortality per capita (year^{-1})						
Flowering events	-Presence of flowing events.	-Presence / absence (as essential data or as additional data).						
Impacts and disturbances	-Presence of pressures.	-Presence of pressures by visual evidence (from a standardised list as WFD or others).						
3. Landscape/Seascape characteristics								
Habitat distribution/ fragmentation	-Area covered by the habitat. -Degree of fragmentation.	- m^2 / ha -% of not covered surface (degree of fragmentation).						

2.2 Definition of ranges and thresholds to obtain condition indicators

This section describes some of the ranges and thresholds used by different Member States to obtain condition indicators for habitat 1120*. The definition of ranges and thresholds for this habitat varies between countries but not all MSs have clear and defined thresholds. Some MSs use specific reference values for indicators, others rely on expert judgement, others use indexes to evaluate the ecological status of *Posidonia oceanica* meadows and some use a combination of these.

The following table (Table 5) gives examples of reference values used by **Spain** to record the condition of this habitat (Díaz & Marbá, 2009), which are mostly focused on biotic variables. Some of the thresholds used are merely descriptive but give an indication on how to define the condition of the variable analysed. Of all the MSs, Spain has the most-defined ranges and thresholds.

Table 5. Thresholds established by Spain for different variables

FV: favourable; U1: unfavourable; U2: unfavourable declining

Variable	Metric	Reference value/ threshold/ condition indicator
Physical		
Water transparency	Light attenuation coefficient k (m^{-1})	FV: potential maximum depth of the meadow > its current limit. U1: water transparency has reduced but potential depth of the meadow ≥ depth of the last depth limit of the meadow measured. U2: current and potential depths have decreased since the last limit measured.
Chemical		
Hydrogen sulphide concentration in calcareous-type sediments	µM/l	FV: <10 µM/l U1: 10-30 µM/l U2: >30 µM/l
Biotic		
Sea urchins (*Paracentrotus lividus*)	ind/m^2	Normal reference density: 0-5 ind/m^2
Holothurians	ind/m^2	Normal reference density: 0.02-5 ind/m^2
Invasive species	Presence/ absence	FV: no presence observed U1: presence observed in meadow clearings U2: presence observed in the meadow
Deep limits	m	FV: depth stable or increased U1: depth decreased ≤ 1 m U2: depth decreased ≥ 1 m
Shallow limits	m	FV: depth stable or decreased U1: depth increased <1 m or meadow area loss <5% U2: depth increased >1 m or meadow area loss >5%
Size of patches	m^2	FV: the mean size of patches has increased. U1: the mean size of patches is stable or has slightly decreased. U2: the mean size of patches has slightly decreased or 10% of patches have disappeared.

Variable	Metric	Reference value/ threshold/ condition indicator
Coverage	%	FV: mean coverage has increased or has not changed significantly between two surveys. U1: mean coverage at the monitoring station has decreased significantly, by less than 30%, between two surveys. Or, with three or more surveys, a significant rate of decrease in coverage, of less than 10% a year. U2: mean meadow coverage at the station has decreased significantly, by more than 30% between two surveys. Or, with three or more surveys, a significant rate of decrease in coverage equal to or greater than 10% a year is observed.
Conservation index	CI = (% living *P. oceanica*) / (% dead matte + % living rhizome)	FV: IC ≥ 0.8 U1: IC = 0.6-0.8 U2: IC < 0.6
Shoot density per depth	N/m²	FV: density normal or high for its depth U1: density low for its depth U2: density very low for its depth
Rhizome burial degree	Cm	FV: -5.5 cm U1: -5.5 cm to -7.7cm U2: > -7.7 cm
Growth of shoots at permanent monitoring plots	N/m²	FV: shoot density ≥ prior to census U1: shoot density 5-10% lower U2: shoot density >10% lower
Shoot mortality	N/m² year	FV: decline <5% and/or compensated by new shoots U1: decline 5%-10% U2: decline ≥10%
New shoots	New shoots/m² year	FV: net growth positive or not less than -5%/year U1: no new shoots but decline <7%/year U2: no new shoots but decline >7%/year
Vertical rhizome growth	Mm	FV: the average annual growth in the last 5 years is not significantly less than the average annual growth of the previous 5 years. U1: the average annual growth in the last 5 years is significantly less than the average annual growth of the previous 5 years, but the average growth in the most recent period is 20% more than the average growth of the previous period. U2: the average annual growth in the last 5 years is significantly less than the average annual growth of the previous 5 years, and the average growth in the most recent period is 5 times less than the average growth in the previous period.
Total sedimentation	g (dry matter) / m² day	FV: <5 g/m² day U1: 5-8 g/m² day U2: >8 g/m² day
Organic sedimentation	g OM (dry matter) / m² day	FV: <1.5 g/m² day U1: 1.5-4 g/m² day U2: >4 g/m² day
Phosphorus sedimentation	mg P (dry matter) / m² day	FV: <50 mg/m² day U1: 50-120 mg/m² day U2: >120 mg/m² day
Iron sedimentation	mg Fe (dry matter) / m² day	FV: >43 mg/m² day U1: <43 mg/m² day

Croatia lacks a baseline, instead using modified standard thresholds of UNEP-RAC/SPA (UNEP-RAC/SPA, 2011) for different biotic variables to evaluate the conservation status of *Posidonia oceanica* (Guala et al., 2014). Those thresholds establish 5 different categories (bad, poor, moderate, good, high) and are used for the following variables (Table 6):

Table 6. Thresholds established by Croatia for different variables

Variable	Metric	Threshold
Lower limit	m depth	High: >34.2 m Good: 34.2 m - 30.4 m Moderate: 30.4 m - 26.6 m Poor: 26.6 m - 22.8 m Bad: <22.8 m
Type of lower limit	Descriptive	High: progressive Good: sharp (cover >25%) Moderate: sharp (cover <25%) Poor: sparse Bad: regressive
Cover of the lower limit	% living *P. oceanica*	High: >35% Good: 35% - 25% Moderate: 25% - 15% Poor: 15% - 5% Bad: <5%
Shoot density	Shoots/m^2	*See details in Guala et al., 2014*

Another variable monitored by **Croatia** with established thresholds is the Conservation Index (% living *P. oceanica*) / (% dead matte + % living rhizome)) of the meadow (Table 7) (Montefalcone, 2009):

Table 7. Thresholds established in the Conservation Index in Croatia

Variable	Metric	Threshold
Conservation index	CI = (% living *P. oceanica*) / (% dead matte + % living rhizome)	High: >0.9 Good: 0.7 - 0.9 Moderate: 0.5 - 0.7 Poor: 0.3 - 0.5 Bad: <0.3

Slovenia has a defined threshold to evaluate specifically the shoot density (number of shoots/m^2) of *P. oceanica* meadows (Table 8), establishing five different categories:

Table 8. Thresholds established to evaluate shoot density in Slovenia.

Variable	Metric	Threshold
Shoot density	Number of shoots / m^2	Very good condition: >750 shoots / m^2 Good condition: 500-749 shoots / m^2 Moderate condition: 250-499 shoots / m^2 Bad condition: 50-249 shoots / m^2 Very bad condition: <50 shoots / m^2

The need to monitor this habitat and fulfil the requirements of European directives (particularly the WFD) led to the creation of different indexes to define its ecological status (e.g., POMI index (Romero et al., 2007), PREI index (Gobert et al., 2009)). These indexes are good tools for evaluating different variables and quantifying them.

One of these indexes, the *Posidonia oceanica* Rapid Easy Index (PREI), is used by **Malta** and **Italy** to partially evaluate the ecological status of *Posidonia* beds. This index was developed to be simple and cost-effective, using different indicators that together provide an Ecological Quality Ratio (EQR) on a scale from 0 to 1. The Index establishes the following thresholds (Table 9) (Gobert et al., 2009):

Table 9. Ecological Quality Ratio

EQR	Ecological status	Colour code
1 − 0.775	High	Blue
0.774 − 0.550	Good	Green
0.549 − 0.325	Moderate	Yellow
0.324 − 0.100	Poor	Orange
<0.100 – 0	Bad	Red

In **France** there are no specific threshold values or ranges of values for the moment, so expert opinion is using the results obtained for the different variables studied to asses ecological status.

2.3 Aggregation methods at the local scale

Most Member States use a common rather than a habitat-specific methodology to aggregate data on indicators at the local scale. Thus, the general conservation status of a natural habitat in a Natura 2000 site is considered "favourable" if its structure, functions and composition allow the sustainability of the habitat over time and its stability or expansion in space (Maciejewski et al., 2016).

This general approach is used in **Spain**, where the evaluation of the conservation status of Posidonia beds, based on the structure and function of a meadow, is determined at two levels: at a smaller scale (monitoring station level) and at a bigger scale (meadow level). This evaluation includes different structural, dynamic and environmental variables that together define the conservation status of the habitat. At the smaller scale (monitoring station level), variables are aggregated to obtain the following overall status:

- **Favourable**: when all structural and environmental variables are classified as 'favourable'.
- **Unfavourable-inadequate**: when some structural variables are classified as 'favourable' and others as 'unfavourable-inadequate'.
- **Unfavourable-bad**: when at least one structural variable is classified as 'unfavourable-bad'.

At the bigger scale (meadow level), variables are aggregated to obtain the following overall status:

- **Favourable**: when the status of all stations is 'favourable'.
- **Unfavourable-inadequate**: when the status of some stations is 'favourable', but 'unfavourable-inadequate' for others, or when the status of all stations is 'unfavourable-inadequate'.
- **Unfavourable-bad**: when the status of some stations is 'unfavourable-bad'.

In **France**, one approach considered for aggregating data at the local scale is the PatriNat method. This method situates the conservation status of a habitat along a gradient ranging from unfavourable to favourable. It evaluates three parameters of the habitat (surface, structure and functions, and alterations), represented by one or more indicators filled in or calculated from metrics recorded in the field. Each indicator evaluated will be compared to a threshold value, which has not yet been clearly defined for this habitat. Following this method, each indicator is given a score (negative or zero) which will be subtracted from a starting score of 100. An indicator result reflecting a good status will have few points subtracted and an indicator reflecting a bad status will have more points subtracted, obtaining an overall status plotted on the gradient (Delavenne & de Bettignies, 2023).

2.4 Aggregation at biogeographical scale

Member States currently follow the recommendations of the Art. 17 reporting guidelines for the period 2013-2018, which establishes that "if 90% of the habitat area is reported as in 'good' condition, then the status of the 'structure and functions' parameter is 'favourable'. If more than 25% of the habitat area is reported as 'not in good condition', then the 'structure and functions' parameter is 'unfavourable-bad'.

2.5 Selection of localities

The selection of localities for monitoring habitat 1120* and establishing its conservation status varies according to Member State. The general criterion is to achieve representativeness of the habitat for the entire biogeographic region in which it is distributed. For this, the Member States consider different criteria such as the availability of a reference cartography for defining sampling areas, the presence of the habitat in protected areas, previous monitoring carried out within the framework of other European directives (mainly the WFD and the MSFD), or the various environmental conditions at the regional level (e.g., areas with a greater or lesser degree of pressures and impacts). Generally, these criteria are used together with expert judgement to define the monitoring localities. The locality-selection methods used by the Member States who specify this process in their methodology is detailed below.

In **Spain**, nineteen localities have been selected, distributed among the autonomous regions of the Spanish Mediterranean coast (Díaz & Marbà, 2009). These were selected on the basis of prior knowledge provided by surveillance and monitoring networks for this habitat (which were already established and operating in the various regions and had defined sampling stations), the availability of updated cartographies for this habitat, and expert judgement. Each locality should have different monitoring stations, and the number of stations depends on the depth range of each meadow: three monitoring stations for meadows with a wide range of depths, following the bathymetric gradient (a shallow station (5-7 m deep), an intermediate station (14-15 m deep) and a deep station, at the edge of the meadow). For meadows with a narrower range of depth, or where it is not possible to carry out sampling, the intermediate station can be omitted, leaving one deep station and one shallow station per locality. For localities with sparse or patchy meadows, a single monitoring station will suffice.

Croatia selects the meadows to be monitored on the basis of different environmental conditions, using criteria from Annex III of the Habitats Directive. Thus, it established 50 monitoring sites, grouped according to 3 priority levels (Guala et al., 2014): 1) relevant areas for conservation reasons (15 sites), including protected areas (MPAs, Natura 2000 sites) and areas where previous data on the conservation status of the habitat were available; 2) areas that suffer anthropic pressures and impacts (20 sites); 3) areas relevant to other ongoing monitoring purposes (WFD or MSFD) (15 sites). Guala et al. (2014) define a site as a continuous area where the meadow is distributed for about 1 km in length. At each site, three zones about 100 m apart are to be selected, and in each zone three monitoring stations are chosen at different bathymetric intervals: shallow (<10 m), intermediate (12-18 m) and deep (as close as possible to the lower limit). Stations are chosen randomly in the first year and remain fixed over the six-year monitoring programme.

In the case of **Slovenia**, given the size of its coast, 5 monitoring localities were established (Lipej et al., 2022), covering all the larger fragments of meadows present. Sampling was defined for three different depth ranges: 1-2 m, 2-3 m and 3-4 m.

Italy does not have established monitoring localities for evaluating the environmental status of habitat 1120* (Ministero dell'Ambiente e della tutela del territorio e del Mare, 2017). The proposed method for defining the localities is similar to that established in Croatia: to use the available cartography, to establish the representativeness of the habitat on the basis of different environmental conditions and considering existing monitoring data of other Directives and MPAs.

Malta has established a network of monitoring stations directly linked to the WFD and its requirements (Environment and Resources Authority, 2024). This network is composed of both “surveillance stations” (representative of the water body, where anthropogenic activity is not significant) and “operational stations” (to assess the impacts from point sources, diffuse sources and hydromorphological changes). These locations are also identified to monitor the condition of habitat 1120* in relevant marine Natura 2000 sites. The process for selecting monitoring localities involves expert judgement, pressures assessment and the results obtained in previous monitoring efforts.

France did not specify a process for defining monitoring localities in their methodology for assessing the habitat condition of *Posidonia* beds, being expert judgement the criterion followed.

2.6 General monitoring and sampling methods

Article 17 of the Habitats Directive requires six-yearly reporting on the conservation status of habitats, including Annex I habitats such as 1120* (*Posidonia* beds). The reviewed methodologies of the different Member States do not specify in all cases the monitoring frequency, but a minimum six-yearly monitoring is required. The monitoring techniques specified in the various methodologies is similar among the MSs, and some of the variables are measured in situ. This is due to the distribution range of habitat 1120*, which makes it technically easy to monitor variables by diving, which is also a cost-effective methodology.

The level of detail in the description of the monitoring and sampling methods used by the Member States is variable. **Spain** (Díaz & Marbà, 2009) and **Croatia** (Guala et al., 2014) give a precise explanation of the methodology used to monitor the habitat, the criteria used to define and select monitoring localities, the monitoring frequency of each variable, and the establishment of thresholds for some of the variables monitored.

Some MSs (**Italy** and **Malta**) use an environmental index, such as the PREI index (Gobert et al., 2009) to assess habitat 1120* *Posidonia* beds. In the case of Malta, this index is intercalibrated under the WFD, hence the assessment of condition in each coastal water body follows the requirements of this European Directive. **Slovenia** also follows the requirements of WFD to assess the condition of *Posidonia* beds.

As previously explained, some MSs indicate some of the variables to measure but do not specifically focus on the methodology to evaluate them. A common ground between all MSs is that they highlight the importance of expert judgement for defining the monitoring localities and for analysing the results obtained for the variables studied.

The key characteristics analysed for this habitat by the MSs, and the methodologies used, are summarised in Table 10.

Table 10. Methodologies used by Member States in the assessment of key characteristics of *Posidonia* beds

Key characteristics	Monitoring methods
Substrate type	Diving, visual observation.
Sediment type, quality and sedimentation rate	Diving, sampling, core, sediment traps, laboratory analysis.
Water (physicochemical parameters)	Multiparameter probe, Secchi disc, water sampling, laboratory analysis.
Presence of characteristic/ endangered species, indicator species and invasive species	Diving, visual observation, census, % coverage.
Meadow morphology and limits	Diving, visual observation, size of patches (patchy meadows), remote mapping.
Shoot density and coverage	Diving, visual observation, counting, estimation of % coverage, remote mapping (satellite, airborne imagery).
Shoot sampling and analysis	Diving, sampling, laboratory analysis.
Burial degree of rhizomes	Diving, visual observation, measurement.
Meadow dynamics (shoot / rhizome evolution, reproduction)	Diving, visual observation, measurement.
Impacts and disturbances	Diving, visual observation, satellite, airborne imagery and/or direct observation, mapping.
Habitat distribution/ fragmentation	Remote mapping (satellite, side-scan sonar, airborne and/or acoustic imagery), diving, visual observation.

As shown in Table 10, apart from in situ monitoring, additional methodologies are needed to analyse some of the key characteristics of habitat 1120*. One of these is laboratory analysis, which is used for water samples, sediment samples or even *Posidonia oceanica* samples. MSs also use this methodology to evaluate the environmental conditions in which the habitat develops and to obtain an in-depth analysis of species condition.

Table 10 also shows the use of remote monitoring and mapping methods to evaluate the distribution of the habitat and the meadow limits. Although these remote methods are listed in some of the methodologies reviewed, they are not described in detail. These methods depend on the availability of precise information and the technological capacity of the MSs.

2.7 Other relevant methodologies

Posidonia oceanica is used as a biological indicator in the monitoring programmes implemented by the Water Framework Directive and the Marine Strategy Framework Directive (Güreşen et al., 2020). Multiple investigation practices, protocols and indexes have been developed using *P. oceanica*, owing to its structural and functional features, providing information about the status of coastal ecosystems (Med-GIG, 2007). Some examples of these indexes and protocols are listed below:

- ***Posidonia oceanica* Rapid Easy Index (PREI)** (Golbert et al., 2009): a method used to assess the ecological status of seawater along French Mediterranean coasts. Guidelines from **Italy** adopt this index as the official national classification method (DM 260/2010). **Malta** also applies this index at specific monitoring stations under the WFD and MSFD (Environmental and Resources Authority, 2015).
- ***Posidonia oceanica* Multivariate Index (POMI)** (Romero et al., 2007): a multivariate index based on the structural and functional attributes of the *Posidonia oceanica* ecosystem. This index has been used for monitoring coastal waters in Catalonia (**Spain**) since 2005 and has been applied more recently in **Croatia** (Nikolić et al., 2009; UNEP-RAC/SPA, 2011; Mascaró et al., 2012). A modification of the POMI (modified POMI) that is more cost-effective has been developed for monitoring *P. oceanica* meadows in **Croatia** and has been tested and partially implemented in **Montenegro** (Guala et al., 2014).
- **Valencian conservation status index** (Fernández-Torquemada et al., 2008): a principal components analysis was used to classify the ecological status of *P. oceanica* at different localities in the Valencian Community (**Spain**) according to five classes as prescribed by the WFD. After evaluating multiple descriptors, an index was developed, which included descriptors that provided the most consistent information on conservation status as well as demonstrating a significant relationship with estimated pressures on the meadows.
- **Biotic Index based on *Posidonia oceanica* (BiPo)** (Lopez y Royo et al., 2010): developed using all *P. oceanica* monitoring data available in the western Mediterranean and based on a standard assessment of anthropogenic pressures, in accordance with the WFD. This index was developed to comply with WFD requirements, to be cost-effective and to be applied over the largest geographical extent possible.
- **Ecosystem-Based Quality Index (EBQI)** (Personnic et al., 2014): an index based on the overall functioning of the *P. oceanica* ecosystem, compliant with MSFD requirements and using its ecosystem-based approach.
- **INTERREG *Posidonia* Programme (POSID)** (Pergent, 2007): a protocol developed to establish monitoring programmes for *P. oceanica* in southern and eastern Mediterranean countries (Algeria, Tunisia, Libya and Turkey).

Lastly, beyond these protocols and indexes, new technologies in the near future may be used to develop new methodologies for studying and evaluating marine habitats such as *Posidonia* beds. As described in section 2.6, the use of new technologies makes it feasible to analyse and evaluate some aspects of habitats remotely. Remote sensing and mapping (i.e., using Copernicus or Sentinel-2) represent a new trend for studying the distribution and fragmentation of habitats and for detecting changes in environmental conditions of the marine environment, such as temperature and primary production. Though it may be a good method for studying marine habitats in the near future, it is still in development and must therefore be verified by field surveys. In the case of habitat 1120*, it may be necessary to continue to have trained personnel measure and evaluate some variables in the meadow; for example, shoot density and percentage coverage of living rhizome.

2.8 Conclusions

The distribution range of *Posidonia oceanica* makes it technically easier to monitor this habitat than other marine habitats. Thus, some of the variables used to evaluate *Posidonia* beds are cost-effective and feasible to measure. For this reason, together with *P. oceanica*'s sensitivity to changes in environmental conditions (making it an excellent indicator species) and the fact that it is an endemic species, *Posidonia* beds are well studied in European Mediterranean countries.

The review of methodologies used by the Member States for assessing and monitoring the condition of habitat 1120* *Posidonia* beds reveals some differences but also common aspects that deserve attention.

The level of detail in the description of the methodologies varied according to MSs. Some of the methodologies were basically a list of variables to measure, without much indication on how, where, when or what results could be obtained to determine good or bad condition of the habitat. All the methodologies reviewed remarked on the need for expert judgement for assessing the condition of the habitat, e.g. to define the variables and monitoring sites. This is especially clear in the case of the methodology used by France, a leading country in the study of *Posidonia oceanica*, with developed indexes to assess the condition of this habitat and ongoing long-term monitoring, which however requires expert judgement throughout the evaluation process.

Regarding the variables used to assess habitat condition, in the various methodologies reviewed it was observed that not all MSs carry out this assessment with the same degree of detail. Some Member States measure a set of variables that give a full spectrum of the habitat condition, categorising them as either "mandatory" or "supplementary" in order to facilitate the application of the methodology and provide a deeper analysis according to available resources (e.g. Spain).

Almost all the MSs (5 out of 6) considered in this review monitor abiotic variables, mainly using multiparametric probes and sampling both water and sediment. Some MSs consider these physicochemical characteristics as supporting parameters to analyse but not specifically as part of the habitat monitoring. Abiotic variables, as the chemical and physical conditions of water and sediment (e.g. quality of the sediment and water, type of substrate where the habitat develops) are present in all the methodologies reviewed.

All the MSs reviewed monitor some biotic characteristics of the habitat but with a different level of detail. The only characteristic monitored by all the MSs is the density of shoots present in the meadow. This variable is easy to measure and gives valuable information on the habitat condition, as the evolution of the shoots density in a meadow can reflect changes in the environmental conditions or the presence of other pressures or impacts. Some MSs also monitor and evaluate other characteristics that provide information on the evolution of the seagrass (e.g. % coverage, shoot sampling, growth/reproductive dynamics, etc.), the associated fauna (e.g., characteristic species, invasive species, etc.) and disturbances and impacts observed. Most of the biotic characteristics analysed are monitored through visual observation and in situ measurements (diving).

Three MSs address seascape characteristics in the methodologies reviewed. It is fundamental to monitor the changes in the area of the habitat in order to evaluate its evolution and detect potential impacts that may lead to its regression and loss. The methodology used to evaluate these characteristics depends on the area that needs to be covered, but it mainly involves remote sensing or, in the case of small areas/patches, in situ evaluation.

It is important to highlight that none of the methodologies used by the different MSs evaluates the density or taxonomic richness of epifauna and infauna, including meiofauna, within the biotic characteristics evaluated. Epifauna is evaluated by some MSs as a structural variable, by assessing the biomass of epiphytes from sampled shoots. The presence of meiofauna is very abundant in *Posidonia oceanica* meadows (>670 meiofaunal species have been documented), exceeding the combined richness of sessile macroflora and macrofauna (García-Gómez et al., 2022). This fauna has short life cycles, low dispersal, and high surface-to-volume ratios, being sensitive and providing early response to eutrophication, toxicity and physical disturbance, while macrofauna usually shows a delayed response, of months or even years (Balsamo et al., 2012; Zeppilli et al., 2015; Ingels et al., 2021; Kim et al., 2023). This is why it could be considered an additional recommended biotic characteristic to be analysed when monitoring *Posidonia* beds.

With regard to general monitoring and sampling methods, most of the methodologies reviewed propose the use of fixed monitoring stations for assessing habitat condition. Fixed stations make it easier to assess the environmental condition of the habitat at the mid- and long-term, allowing a comparison of the data series obtained for the variables measured. Most MSs also consider it important to establish truly representative monitoring stations that can give a full picture of the habitat and allow for an in-depth assessment of its condition. To select these monitoring sites and stations in each country, the MSs also agree on the need for expert judgement, as extensive local knowledge of the habitat distribution and environmental conditions present is fundamental for selecting representative sites.

In conclusion, the methodologies of most of the MSs for assessing the condition of *Posidonia* beds share common aspects, measuring different variables that should allow a detailed assessment of the habitat condition. However, not all the methodologies reviewed were descriptive or appeared to be already in use, which made it difficult to conduct a complete analysis of all the MSs. Nevertheless, there were good examples of the methodologies used by different MSs, such as Spain and Croatia, to assess the condition of habitat 1120*, that could provide a good starting point for harmonising the methodology used for assessing the environmental condition of *Posidonia* beds.

3 Guidance for the harmonisation of methodologies for assessment and monitoring of habitat condition

3.1 Selection of condition variables, metrics and measurement methods

Variables identified for assessing and monitoring habitat condition need to be robustly associated to the key characteristics and processes (functions) that determine habitat condition and must be sensitive to natural threats or human pressures that decrease favourable condition (Maes et al., 2023). A set of variables associated to all types of characteristics (abiotic physical and chemical, biotic compositional, structural and functional, landscape/seascape variables) should be measured.

Description of the condition variables, metrics and measurement methods need to be informed and clear so that they can be applied in all Member States.

The ecological characteristics, methodologies, variables and metrics used to investigate and assess the condition of habitat type 1120* are presented earlier in this report. A proposed list of essential and recommended condition variables is presented in Table 11.

- **Essential variables** describe essential characteristics of the habitat that reflect its quality and condition. These variables are selected on the basis of their instrumental relevance, validity and reliability, and should be assessed in all MSs following equivalent measurement procedures.
- **Recommended variables** are optional, additional, complementary variables that may be measured when relevant, to gain further insight on the habitat condition, e.g. according to contextual factors operating on habitats in the different MSs. These are complementary to the essential variables, can help improve the assessment and understanding or interpretation of the overall results; it is recommended that the MSs assess these variables if considered necessary or if the results obtained from other variables suggest to do so.

In addition to the essential and recommended condition variables described above, some **descriptive or contextual** variables are also proposed. These variables define environmental characteristics (e.g. climate, topography, lithology) that can influence the habitat condition, are useful for defining thresholds for the condition variables and interpret the results of the assessment, but are not used in the aggregation of variables to determine the condition of the habitat. There are contextual factors operating in the different MSs, including biogeographical gradients, which may determine the values of the variables characterizing the habitat condition as favourable and should be taken into consideration when defining the thresholds for the condition variables.

The **selection of variables** for assessing and monitoring habitat condition is based on the following main criteria:

- **Addressing all types of habitat characteristics**: abiotic (physical and chemical), biotic (compositional, structural and functional) and seascape characteristics should be measured; at least one variable per characteristic type should be included.
- **Proper description**: the selected variables, metrics and measurement methods should be easily interpretable and useful for the implementation of the assessment and monitoring of the habitat in all MSs.

- **Cost-effectiveness**: the variables and methods selected should be cost-effective in terms of data collection and processing, as far as possible should not require advanced knowledge or the use of highly specific equipment, and basic training should be enough for collecting the data.
- **Robustness**: the variables selected must clearly address the key characteristics and processes of the habitat for the proper assessment of its condition.
- **Harmonisation**: the final assessment of habitat condition, based on the variables selected and methods applied, after considering the contextual factors of each MS, should allow the comparability of results among the MSs and their aggregation at the EU scale.

Different variables have been selected for assessing and monitoring habitat type 1120* *Posidonia* beds (*Posidonion oceanicae*) considering the above-mentioned criteria and the review of the methodologies used by the EU MSs. Note that the variables presented below (and the parameters measured) have been selected to provide an overall assessment of the habitat condition in order to harmonise those methodologies.

Abiotic variables for physical state

- **Substrate type:** rock, pebble, gravel, sand, matte or mixed substrate.
- **Sediment type:** granulometry.
- **Water (physical parameters):** temperature and transparency/turbidity.

Abiotic variables for chemical state

- **Sediment quality**: total organic carbon (TOC) in the sediment.
- **Water (chemical parameters):** nutrient concentration, pH and salinity.

Biotic variables for compositional state

- **Presence of indicator and/or endangered species**: density of sea urchins (*Paracentrotus lividus*), sea cucumbers (*Holothuria* spp.) and presence of other seagrass or endangered species.
- **Presence of invasive species**: presence and abundance of non-indigenous species such as *Caulerpa* spp.
- **Density and diversity of meiofauna**: study of the biodiversity and density of individuals.
- **Diversity and abundance of characteristic fish species**: visual census in a 50m transect (Harmelin-Vivien et al., 1985) of the fish diversity and its relative abundance present in the meadow.

Biotic variables for structural state

- **Meadow limits:** evaluation of the lower limit of the meadow.
- **Meadow configuration**: type of meadow (continuous, patchy (fragmented) or mixed).
- **Coverage of living rhizomes/dead matte**: surface covered by living rhizomes and dead matte measured in defined monitoring stations.
- **Shoot density**: shoot density measured in defined monitoring stations.
- **Analysis of shoots:** morphometric, biochemical, biomass and lepidochronological parameters.
- **Burial degree of the rhizome:** distance (cm) between the sediment surface and the ligule of an external leaf of a shoot.

Biotic variables for functional state

- **Impacts and disturbances**: presence of pressures and impacts, visually evaluating its importance.
- **Meadow dynamics**: vertical rhizome growth and shoot dynamics (birth, growth, mortality).
- **Sedimentation rate:** evaluation of the sediment inputs.

Landscape/seascape variables

- **Habitat distribution/fragmentation:** evaluation of the area covered by the meadow and its degree of fragmentation of the meadow.

Most of the variables included in Table 11 are measured at the monitoring station. Some variables (e.g., meadow morphology, presence of impacts and disturbances, habitat distribution/fragmentation) could also be addressed on a larger scale, using remote sensing, and be measured at locality level. The results obtained should then be aggregated at a local and biogeographic region level, as explained in sections 3.3 and 3.4. A description of sampling methods and of the methodologies suggested to measure the essential variables can be found in section 3.5.

Table 11. Proposals for condition variables for assessing and monitoring habitat type 1120* (*Posidonia* beds)

Characteristics	Variables	Metrics	Application	Standardised measurement procedures
1. Abiotic characteristics				
1.1 Physical state characteristics				
Substrate type	- Substrate type.	Type/s in five classes (rock, pebble, gravel, sand, matte or mixed substrate (indicating which substrates are present).	Descriptive/ contextual	Visual observation by diving.
Sediment composition	- Granulometry.	Particle size and distribution.	Recommended	Diving, sampling with a hand core, laboratory analysis of the samples (granulometric analysis).
Water (physical parameters)	- Temperature. - Transparency/turbidity.	- Temperature: degrees Celsius. - Transparency/ turbidity: light attenuation coefficient (m-1).	Essential	1) In situ analysis using a multiparameter probe (CTD) for temperature and turbidity (if a turbidity sensor is available) or measuring temperature with a thermometer, and light attenuation with a Secchi disc. 2) Sampling water using a Niskin bottle, analysis of water turbidity at the laboratory.
1.2 Chemical state characteristics				
Sediment quality	- Total organic carbon (TOC)	- g/m^2 per day.	Essential	Diving, sampling with a hand core, laboratory analysis of the samples.
Water (chemical parameters)	- Nutrient concentration - pH - Salinity	- Nutrient concentration: phosphates and nitrates - pH: pH scale - Salinity: PSU	Essential	1) In situ analysis using a multiparameter probe (CTD) for pH and salinity. 2) Sampling water using a Niskin bottle, analysis of nutrients in the water at the laboratory
2. Biotic characteristics				
2.1 Compositional state characteristics				
Presence of indicator and/ or endangered species	- Presence of indicator species: sea urchins (*Paracentrotus lividus*), sea cucumbers (*Holothuria* spp.).	- Indicator species: density of individuals (ind/m^2). - Seagrass species: estimated % coverage.	Essential	Visual observation by diving, census. 1) Indicator species: density of individuals per m^2 in the monitoring station using quadrats of a known area (e.g., 40x40 cm). These can be randomly or

Characteristics	Variables	Metrics	Application	Standardised measurement procedures
	- Presence of other seagrass species (*Cymodocea nodosa* or *Zostera noltii*). - Presence of endangered species (especially *Pinna nobilis*, but also the presence of other species (e.g., *Cladocora caespitosa*).	- Endangered species: presence/absence in a known area.		systematically (using the measurements of shoot density and % coverage) placed over the station. 10-20 measurements per monitoring station. 2) Seagrass species: % coverage of substrate colonised by other seagrass species in 4 random 10 m linear transects in the monitoring station. 3) Endangered species: one method for evaluating its presence/absence is to establish a circular census area, with a 5-15 m radius (usually 10 m), around the central point of the monitoring station, and mark the living individuals.
Presence of invasive species	- Presence of non-indigenous species such as *Caulerpa* spp. - Abundance of non-indigenous species such as *Caulerpa* spp.	- Presence/absence. - Abundance (only if there is presence): surface occupied per m^2 or % coverage.	Essential	Visual observation by diving, census. 1) Presence/absence: visually exploring 20 m around the central point of the monitoring station to note the presence of invasive algal species. 2) Abundance: % present in the same quadrats used in the meadow to measure shoot density and % coverage of living rhizome.
Density and diversity of meiofauna	- Density of individuals. - Diversity of species.	- Density (ind/10 cm^2), - Taxonomic richness (S), Shannon–Wiener H.	Recommended	Diving, sampling 20x20cm quadrats (taking the surface substrate), taking a total of 3 replicate sediment samples between meadow patches. Taxonomic laboratory analysis.
Diversity and abundance of characteristic fish species	- Richness of fish species. - Number of individuals.	Taxonomic richness (S), Shannon–Wiener H.	Recommended	Diving, doing a visual census in a 50m transect in the meadow. Counting the number of individuals of each species found.
2.2 Structural state characteristics				
Meadow limits	-Evaluation of the lower limit of the meadow.	Geographical coordinates of the limit and depth (m).	Essential	1) Visual observation by diving. 2) Remote sensing (satellite, side-scan sonar, airborne and/or acoustic imagery). 3) Towed video/ROV. The limit may not necessarily be in the monitoring station.
Meadow configuration	- Type of meadow	Defined in three classes (large continuous meadow, patchy meadow or mixed meadow).	Descriptive/ contextual	1) Visual observation by diving. 2) Remote sensing (satellite, side-scan, airborne and/or acoustic imagery). 3) Towed video/ROV.

Characteristics	Variables	Metrics	Application	Standardised measurement procedures
Patch size (in patchy meadows)	- Area covered by control patches. - Study of its evolution (growth/regression).	- m^2 - Distance (m) between old and new patch limits.	Essential (only in patchy meadows)	Visual observation by diving, measuring surface and marking patch limits to evaluate its evolution.
Coverage of living rhizomes/dead matte	- Surface covered by living rhizomes and dead matte measured in defined monitoring stations.	% coverage.	Essential	Visual observation by diving.
Shoot density	- Shoot density measured in defined monitoring stations.	Shoots per m^2.	Essential	Visual observation by diving, counting.
Burial degree of rhizomes	- Distance between the sediment surface and the ligule of an external leaf of a shoot.	cm	Essential	Visual observation by diving, measuring.
Analysis of shoots	- Morphometric and biometric parameters. - Biochemical parameters. - Biomass parameters (Note* lepidochronological analysis is part of the methodology used by different MSs reviewed. It has not been included here but it could be added. This is a selection of parameters that have been considered to provide more information on habitat condition).	Different units/metrics per variable: - Morphometric and biometric parameters, such as: number of leaves; length / width of leaves (cm), % of necrosed area on leaves, etc. - Biochemical parameters, such as: C content; N content; isotopic signature of N, etc. - Biomass parameters: epiphyte load (mg (dry weight) of epiphytes/g (dry weight) of leaf).	Recommended	1) Diving. 2) Sampling shoots at each monitoring station. 3) Analysing the samples at the laboratory.
2.3 Functional state characteristics				
Impacts and disturbances	- Presence and effect of pressures and impacts, visually evaluating its importance.	- Effect of pressures or impacts in the % surface of the meadow (e.g., anchoring boats, mooring buoy fields installed over the meadow), infrastructures (e.g., pipelines, aquaculture facilities, harbour extensions, etc.), large-sized litter (e.g., lost fishing gear, sunken ships, illegal or abandoned moorings, etc.).	Essential	1) In situ evaluation: visual observation by diving in the different monitoring stations, visual rating in case of presence. 2) Remote sensing (satellite, side-scan, airborne and/or acoustic imagery).

Characteristics	Variables	Metrics	Application	Standardised measurement procedures
Sedimentation rate	- Sediment input.	cm^3	Recommended	Diving, sampling using sediment traps, laboratory analysis of the samples.
Meadow dynamics	- Vertical rhizome growth - Shoot dynamics (birth, growth, mortality).	- Rhizome growth (cm). - Shoot dynamics (relative number of shoots present in a fixed quadrant).	Recommended	Visual observation by diving, measurement.
3. Landscape/Seascape characteristics				
Habitat distribution and fragmentation	-Area covered by the meadow. -Degree of fragmentation of the meadow.	- m^2/ha -Continuous, fragmented (number of fragments).	Essential	1) Remote sensing (satellite, side-scan sonar, airborne and/or acoustic imagery). 2) A detailed mapping and updated cartography are essential for evaluating the evolution of the habitat.

3.2 Guidelines for the establishment of reference and threshold values, and obtaining condition indicators for the variables measured

The observed measurements of the condition variables need to be compared to reference values and critical thresholds, in order to assess the condition for each variable. A reference level is the value of a variable at the reference condition, against which it is meaningful to compare past, present or future measured values of the variable. The difference between the value of a variable and its reference level represents the distance to the reference condition.

Reference levels should be defined in a consistent manner across different variables within an ecosystem type, and for the same variable across different ecosystem types. This ensures that the derived indicators are compatible and comparable, and that their aggregation is ecologically meaningful (United Nations, 2021).

Reference levels are usually set with high and low levels reflecting the limits or endpoints of the range of a condition variable that can be used in re-scaling. For example, the high level may refer to a natural state and the low level may refer to a degraded state where ecosystem processes are below a threshold for maintaining function (Keith et al., 2013, in United Nations, 2021).

Establishing reference values and thresholds is therefore essential to determine whether habitats are in good condition or have become degraded. Reference values represent the desired state of an ecosystem, often reflecting intact or minimally disturbed conditions. These values serve as benchmarks for assessing habitat condition.

In the methodologies reviewed for habitat type 1120*, many MSs did not indicate specific threshold values for the variables measured to determine whether the habitat 1120* is in good or bad condition. Some MSs did an intercalibration process with the existing monitoring for other European Directives (e.g., WFD), aligning the classification for the different methodologies and using indexes and reference levels that were already in use.

Expert judgement was used to determine the habitat condition in MSs that did not provide specific threshold for it. It is important to remark this aspect, as the lack of threshold values does not mean that there is no monitoring or evaluation on the habitat condition in place.

These guidelines do not intend to provide specific rules or values for these thresholds, but to define the main criteria and guide on the establishment of such values that would help determining whether the habitat is in good condition or not.

In a review of approaches for setting reference conditions for assessing marine ecosystem quality, Borja et al. (2012) recommend that they should be defined/described with reference to:

1) Multiple sites with similar physical characteristics within an ecoregion or habitat type.
2) Ideally represent minimally impaired or undisturbed conditions (i.e. absence or minimal human pressure).
3) Provide an estimate of the variability in biological communities and habitat quality due to natural physical and climatic factors.

They identified four main approaches: crossing referencing pristine areas, hindcasting, modelling and best professional judgement.

Pristine areas: Reference values set against "pristine" areas could be developed with knowledge of either undisturbed habitats or habitats that are considered to be in good condition. However, for this to be credible, it would require comprehensive knowledge of the pressures and impacts on the different habitats, and the implications for their condition. Finding such locations is also likely to be problematic, especially as many examples of the habitats which are being assessed are adjacent to the coast or within territorial waters and therefore likely to have been subject to many pressures/impacts sometimes over significant periods of time.

Hindcasting: Using hindcasting to set reference levels requires cross reference to some historical reference condition. This may be a condition which is considered unimpacted (see above) or a set date (as with the Habitats Directive where 1994 is used as a baseline). Issues will arise around deciding when to set any baseline, the reliability and availability of historic data, and how to account for any natural oscillations in condition. For example, a habitat may not be in favourable condition in the selected baseline year, there may be a lack of sufficient data to inform decisions on the most appropriate baseline year, and global changes in recent decades could alter the former reference conditions making any comparisons with datasets from 50, or 100 years ago impossible.

Modelling: Modelling by extrapolating biological attributes can be used to summarise/simplify, visualize and explain actual or predicted situations e.g. the Driver-Pressure-State-Impact-Response (DPSIR) framework (OECD, 1993). There are, however, many considerations with developing and applying such models (Patricio et al., 2016). They include data availability, the level of confidence in the outcomes, how to scale up interpretations, for example from a site to a region, and how to assess cumulative impacts. There is an additional consideration that modelling approaches can be complex with less transparency and comprehensibility for stakeholders and policymakers.

Expert judgement: This is widely used when there is limited data but should ideally be underpinned by some clearly stated criteria and it has less transparency and comprehensibility for stakeholders.

The analysis carried out by Borja et al. (2012) (summarized in Table 12) considered that whilst using pristine or minimally impacted conditions was the best single method, others were also adequate when combined with expert judgement. Setting targets was seen as an alternative approach where none of the traditional reference conditions approaches were applicable, which implicitly indicates conditions where the indicator in question is not adversely affected or only slightly affected. Their conclusions, looking specifically at assessing benthic ecological status, were that a combination of methods in setting reference conditions is more adequate in obtaining final quality assessments related to the pressures on a habitat than one method alone.

Also relevant is the consideration that, regardless of the approach, there may be existing relevant thresholds and reference values set within legal obligations. For marine habitats this is the case at a European level, under the Marine Strategy Framework Directive (MSFD) and the Water Framework Directive (WFD).

Table 12. Evaluation of target and reference condition setting methods, regarding different issues (from Borja et al., 2012)

Methods / issues	Reference conditions			Expert judgement	Targets		
Main issues	**Pristine areas**	**Historical data**	**Modelling**	**Best professional judgement**	**Baseline set in the past**	**Current baseline**	**Directional/ trends target**
Legislation using/ proposing it	WFD, CWA	WFD, CWA, OSPAR	WFD, CWA	WFD, CWA	OSPAR	HD	OSPAR
Data needs	Moderate (2)	High (1)	High (1)	Low (3)	Moderate (2)	Moderate (2)	Moderate (2)
Scientific robustness	High (3)	Moderate (2)	Moderate/ high (2.5)	High (3)	High (3)	High (3)	High (3)
Confidence of the method	High (3)	Moderate (2)	Moderate/ high (2.5)	High (3)	Moderate (2)	High (3)	Moderate (2)
Applicability	High (3)	Low (1)	High (3)	High (3)	Moderate (2)	Moderate (2)	Moderate (2)
Practicality of the method within available time scales	High (3)	Moderate (2)	High (3)	High (3)	Moderate (2)	Moderate (2)	Moderate (2)
Transparency and comprehensibility	High (3)	High (3)	Low (1)	Low (1)	High (3)	High (3)	High (3)
Total scores	17	11	13	16	14	15	14

General guidance on setting environmental thresholds is included in the MSFD which requires that Good Environmental Status (GES) should be achieved in EU marine waters as described by eleven environmental Descriptors.

At the core of the GES assessment lies the need for threshold values which enable a quantitative assessment of environmental status for the indicators and elements used for each GES criterion. Principles and guidelines on how these thresholds should be set are specified in Article 4(1) of Commission Decision (EU) 2017/848 (European Union, 2017[2]) (Box 1).

[2] Commission Decision (EU) 2017/848 of 17 May 2017 laying down criteria and methodological standards on good environmental status of marine waters and specifications and standardised methods for monitoring and assessment, and repealing Decision 2010/477/EU) Art 13.

Box 1. Article 4 of Commission Decision (EU) 2017/848 (EU, 2017)

Article 4 - Setting of threshold values through Union, regional or subregional cooperation

1. Where Member States are required under this Decision to establish threshold values through Union, regional or subregional cooperation, those values shall:

(a) be part of the set of characteristics used by Member States in their determination of good environmental status;

(b) be consistent with Union legislation;

(c) where appropriate, distinguish the quality level that reflects the significance of an adverse effect for a criterion and be set in relation to a reference condition;

(d) be set at appropriate geographic scales of assessment to reflect the different biotic and abiotic characteristics of the regions, subregions and subdivisions;

(e) be set on the basis of the precautionary principle, reflecting the potential risks to the marine environment;

(f) be consistent across different criteria when they relate to the same ecosystem element;

(g) make use of best available science;

(h) be based on long time-series data, where available, to help determine the most appropriate value;

(i) reflect natural ecosystem dynamics, including predator-prey relationships and hydrological and climatic variation, also acknowledging that the ecosystem or parts thereof may recover, if deteriorated, to a state that reflects prevailing physiographic, geographic, climatic and biological conditions, rather than return to a specific state of the past;

(j) be consistent, where practical and appropriate, with relevant values set under regional institutional cooperation structures, including those agreed in the Regional Sea Conventions.

A review of the state of play with thresholds for MSFD criteria used by Member States, published in 2022, shows the progress but it also indicates that there is still some way to go before this is achieved for all eleven Descriptors (Vasilakopoulos et al., 2022). On the other hand, the WFD requires Member States to protect and where necessary restore water bodies in order to reach good status (chemical and ecological) and to prevent deterioration. Standards for priority substances and certain other pollutants are set out in the Environmental Quality Standards Directive (2008/105/EC)[3].

Habitat 1120* is evaluated under both MSFD and WFD. This is why a consistent approach, cross-referencing agreed thresholds for MSFD Descriptors and WFD thresholds, with those that are also relevant to assessing the condition of the structure and function of marine and coastal habitats covered by the Habitats Directive is clearly desirable. As mentioned before, some MSs already did an intercalibration process with the existing monitoring for other European Directives (e.g. WFD), aligning the classification for the different methodologies and using indexes and reference levels that were already in use.

The evaluation of the condition of the habitats is based on determining whether the variables used in the assessment indicate good or not good condition. Nevertheless, it is common practice to define more than two categories in the assessment of each variable, e.g. good,

[3] Directive 2008/105/EC of the European Parliament and of the Council of 16 December 2008 on environmental quality standards in the field of water policy, amending and subsequently repealing Council Directives 82/176/EEC, 83/513/EEC, 84/156/EEC, 84/491/EEC, 86/280/EEC and amending Directive 2000/60/EC of the European Parliament and of the Council

medium, bad, as observed in the analysis of methodologies used by the MSs. Different criteria are applied to attribute these condition categories according to the characteristics of each variable, for example, whether they are categorical (e.g. no alien species allowed), or quantitative variables which may obey to linear or non-linear relationships with the condition (Jakobsson et al., 2020). This assimilation of the values (quantitative or categorical) of the variables to the condition categories (i.e., good and not good, or good, medium and bad) would correspond to the scaling necessary to later evaluate them jointly, through aggregation procedures, as described in the following section. So, these condition variable categories can be translated to values, such as good=2, medium=1, bad=0. Alternatively, when quantitative values for the variables are available, they can be directly standardized to apply aggregation procedures.

In the assessment of habitat condition, each characteristic and associated variable is likely to involve the use of different measurement units. Owing to the different metrics and magnitudes applied to the variables, the values obtained from their measurement require some form of standardisation, e.g., by re-scaling, to build indicators combining different variables. The values obtained from the measurement of the variables are scaled according to their reference levels, thus normalised to a common scale and direction of change, and can then be combined to form a composite index or to obtain an overall result of the assessment using appropriate aggregation approaches (see further details in Section 3.3. on Aggregation).

Lastly, in the specific context of habitat 1120*, it is important to highlight the need of considering the key ecological characteristics of *Posidonia oceanica* when defining reference values. Some factors that should be taken into consideration are listed below:

- **Depth:** Neptune grass is mainly distributed at depths ranging from a few dozen centimetres down to 30 - 40 metres. It requires light for photosynthesis, so variables such as shoot density or percentage of coverage are conditioned by depth. Therefore, these variables should have different threshold values at each depth range. For this reason, some research groups take all measurements at a single depth to be able to establish a threshold (Lepareur, 2011) or have multiple threshold values at different depths for variables such as density or coverage (Guala et al., 2014).
- **Substrate type:** *Posidonia oceanica* is mainly found on sandy substrates, nevertheless, it can also be present on rock, pebble or gravel substrates. Depending on the substrate the threshold values should differ (Ministero dell'Ambiente e della tutela del Territorio e del Mare, 2017).
- **Water transparency:** Neptune grass grows better in clear waters. Threshold values should take into consideration the distance between meadows and rivers or streams.
- **Hydrodynamism:** sand-bottom swell areas have high sediment dynamics, which could affect the distribution of *P. oceanica* meadows. This should be taken into consideration when defining threshold values compared to areas with low swell and sediment dynamics.
- **Temperature:** Neptune grass can withstand a wide range of temperatures. However, global warming and the consequent increase in sea temperature could affect the survival and distribution of the species (Savva et al., 2018; Ontoria et al., 2019).
- **Salinity:** Neptune grass growth is very sensitive to environmental conditions, especially desalination. Hence, the threshold values adopted to measure the environmental state of this habitat should take into account its proximity to rivers, streams, and other freshwater inputs.

3.3 Guidelines for the aggregation of variables at the local level

Ecological assessments require the integration of physical, chemical, and biological quality parameters. The choice of the aggregation method of such partial assessments into an overall assessment has been widely discussed within the scientific community, since the methodology can have a considerable influence on the outcome of the assessment. Different approaches can be used to integrate the values of the measured variables to give an overall value that indicates the overall condition of habitat types at the local scale, i.e. the monitoring plot, station or site.

An appropriate aggregation method is crucial to categorising local-scale condition into good/not good. This is because the proportion of the habitat type in **good/not good condition** is the main information required for assessment of the structure and function of the habitat type at the biogeographical level.

3.3.1 Overview of aggregation methods

In a review of methods for aggregating and integrating information when assessing the status of marine ecosystems under the MSFD, focusing mostly on the descriptors related to biodiversity, Borja et al. (2014) discussed the advantages and disadvantages of several different approaches used to combine a number of variables into an overall assessment. A review and discussion of advantages and disadvantages of several different aggregation methods for marine biodiversity status assessments has also been carried out by Barnard & Strong (2014).

The main approaches are summarised below.

One-out, all out (OOAO)

The OOAO rule has been recommended for assessment of Ecological Status under the Water Framework Directive (CIS, 2003). The logic behind this is that a water body could not achieve good ecological status if any of the quality elements measured fail. This means that an OOAO approach using the “worst case” scenario for its assessment. This can be viewed as a rigorous precautionary approach. One criticism, however, is that it could lead to an underestimation of the true overall status.

A precautionary one-out, all-out approach is also used in the aggregation of the parameters used in the assessment of conservation status under the Habitats Directives and the IUCN Red List of Species and the IUCN Red List of Ecosystems.

The OOAO rule is a rigorous and conservative approach which follows the precautionary principle, and works well where all the necessary information is available. In the marine environment, where there may be significant data gaps it is important to clarify the extent to which such an approach may be “preliminary”, “partial” or “incomplete” due to lack of data.

Averaging approach

The averaging approach is the most commonly used method to aggregate indicators (Shin et al., 2012) and consists of simple calculations, using methods such as arithmetic average, hierarchical average, weighted average, median, sum, product or combinations of those rules, to come up with an overall assessment. Differential weighting applied to the various indicators can be used when calculating means or medians. An adequate basis for assigning weights is not always available and assigning weights often involves expert judgment: However, expert opinions applied in such a way can show important differences.

This approach needs a normalisation of the obtained data to be used.

Conditional rules

Conditional rules are an approach where indicators can be combined in different ways to generate an overall assessment, depending on specified criteria. For instance, it can be formulated in a way that requires that specific proportion of the variables to achieve good status or if a certain number of variables do not meet the threshold, the overall status fails.

Scoring or rating

In this method different scores are assigned to a particular status for a number of different elements, e.g. ranging from 1 to 5 for poor to good). The scores may then be summed to derive a total score which is then rated according to the number of elements taken into account. Different weights can be assigned to the various elements.

Multimetric indices to combine indicators

Within the WFD there are many examples of multimetric indices developed for different biological elements. Within the MSFD, the use of multimetric indices or multivariate techniques for integrating indicators of seafloor integrity have been recommended (Rice et al., 2010). Multimetric methods that are used to combine multiple parameters in one assessment may result in robust indicators, but ideally the various parameters should not be inter-correlated.

Multidimensional approaches

Multivariate methods, such as Discriminant Analysis or Factor Analysis combine parameters in a multi-dimensional space. Multivariate methods have the advantage of being more robust and less sensitive to correlation between indicators. However, interpretation is less intuitive than other methods, as information on individual indicators in each ecosystem is lost and links to management options are less obvious.

Decision tree

Decision trees provide the opportunity to apply different, specific, rules to combine individual assessments into an overall assessment. A decision tree allows implementing individual rules at each of its nodes and thus incorporates decisions at each step within the decision tree. The decision rules can be quantitative or qualitative as well as based on expert judgment. This gives room for a high degree of flexibility in reaching the final assessment.

Probabilistic approach

In some cases the results for each indicator may be uncertain due to several factors e.g., natural variation in the sampling sites, random variation in the samples, insufficient scientific understanding about what should be the reference value for good status, etc. If these uncertainties can be approximated, this gives rise to the possibility of taking this information into account when integrating the indicators. The more uncertain indicators will get less weight in the integrated assessment, while the more certain ones will be more reliable and hence get more weight.

High level integration

This approach, which includes the selection of an agreed reduced set of indicators and agreed weighting rules, could be considered a pragmatic compromise, reducing the risks associated with OOAO while still giving an overall assessment. An example of a high-level integration, where assessments for several ecosystem components are merged into a final assessment, is the HELCOM-HOLAS project (HELCOM, 2010).

As seen in Section 2.3, across EU Member States, the aggregation at local-scale assessments relies on integrating information from multiple variables, though specific approaches varying by country and habitat. Several MSs however apply a conditional rule, whereby a number of relevant variables measured must reach or pass the defined thresholds for good condition, or even the one-out, all-out rule, which requires that all the variables reach the threshold, for the overall habitat condition at the local scale to be considered good.

3.3.2 Recommendations for the aggregation of the measured variables to determine the habitat type condition at the local scale

A common aggregation method to integrate all essential and specific variables measured to assess the habitat should be applied consistently across the habitat range in the EU in order to obtain comparable results.

Considering the various approaches described above and with the aim of harmonising the assessment of marine habitat types at the local scale, we suggest a two-step approach, in which a first aggregation is carried out separately for each group of variables associated to abiotic, biotic and landscape characteristics, and then, the results of such partial assessments are then integrated into the overall local assessment of the habitat condition following a one-out , all-out rule, as described below but being clear about where data is limited or insufficient to make such an assessment (Figure 1).

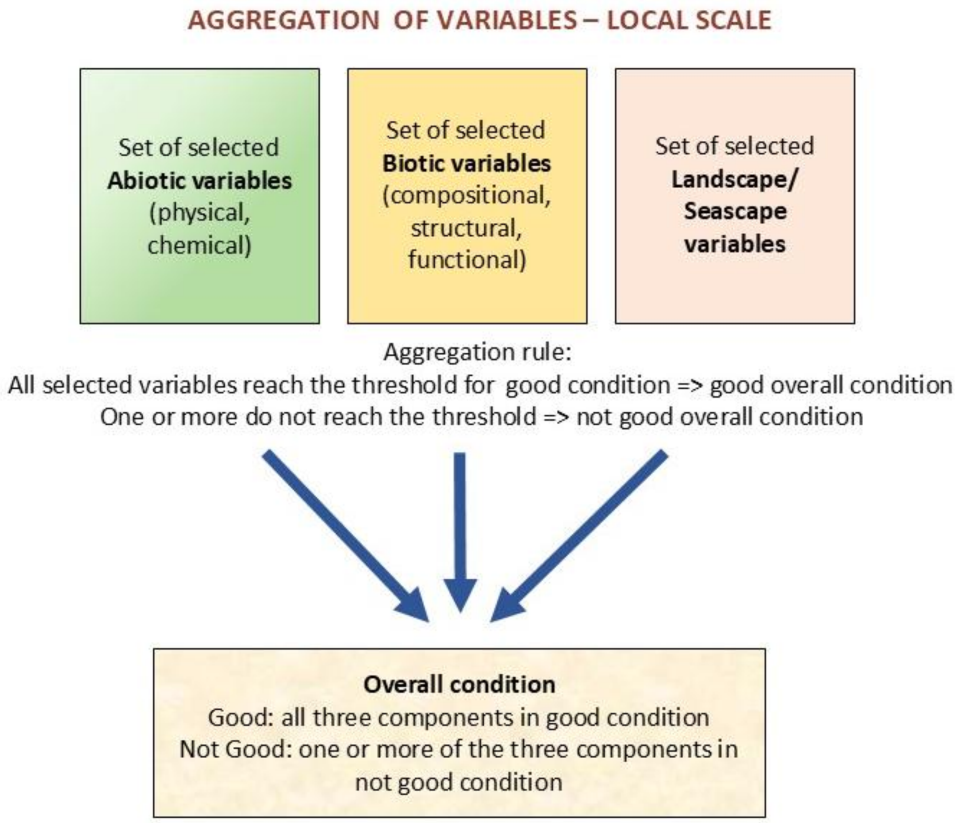
Step 1 – Aggregation of the variables measured in each group of characteristics

A first step would involve using a conditional rule. This would aggregate the variables for each group of characteristics whilst noting that a minimum set of essential variables in each group must reach/exceed the defined threshold for good condition. This would be done for each habitat component (abiotic, biotic, landscape). The selection of the set of variables that must reach the threshold is made considering their indicator value, i.e. their relative importance or relevance to determine whether the habitat is in good condition or not. These should be variables for which a clear threshold can be defined to distinguish good and not good condition. If any of those selected variables do not reach/exceed the minimum thresholds, the condition cannot be considered good for the corresponding component of the habitat (abiotic, biotic, landscape). If there are insufficient data on any particular variables to make such an assessment this should be noted.

Step 2 – Aggregation of the three groups of variables or habitat components

In a second step, the results achieved in each the three components or groups of characteristics (abiotic, biotic, landscape) would be aggregated following the “one-out, all-out” rule, which requires that all the three components have been assessed in good status for the overall condition of the habitat at the local scale to be considered good. If any of these components do not reach an overall good status, the condition of the habitat at the local scale cannot be considered good. Where there are data gaps on any of the habitat components (biotic, abiotic or landscape), a clarification should be provided to show that the assessment is incomplete or preliminary.

Figure 1. Scheme of the proposed aggregation of variables at the local level



3.4 Guidelines for aggregation at the biogeographical region scale

Habitat condition at the biogeographic level is evaluated aggregating the available information at the local scale for each MS. Currently, MSs follow the recommendations from the Art. 17 reporting guidelines for the period 2013-2018. Thus, the status of "structure and functions" parameter at the biogeographical scale for habitat 1120* *Posidonia* beds (*Posidonion oceanicae*) is considered:

- **Favourable**: if more than 90% of the *Posidonia oceanica* meadows assessed are considered as in "good" condition.
- **Unfavourable-bad**: if more than 25% of the *Posidonia oceanica* meadows assessed are reported as "not in good condition".

This rule highlights the importance of a sampling design that ensures sufficient representativeness of the total habitat area and diversity (see section 3.6 below).

3.5 Guidelines on general sampling methods and protocols

After reviewing the methodologies used by different MSs to evaluate habitat type 1120* *Posidonia* beds (*Posidonion oceanicae*), in situ methods are clearly the ones more used, although remote sensing monitoring methods have recently emerged as an interesting new approach. Specifically, the most widely used method to study and monitor this habitat is scuba diving, as its distribution in shallow coastal waters makes it very feasible for expert divers. It should be noted that videography is another method often used to monitor some variables such as the meadows' limits or their morphology, even though future research should be addressed to test the effectiveness of videography as additional method for assessing the conservation status of *Posidonia oceanica* meadows (Guala et al., 2014).

Therefore, the sampling recommendations and monitoring protocols for habitat 1120* showed next are mainly focused on in situ methodology by scuba diving at defined monitoring stations. The aggregation of data into different spatial units (e.g. biogeographical scale, Natura 2000 network, site level scale) has been detailed in sections 3.3. and 3.4.

3.5.1 Recommendations on sampling design and monitoring protocols

The proposed methodology to sample and monitor habitat type 1120* has been defined after reviewing the existing methodologies used by different MSs. Some of the variables described here are based on the methodology used in Spain (Díaz & Marbà, 2009) as it is well-defined and standardised.

Quantitative essential variables are suggested to be monitored once a year in threatened meadows and every two to five years in non-threatened meadows. **Descriptive variables** are suggested to be monitored the first time the monitoring stations are surveyed and whenever a change is observed on them. **Recommended variables** could also be assessed to get a better insight into the habitat condition. Regarding these variables, it is suggested to assess them at least every six years to have a general view of the habitat status to support the reporting required by Article 17 of the Habitats Directive.

Variables are recommended to be measured always at the same time of the year, from July to September, when the weather conditions are suitable (Díaz & Marbà, 2009). However, some scientists suggest that the monitoring of some variables should be preferably carried out when the leaves are shorter, enabling data to be easily collected for some variables as living rhizome coverage or shoot density (Guala et al., 2014). Variables such as the epiphytes attached on the leaves (epiphyte load) are analyzed from sampled shoots (which has not been considered an essential variable) and should be monitored in September, when *Posidonia oceanica* shoots achieve maximum biomass (Alcoverro et al., 1995), or October (Martínez-Crego et al., 2010), using the oldest leaf of each shoot (Cinelli et al., 1984) as it has the higher number of epiphytes.

Finally, the number of replicates to be carried out for the different quantitative variables at each monitoring station varies depending on the different methodologies reviewed. Local expert judgement should be considered in each MS when making this decision.

The methodologies suggested to measure the essential variables are described below.

- **Substrate type**

The seafloor of the monitoring station area is visually examined to determine the substrate nature. Substrate categories in which meadows could develop are: rock, pebble, gravel, sand, *Posidonia* "matte" or a mixed one.

- **Sediment quality**

The sediment of the monitoring station is sampled using a hand core and analysed at the laboratory to determine the total organic carbon (TOC) present.

- **Water (chemical parameters)**

The water chemical parameters, mainly salinity, can be measured by taking surface and depth water samples at the monitoring station. Later these samples are analysed in the laboratory to obtain the results.

A cost-efficient alternative would be to use a multiparameter probe (CTD). It would be deployed at each monitoring station to get surface and depth salinity data as well as additional parameters that could be also measured with this device.

- **Presence of indicator, endangered and/or other seagrass species**

The indicator species presence is monitored by counting individuals (usually sea urchins and sea cucumbers) using the same squares used to measure the meadow coverage. The 40x40cm squares can be randomly placed over the meadow at the monitoring station or use the same deployment of the squares when measuring the percentage of coverage or the shoot density (even though it can be used a 20x20cm square to measure shoot density) to do the counting of indicator species' individuals. Further information on how to conduct the sample monitoring can be found in Díaz & Marbà (2009).

The presence of endangered species such as *Pinna nobilis* can be evaluated by different methods (Hendricks et al., 2008; García-March, 2005). One example is presence/absence by doing a circular census of 5 to 15m meter radius (it is normally used a 10m radius) around the central point of the monitoring station, and marking living individuals. More information about this method can be found in García-March (2005). In case of finding other endangered species while doing the census (e.g. *Cladocora caespitosa*) it should be noted and the individual should be marked in order to monitor any subsequent changes.

Lastly, the presence of other seagrass species could be monitored by doing four random 10m linear transects within the monitoring station and estimating the percentage of substrate colonized by those species. Any other seagrass species present should be noted, along with the percentage of coverage and any changes monitored in the future. More information can be found in Guala et al. (2014).

- **Presence of invasive species**

In areas where there is a clear invasive species occurrence, an estimation of the percentage of coverage by the species should be undertaken while measuring the percentage of coverage (and even shoot density) of *Posidonia oceanica* using a 40x40cm square.

Moreover, the monitoring station area should be visually scanned looking for any invasive species in a 20m radius from the central point and sorted into presence/absence. Further information on how to conduct the monitoring of this variable can be found in Díaz & Marbà (2009).

- **Meadow lower limits**

The lower limit of a meadow may not be found in an established monitoring station. There are different methodologies that could be used to evaluate the lower limit of the meadow in situ (Díaz & Marbà, 2009; Pergent, 2007). One of those methodologies (Díaz & Marbà, 2009) is briefly explained below together with other methods, such as remote sensing: mapping the lower limit of habitat 1120* using side-scan, acoustic imagery, aerial or satellite precision orthophotography wherever it is possible).

In situ methods:

- **Diving**: in large meadows a variable number of measures are taken along a 100m transect following the depth limit. In smaller meadows, the whole limit can be monitored. In both cases, at each measurement divers note the existing depth, take the coordinates and mark the limit position of the meadow with permanent stakes to evaluate its evolution. It is also important to describe the type of limit observed, following the indicators described in Meinesz & Laurent (1978).
- **Towed video/ROV:** a towed video or a ROV can be used to define the lower limit of the meadow, as well as establishing its coordinates and depth.

Further information on how to conduct the monitoring of the lower limit can be found in Díaz & Marbà (2009).

- **Meadow configuration**

The area is visually scanned at the monitoring station. The meadow morphology is assessed regarding its continuity as well as the number of patches observed in fragmented meadows. Meadows are classified into:

- **Continuous**: no fragmentation is observed in the meadow.
- **Patchy**: fragmentation is observed in the meadow and no continuous patch is visible.
- **Mixed**: a continuous meadow with presence of some patches.

It would be also possible to use remote sensing to assess the meadow configuration, but it is a simple qualitative variable that could be visually characterized when evaluating other variables.

- **Patch size (only for patchy meadows)**

In patchy meadows, a variable number of patches should be selected and measured to calculate its area. Marking the limits of patches with stakes will make it possible to monitor their evolution and determine whether their area is increasing or decreasing. To be able to monitor the patches they should be geo-referenced and identified with a numbered stake (Díaz & Marbà, 2009).

- **Coverage of living rhizomes/dead matte**

Coverage is assessed carrying out a defined number of replicates (at least 3 replicates but the number should be defined by the expert judgement) at each monitoring station by estimating the percentage of area covered by *Posidonia oceanica* living rhizomes within a 40x40cm square, subdivided into 20x20cm squares. The 40x40cm square should be always placed over the meadow at the monitoring station (also when its morphology is patchy, specifically avoiding substrate to deploy the whole square over the meadow). There are multiple existing methodologies that define where the coverage should be measured, normally using fixed 10m transects that start at a defined coordinate (at the central point of the monitoring station). Other methodologies place the square randomly on the meadow, normally used at patchy meadows to avoid measuring areas where no meadow cover is present.

Further information on this methodology used can be found in Díaz & Marbà (2009).

- **Shoot density**

Shoot density is normally measured at the same 10m linear transects used to measure the percentage of coverage at the monitoring station. Five replicate measures of density are recommended along the transect where a high coverage is found (if possible, with a 100% of coverage), for both continuous and patchy meadows. Thus, 20x20cm squares are normally used to count all the shoots that fit inside the square to estimate its density in the monitoring station. 40x40cm squares can be also used, as the important thing is to know the area measured. It is recommended to use 20x20cm squares as it is easier to find in the meadow high coverage spots of 20x20cm than 40x40cm spots to measure the present density.

Further information on the methodology procedure can be found in Díaz & Marbà (2009).

- **Burial degree of rhizomes**

At the monitoring station a shoot is selected randomly to measure the distance between the sediment surface and the ligule of an external leaf. The measurement will be positive if the sediment is below the leaves and negative if the sediment is above the leaves. This measurement will be carried out 12 times at each monitoring station.

Further information on the methodology procedure can be found in Díaz & Marbà (2009).

- **Impacts and disturbances**

The monitoring station area is visually scanned looking for any impact and/or disturbance signal and sorted into presence/absence (modified from Lepareur, 2011; Guala et al., 2014). The types of pressures and impacts that could be found on a *Posidonia oceanica* meadow are, for example, anchoring pressure, mooring buoy fields installed on top of the meadow (e.g. concrete blocks, chains, ropes, etc.), presence of infrastructures (e.g. pipelines, aquaculture facilities, constructions like harbour ampliations, sewer outfalls, pollution inputs etc.) or presence of big-sized litter (e.g. lost fishing gear, sunken ships, illegal or abandoned moorings, etc.).

If present, it is important to visually evaluate any effects and rate them as high impact, medium impact, or low impact. Even though this is a subjective and preliminary rating and further evaluation will be needed, it gives an idea of the degree of impact that it causes on the habitat. This rating cannot be used as a threshold to evaluate this variable, and proper thresholds should be developed by the MSs.

Whenever it is possible, remote sensing images should be also used to assess large-scale impacts and disturbances and locate them beyond the monitoring station level.

- **Habitat distribution and fragmentation**

The habitat distribution should be monitored on a larger scale by mapping the meadow and having an actualized cartography of its distribution, including both its upper and lower limits.

Besides mapping the habitat distribution, it is fundamental to measure its area cover and to evaluate its degree of fragmentation. This information will make it possible to effectively monitor its evolution on a larger scale.

3.6 Selecting monitoring localities and sampling design

The identification and selection of localities for the assessment and monitoring of this habitat requires a systematic approach to ensure that the selected sites provide comprehensive and representative data. The selection of localities for sampling along with the sample size (number of plots) and power (statistical significance) are crucial to ensure the representativity of the results obtained in the assessment and monitoring of each habitat at the biogeographical scale.

Different approaches are recommended for the selection of monitoring localities:

- **Geospatial Analysis**: remote sensing techniques (e.g., acoustic surveys - side scan sonar, AGDS, MBES) as well as geographic information systems are essential tools to identify, analyse and integrate spatial data (e.g. extent, topography, and changes over time) and to identify areas of interest based on various criteria such as biodiversity, threats, and ecosystem services.
- **Field Surveys**: initial visits to potential sites are advisory to gather on-the-ground information about zonation patterns, accessibility, and logistics.
- **Review of existing data/knowledge:** reviews can help prioritise areas based on scientific knowledge.

Selecting a minimum number of localities for monitoring involves balancing several criteria to ensure comprehensive and effective coverage:

1. **Biogeographical or marine heterogeneity:** this habitat is present in all marine biogeographic regions but with different characteristics especially because of differences in tidal range and salinity profiles. Monitoring sites should be distributed across the entire area to represent the full range of ecological diversity and capture regional variations as well as habitat heterogeneity.
2. **Spatial Distribution:** monitoring sites must be distributed across the full geographical range of the habitat (subject to accessibility constraints) to avoid geographical bias and to capture regional variations and ensuring they represent a significant proportion of the habitat's area.
3. **Statistical Criterion**: It would be advisable to ensure that the number of sites is statistically sufficient to detect changes and trends with desired confidence levels (e.g., 95%). Multiple sites within similar ecological contexts should be included for data reliability and robustness.
4. **Existing data and monitoring sites:** Due to potential limitations in surface area and/or budget, previous research can help determine a more realistic number of monitoring locations. Making use of existing monitoring sites with historical data can also strengthen the understanding of long-term trends and changes in habitat condition. Such sites provide valuable baselines for comparison and support robust trend analyses over time.
5. **Degree of conservation and exposure to threat levels:** Monitoring locations should include both protected and high-risk areas experiencing significant threats. The selection should include areas that show different degrees of conservation or degradation, in order to capture the existing variation in the habitat condition across its range. This requires including localities representing well-conserved habitat areas, with minimal human impact, as well as areas subjected to degradation and different pressures and threats. To capture the range of pressures affecting the habitat, localities should be selected across a spectrum of threat levels, from low to high and considering different sources of threats, such as water quality, disturbance/accessibility and resource extraction.
6. **Presence inside and outside Natura 2000 sites:** The assessment and monitoring of habitats conservation status must be done both inside and outside Natura 2000 sites, which requires selecting localities – and an appropriate number of sampling stations/transects – that reflect the proportion to the habitat’s distribution within and outside the Natura 2000 network.

7. **Accessibility and practicality:** Monitoring localities should be accessible for regular visits, taking into account logistical factors and ease of access. Practical considerations also include the safety of field personnel and the feasibility of transporting equipment to and from the site.

Once the sampling localities have been identified for each habitat type, the minimum number of sampling stations in each locality and across the biogeographical region must be calculated in order to balance the sampling effort with representative data.

The **size of the sample** influences two statistical properties: 1) the precision of estimates and 2) the power of the assessment to draw conclusions. The number of sampling stations must be **statistically sufficient** to be able to detect changes and trends with desired confidence levels. Appropriate statistical methods should be used to determine an adequate sample size.

Considering the heterogeneity of the habitat type, it is highly recommended to consult with a sampling statistician regarding the sample size, i.e. the minimum number of sampling stations/transects required to ensure representativity and statistical significance.

Some key elements to ensure a proper representation of the habitat condition in the sample are summarised below.

Key elements for statistical representation

Sample size and distribution:

- The number of localities/transects etc. should be sufficient to provide a statistically robust sample size. This ensures that the data collected can be generalized to the entire habitat type within the region.
- Statistical methods such as stratified random sampling are often used to ensure that all habitat subtypes and environmental gradients are adequately represented.

Sampling design:

- Within each sampling area or locality, multiple plots are established to collect detailed data on benthos, infauna, mobile species and other ecological indicators. The distribution and number of sampling stations depend on the variability and size of the habitat patch. Sampling areas (plots, transects) are laid out considering the existing main ecological gradients, e.g., exposure to waves/currents/tides, depth, sediment characteristics.

Replication and randomization:

- Replication of sampling units within each locality and randomization of sampling plots location help to reduce bias and increase the reliability of the data.
- Randomized plot locations ensure that the sampling captures the natural variability within the habitat.

3.7 Use of available data sources, open data bases, new technologies and modelling

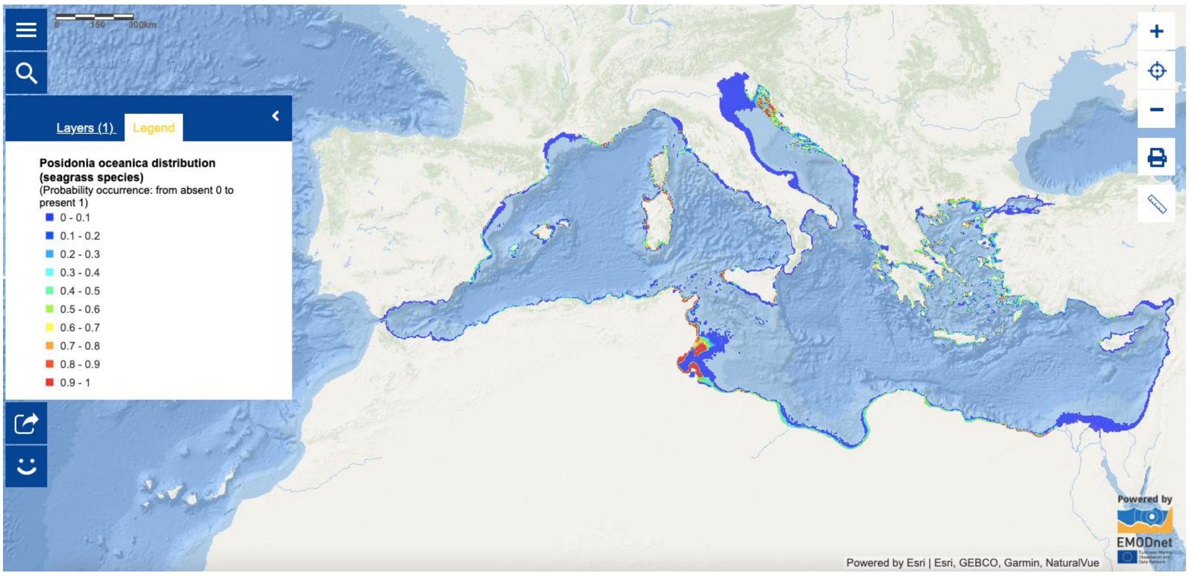
Currently, there are few open data sources that provide data on habitat 1120* *Posidonia* beds (*Posidonion oceanicae*). However, some of the information available (provided by the European Commission) gives a good idea on the importance of the species distribution along the Mediterranean coastline.

The European Marine Observation and Data Network (EMODnet) of the European Commission provides two useful tools for studying this habitat. The first is the EMODnet Map Viewer[4], which includes the current known extent and distribution of seagrass meadows in European waters, among multiple layers, collated by EMODnet Seabed Habitats (Figure 2). The second is the European Atlas of the Seas[5], which also provides a distribution map of *Posidonia oceanica* meadows, although it only shows the probability of occurrence of this habitat along the Mediterranean coastline. Both these data sources could provide a useful starting point for Member States who do not have adequate habitat mapping.

Another European Commission data source useful for studying this habitat is Copernicus[6], the Earth Observation component of the European Union's Space Programme. The data collected includes information on marine environment variables such as temperature, river inputs and chlorophyl, among others. This could be a useful tool during the process of selecting localities.

Finally, new remote sensing technologies have recently emerged as an alternative for larger-scale studies. Moreover, these technologies are considered a suitable tool for mapping and monitoring *Posidonia oceanica* meadows owing to their high precision, statistical power and cost-effectiveness (Puhr et al., 2014). As a result, more studies involving such resources are being carried out lately (Lipej et al., 2022), although it still needs time to become a recurrent methodology in studies. However, field verifications will always be necessary to study this habitat (Díaz & Marbà, 2009).

Figure 2. *Posidonia oceanica* distribution (probability of occurrence)



Source: EMODnet Map Viewer

[4] https://emodnet.ec.europa.eu/geoviewer/#!/
[5] https://ec.europa.eu/maritimeaffairs/atlas/maritime_atlas/
[6] https://data.marine.copernicus.eu/products

4 Guidelines to assess fragmentation at appropriate scales

Posidonia oceanica beds are suffering a general regression in the Mediterranean Sea. As mentioned in the first section of these guidelines, this regression has been estimated to be around 34% of the area covered by the habitat in the last 50 years (Telesca et al., 2015). The regression and fragmentation of *Posidonia* beds are affecting their normal functioning, thus threatening their ecological integrity and the biodiversity they support.

Human activities and natural events both play a significant part in causing the fragmentation of *Posidonia* beds. Other anthropogenic drivers (e.g., climate change) also cause great concern as they also condition the severity and frequency of natural events. Human-induced pressures on *P. oceanica* meadows are multiple, and include the following: coastal development (Holon et al., 2015), including urbanisation and infrastructure-building projects along the coastline (e.g., ports, marinas, coastal defences, pontoons, submarine pipelines); nautical sector growth (Carreño & Lloret, 2021), causing important mechanical impacts on meadows, such as anchoring (Pergent-Martini et al., 2022) and mooring; runoff from agricultural and urban areas (Micheli et al., 2013), causing an input of pollutants and wastewater (Boudouresque et al., 2012) into coastal environments, affecting water quality; dredging and illegal bottom trawling (Kiparissis et al., 2011), physically damaging the seafloor and the seagrass beds, creating gaps and reducing overall habitat connectivity; global warming, which is a major threat for the plant due to increases in water temperature (Litsi-Mizan et al., 2023). With regard to pressures caused by natural processes, storms and severe weather events can uproot shoots and rhizomes, creating gaps in the meadows and leading to fragmentation. Natural grazing by herbivores can also contribute to fragmentation, especially when herbivore populations are high, leading to localised areas of seagrass defoliation (Ruiz et al., 2001).

Some of the main effects of fragmentation on the condition of these seagrass meadows are described below:

- **Alteration of meadow structure and density:** fragmentation transforms continuous *Posidonia oceanica* meadows into patchier landscapes, resulting in bare spaces within the meadow, disrupting the continuous habitat structure. This change can influence the density of Neptune grass shoots, particularly at the edges of patches (Barcelona et al., 2021). The process of recovery is difficult as it is a slow-growing species (Hemminga & Duarte, 2000), making it hard for a meadow to overcome an impact and regenerate effectively.
- **Altered hydrodynamics and sediment dynamics:** the fragmentation of *Posidonia* beds modifies local hydrodynamics, affecting sediment dynamics and ecological interactions. Seagrass beds play a critical role in modifying hydrodynamics, which has a direct effect on sediment distribution and coastal protection. Fragmented areas can lead to changes in sediment particle sizes, with smaller particles being more prevalent near bare soil than in the meadow. This alteration can lead to changes in sedimentation patterns, where the distribution and typology of sediment differ between fragmented and continuous meadows (Serra et al., 2020).
- **Biodiversity and habitat:** the fragmentation of meadows impacts the distribution and diversity of associated marine life. The diversity and abundance of species are more likely to decrease in fragmented areas than in more continuous habitats. For instance, fish assemblages and other organisms are influenced by the fragmentation patterns, with habitat quality declining as meadows become more fragmented (Vega Fernadez et al., 2005).

To properly evaluate *Posidonia* bed fragmentation, long-term monitoring of specific meadows is needed in order to reveal trends over time associated with its regression and recovery processes. This approach can help identify critical thresholds beyond which recovery becomes difficult, providing essential data for conservation strategies. It is also fundamental to have a detailed and updated mapping/cartography of the habitat to be able to evaluate its general distribution and evolution over time.

Specific monitoring of habitat fragmentation would be helpful for completing the information obtained on the structural features of meadows (e.g., coverage, shoot density). For instance, how meadow cover varies across space is as important as the mean cover obtained, as spatial autocorrelation and autocovariance are measures of the spatial scale at which meadow cover varies, allowing an estimate of the degree and scale of fragmentation. It may also provide critical information concerning identity and frequency of anthropogenic processes causing meadow disturbance (Guala et al., 2014).

Monitoring and evaluating the distribution and fragmentation of habitat type 1120* *Posidonia* beds is of major importance for fully assessing the condition of this habitat.

5 Next steps to address future needs

After reviewing the methodologies used by the MSs for assessing and monitoring the condition of habitat type 1120* *Posidonia* beds and with a view to harmonise these methodologies, there are some aspects that should be further addressed in the future:

- It would be useful if the established methodologies of each MS for assessing habitat condition had a similar level of detail. This would also allow the MSs to share information and increase their knowledge about the habitat condition at a Mediterranean level.
- Testing the variables and methods proposed in these guidelines in all the Member States would allow evaluating their feasibility and refine or adjust them to different contexts as required.
- Appropriate reference values and thresholds should be defined for opposed variables to use for assessing habitat condition. It would be good to describe and clearly specify how the thresholds are established. This would help improve the comparability and harmonisation of methodologies in the future and to understand the process for assessing and determining the condition of this habitat in each MS.
- Appropriate methods for selecting the monitoring localities and sampling stations should be applied in all the MSs (this information is lacking in some of the methodologies reviewed). Local expert judgement will always provide a necessary support for this task but some relevant aspects recommended in these guidelines should be considered in this regard.
- Every MS should have a detailed, updated map of the *Posidonia oceanica* meadows along their coastline (some do not have it), which would serve as a key tool for effectively monitoring the distribution and evolution of the habitat.
- Further research on habitat mapping and condition monitoring using remote sensing methods would be useful for developing cost-effective methodologies and facilitating their use by all MSs.
- Further research and detailed monitoring of the effects of fragmentation and climate change on this sensitive habitat should be promoted.

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