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Agenda Item 3: Development of EcAp Ecological Objectives

3.2. Update regarding the development of the IMAP Ecological Objective 4 on Marine food webs under the Barcelona Convention

Desk review on the available information on marine food webs in the mediterranean.

SPA/RAC
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1. Introduction

1. At the 19th Meeting of the Contracting Parties to the Barcelona Convention (COP19, Athens 2016), the Integrated Monitoring and Assessment Programme of the Mediterranean Sea (IMAP) was adopted (Decision IG.22/7).
2. The ecological objective 4 (EO4) on marine food webs ("Alterations to components of marine food webs caused by resource extraction or human-induced environmental changes do not have long-term adverse effects on food web dynamics and related viability") was not included in the IMAP. Proposals of indicators, good environmental status (GES) description and related targets of EO4 were discussed in the early stage of the Ecosystem Approach (EcAp) process implementation. Lack of data and knowledge gaps were recognized on marine food webs in the Mediterranean Sea.
3. In order to be in synergy with the EU Marine Strategy Framework Directive (MSFD) in terms of monitoring of the ecosystem components and assessment of GES in the Mediterranean Sea, the need of a full set of common indicators related to marine food webs was ascertained.

4. This desk review study has the purpose to inventory data sources, best practices and methodologies for the monitoring and assessment of marine food webs in the Mediterranean in view of the development of IMAP EO4 in the framework of the EcAp process of the Barcelona Convention.

5. This desk review is composed of the following sections:
- scientific publications;
- existing and potential data sources;
- methodologies for monitoring and assessment (under the MSFD and other EU sea conventions, i.e. OSPAR, HELCOM);
- relevant ongoing/concluded initiatives/projects at regional, sub-regional or national levels;
- regional/national institutions or key persons/experts working on monitoring and assessment of food webs in the Mediterranean;
- knowledge gaps.

2. Scientific publications

6. A huge number of scientific publications (both academic and grey literature) exists about the application of methods for the study of marine food webs in the Mediterranean. The topic has been extensively explored in the last few decades, with special reference to the diet of single species and, to a much lesser extent, to the analysis of whole food webs. The publications deemed useful to provide a knowledge base on the assessment and monitoring of Mediterranean food webs is provided in the following chapters, in particular Chapter 4, and listed at the end of this review.

3. Existing and potential data sources

7. Data sources useful to the development of indicators and methods aimed at the monitoring of Mediterranean food webs albeit scarce, encompass a large number of species.
8. Stergiou & Karpouzi (2002) considered 146 Mediterranean species and provided (i) the bibliographic sources for their feeding habits based on stomach content analysis, (ii) the contribution of the main prey to their diet, and (iii) their estimated trophic level.
9. Karachle & Stergiou (2017) increased the previous data set providing stomach contents data (including main prey), trophic guild and trophic level for 204 Mediterranean species.

10. De Lope Arias et al. (2016) estimated the trophic level of eleven coastal fish species sampled in and around Mediterranean marine protected areas.
11. Thompson et al. (2020) carried out a meta-analysis on a huge data set from NE Atlantic and provided the feeding guild information for 73 fish species.
12. Silva et al. (2022) produced a georeferenced database (MesopTroph) of diet, trophic markers, and energy content of mesopelagic and other marine taxa encompassing 498 predator taxa from the NW and NE Atlantic and the Mediterranean. The following seven data sets can be directly accessed, visualized and downloaded: stomach contents, stable isotopes, major and trace elements, energy density, fatty acid markers, trophic positions and estimates of preys to predators' diet.
13. Daniela Berto and colleagues (ISPRA, Italy) are preparing a regional database with stable isotopes values that will be available for consultation at some point in the future (Berto, personal communication).
14. The ICES stomach data portal contains historical and new data from various projects and scientific surveys conducted in the NE Atlantic.
15. The European marine modelling resources (MMF-BLUE2) data portal provides access to 12 datasets on biological and environmental parameters that can be used in modelling exercises.

4. Methodologies for monitoring and assessment

16. An array of methodologies and approaches exists aimed at studying marine food webs (Kytinou et al. 2020). Models and indicators can be used to analyze food web structures also in order to assess the environmental status of marine ecosystems (MSFD Descriptor 4) and to attain the objectives of EU conventions. Most of them were developed and applied in NE Atlantic and Baltic ecosystems, but a number of original contributions as well as applications from the Mediterranean exists.
17. This chapter features the basic methods that are the foundations of trophic studies, as well as the main models and indicators that can be used to assess and monitor marine food webs.

4.1 BASIC METHODS

- STOMACH CONTENT ANALYSIS

18. For decades, the only method used for the study of fish diets has been based on visual stomach content analysis. Still widely used, this method gives a snapshot representation of the diet in a species; appropriate temporal (e.g., seasonal) sampling encompassing different life stages gives a reliable picture of the feeding habits of a fish population. The main advantage of this method is the provision of the whole spectrum of food consumed by a species. The main disadvantage is the need for high-level taxonomic expertise and the vast amount of man-hours required in the lab.
19. Hyslop (1980) reviewed stomach content analysis indexes providing a major reference for all future studies. Among the successive reviews of the same type, Mahesh et al. (2018) gave a rather comprehensive and well-done contribution. Cortés (1997), Baker et al. (2014) and Buckland et al. (2017) offered methodological insight to the diet analysis in cartilaginous and bony fishes.
20. Stomach content analysis has been compared to other methods (namely, DNA barcoding and stable isotopes analysis) in an attempt of identifying the best approach to obtain the highest quantity and quality of information in dietary studies according to the research objectives and the type of material available (Vinson & Budy 2011, Matley et al. 2018, Amundsen & Sanchez-Hernandez 2019).
21. Stomach content data can also be used to assess resources partitioning and dietary niche overlap among species (Wootton 1991, Krebs in prep.).

- METABARCODING

22. Another method that became available and started to be used since the late 1990's is the molecular analysis of food remains in the stomach or the digestive tract, in particular through the application of metabarcoding (Symondson 2002, King et al. 2008, Kress et al. 2015, Traugott et al. 2021, Gostel & Kress 2022). This method can also be applied with non-invasive techniques like e.g., on faeces or on regurgitated stomach content, although such techniques are hardly applicable in marine animals. Valentini et al. (2009) proposed a new method based on the trnL approach. Although metabarcoding has the potential of identifying ideally all species in a stomach content, it does not provide any information on their abundance, weight or size.

23. The relative pros and cons of molecular vs. visual analysis with a comparison of the two techniques were analyzed and discussed by (Soininen et al. 2009, Berry et al. 2015, Maes et al. 2022, Gul et al. 2023).

- STABLE ISOTOPES ANALYSIS

24. Visual and molecular analysis of stomach contents give a picture of consumers' diet at a given time and space, and need appropriate - and expensive - temporal and spatial sampling to offer a thorough information on diets. On the contrary, the analysis of carbon (δ13C) and nitrogen (δ15N) stable isotopes gives a dietary information over time that integrates the assimilation of energy through all the different trophic pathways leading to an individual consumer (Schmidt et al. 2007, Eglite et al. 2023). More specifically, the δ15N ratio can be used to estimate the trophic position (or trophic level) of a consumer in the food web, while the δ13C ratio can elucidate the ultimate sources of carbon for an organism (Wada et al. 1991, Post 2002, Fry 2006, Jennings et al. 2008). Post (2002) discussed also the critical issues related to the selection of an appropriate baseline.

25. The topic of sample treatment and process standardization has been tackled by Jacob et al. (2005), Carabel et al. (2006) and González-Bergonzoni et al. (2015). Boecklen et al. (2011) besides reviewing the general topic of stable isotope analysis, discussed thoroughly the limitations and uncertainties of the method, mainly due to the numerous possible sources of variation. A more accurate estimate of δ15N has been offered by Hussey et al. (2014).

26. C and N stable isotopes provide the input data in a number of models that can be used to assess and analyze food webs (Layman et al. 2012) (see Chapter 4.3).

27. The analysis of the sulfur stable isotope δ34S (Fry 2006, Mancinelli & Vizzini 2015) has been much less utilized in food web studies, although it can be successfully employed to distinguish the contribution of different producers to aquatic food webs (Connolly et al. 2004).

- FATTY ACIDS

28. Fatty acids tend to be stored in specific departments of consumers' body and can be used as trophic tracers to ascertain the origin of ingested food (Iverson 2009, Graeve & Greenacre 2020, Burian et al. 2020). Although they are more easily employed in pelagic food webs, they may be applied with some limitations also in the benthic environment (Kelly & Scheibling 2012). Kattner et al. (2007) discussed the role of lipids in the assessment of the impact of climate change at high latitudes.

4.2 INDICATORS

29. Among the above-listed basic methods for the analysis of diets in marine organisms, stable isotopes (especially of C and N) have had the highest success in providing simple (or univariate) indicators of the trophic status of marine communities as well as input to more complex models.

30. Gascuel et al. (2005) proposed the Trophic Spectrum of the total consumer biomass - or alternatively the trophic spectrum of the total commercial catch or of the abundance - as an indicator of the trophic structure in a fisheries context.

31. The Mean Trophic Level (MTL) or Mean Trophic Index (MTI) is based on catch (or landings) data and on the trophic level (TL) of each species caught, and indicates the mean trophic position of a species assemblage in the food web (Pauly et al. 1998). Since it is supposed to decrease with increased fishing mortality of consumers high in the food chain (i.e., high TL species) according to the “Fishing down the food web” principle (Pauly et al. 1998), it has been generally used to assess the ecological status of a fishery. As such it can also be considered a general indicator of the status of a food web, although it is prone to misinterpretation (Hornborg et al. 2013). Fey-Hofstede & Meesters (2007), Branch et al. (2010) and Stergiou & Tsikliras (2011) highlighted issues and limitations of this index.

32. Rountos et al. (2015) proposed the mean trophic level of predator consumption (mTLQ) and used it along with MTL using data from forty Ecopath food web models.

33. A set of indicators of food web ecological status have been identified for Descriptor 4 (D4 - Food webs) in the MSFD[1]:

- Performance of key predator species using their production per unit biomass (productivity);
- Large fish (by weight);
- Abundance trends of functionally important selected groups/species, such as:
    - groups with fast turnover rates (e.g. phytoplankton, zooplankton, jellyfish, bivalve molluscs, short-living pelagic fish) that will respond quickly to ecosystem change and are useful as early warning indicators,
    - groups/species that are targeted by human activities or that are indirectly affected by them (in particular, by-catch and discards),
    - habitat-defining groups/species,
    - groups/species at the top of the food web,
    - long-distance anadromous and catadromous migrating species,
    - groups/species that are tightly linked to specific groups/species at another trophic level.

34. Tam et al. (2017) analyzed over 60 potential food web indicators to extract a final list of 9 operational indicators:

- Total biomass of small fish
- Biomass of trophic guilds
- Primary production required to support fishery (PPR)
- Primary production required to support fishery
- Zooplankton spatial distribution and total biomass
- Mean trophic level of catch (MTL)
- Marine trophic index of the community
- Mean trophic level of the community
- Mean trophic links per species.

35. Safi et al. (2019) used Ecological Network Analysis (ENA) to propose a list of indicators drawn from the ecological literature that can be used in food web assessments:

- Detritivory over Herbivory ratio (D/H)
- Connectance Index (CI)
- Transfer Efficiency over trophic levels (TE)
- System Omnivory Index (SOI)
- Finn's Cycling Index (FCI)
- Relative Redundancy (R/DC)
- Average Mutual Information (AMI)
- Interaction Strength (IS).

36. Thompson et al. (2020) proposed a single indicator based on feeding guild assessment and explicitly suggested its use as an assessment tool in both MSFD and OSPAR frameworks.

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[1] Commission Decision 2010/477/EU of 1 September 2010 on criteria and methodological standards on good environmental status of marine waters (repealed and replaced by Comm. Dec. 2017/848/EU).

37. Machado et al. (2021b) proposed four food web indicators:
- mean trophic level (MTL)
- mean trophic level with cut-offs ($MTL\_{3.25}$ and MTL_{4})
- large fish indicator (LFI)
- mean abundance across trophic guild (MATG).

4.3 MODELS

38. Ecological models are one of the strongest approaches for predicting and understanding the consequences of anthropogenic and climate-driven changes in the natural environment (Piroddi et al. 2015). Plaganyi (2007) reviewed thoroughly the main models that could be used in the frame of ecosystem-based fisheries management - including the then available food web models. Several models have been developed with the aim of assessing and monitoring marine food webs either by quantitatively characterizing the system or by simulating different scenarios dominated by varying natural or anthropogenic pressures. Most models use C and N stable isotopes as input data (see Boecklen et al. (2011) for the limitations due to numerous sources of variation in isotopic signatures of organisms, and McCormack et al. (2019) for a comparison of the output from stable isotope analysis and Ecopath models).

39. Here follows a list of stable isotope-based models available in the literature (see Boecklen et al. (2011) and Layman et al. (2012) for detailed, in-depth reviews).
- Linear mixing models (one isotope, two sources, e.g. Raikow & Hamilton (2001); two isotopes, three sources, e.g. Kwak & Zedler (1997)).
- End-member models (one isotope, two sources, e.g. Forsberg et al. (1993)).
- Euclidean-distance models (two isotopes, three or more sources, e.g. Kline et al. (1993), Phillips & Gregg (2001)).
- SOURCE and STEP linear mixing models (Lubetkin & Simenstad 2004).
- Linear programming model (Bugalho et al. 2008).
- IsoSource mixing model (all possible contributions of each source: Phillips & Gregg (2003)).
- Moore-Penrose pseudoinverse model (M-P) (Hall-Aspland et al. 2005a, 2005b)
- Bayesian mixing models (MixSIR (Moore & Semmens 2008), SIAR (Parnell et al. 2010), FRUITS (Fernandes et al. 2014).
- MixSIAR (Stock et al. 2018, García-Seoane et al. 2023), which represents an improvement over its predecessors, MixSIR and SIAR.
- Spatial correlation model (Melville & Connolly 2003).
- Spatial gradient model (Rasmussen 2010).
- Quantification of group distribution in niche space (e.g. Layman et al. (2007)).
- Directional change in $\delta^{13}C$-$\delta^{15}N$ bi-plot space (Wantzen et al. 2002).
- Numerical simulations (Matthews & Mazumder 2004).
- Tissue type-based comparisons (Tieszen et al. 1983).
- Isotope incorporation models (growth-dependent models, e.g. Fry & Arnold (1982); time-dependent models, e.g. Hobson & Clark (1992); growth-and-time-dependent models, e.g. Carleton & Martinez Del Rio (2010); multi-compartment models, e.g. Cerling et al. (2007)).
- Lipid Correction Models as an alternative to chemical extraction (Kiljunen et al. 2006, Post et al. 2007).

40. OSMOSE (Object-oriented Simulator of Marine Ecosystems)2 is an individual-based model developed to explore the functional role of biodiversity in the exploitation of multispecies systems. The modeled species interact inside a food web framework. Input data are basic

[2] https://osmose-model.org/

biological parameters available to a large extent from open-access sources such as FishBase (Shin & Cury, 1999).

41. StrathE2E[3] is an end-to-end model that allows food web modeling and aims at aiding the management of fisheries active on the continental shelf with an ecosystem-based approach. The web app runs "what if" experiments using environmental data and fishing rates as its input.

42. Phillips et al. (2014) suggested best practices for using mixing models in food web studies. Arostegui et al. (2019) and Silberberger et al. (2021) addressed the biases introduced by sample pre-treatment in mixing model approaches.

43. Link et al. (2015) proposed cumulative trophic patterns based on sigmoidal cumulative biomass (cumB)-trophic level (TL) and 'hockey-stick' production (cumP)-cumB curves, which are strongly supported by their observed occurrence in over 120 marine ecosystems globally. Curve parameters are suggested as possible ecosystem thresholds, and supposed helpful to manage the marine ecosystems of the world.

44. A widely used ecosystem modelling approach considered a major tool in the EcAp, which uses food web data - along with environmental variables and human pressures, e.g. fishery data - as its inputs, is the "Ecopath system"[4]. Born in the early 1990s (Christensen & Pauly 1992) as a free ecological/ecosystem modeling software, in its current form it is made of three modules: Ecopath, Ecosim and Ecospace (Pauly et al. 2000). Among its several potential uses, this modelling tool takes abundance data and trophic guild of marine organisms to assess the impact of fishing and other human activities on the ecosystem in general and on the food web in particular. Hundreds of scientific papers have been published that employ the Ecopath suite as a modelling tool (Colleter et al., 2013). Among the Mediterranean case studies the following should be kept in mind for their wider scope and/or the scientific advancement provided: Coll & Libralato (2012), Steenbeek et al. (2013), Pennino et al. (2020), and Keramidas et al. (2023).

4.4 Methods of monitoring and assessment under MSFD and European conventions

- *MSFD*

45. The Marine Strategy Framework Directive (MSFD)[5] establishes in Art. 1 that *"Member States shall take the necessary measures to achieve or maintain good environmental status in the marine environment by the year 2020 at the latest"*. To assist in this overarching goal eleven qualitative descriptors are considered, including Descriptor 4 (**D4 - Food webs**): *All elements of the marine food webs (...) occur at normal abundance and diversity and levels capable of ensuring the long-term abundance of the species and the retention of their full reproductive capacity*.

46. The attainment of MSFD targets implies the respect of descriptors. To this purpose a number of indicators have ben identified and more can ideally be proposed. The main indicators of food web status have been listed in Chapter 4.2 above. Comm. Dec. 2017/848[6] provides four criteria to be met based on the identification and quantification of trophic guilds, which should be established along with their threshold values by Member States. Those criteria are based on the assessment of indicators to be assessed inside trophic guilds: two primary (diversity: D4C1, and abundance: D4C2), and two secondary (size distribution; D4C3, and productivity: D4C4). Trophic guilds can be drawn from methods described in Chapter 4.1 above and the literature cited therein. ICES gave technical guidance on undertaking technical assessment for D4 (ICES 2021).

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[3] https://outreach.mathstat.strath.ac.uk/apps/StrathE2EApp/

[4] https://ecopath.org/about/

[5] Directive 2008/56/EC of the European Parliament and of the Council of 17 June 2008 establishing a framework for community action in the field of marine environmental policy (consolidated version)

[6] Commission Decision 2017/848/EU 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.

Thompson et al. (2020) proposed a feeding guild assessment indicator suggesting the choice of four to nine guilds according to the ecosystem characteristics.

47. It is worth mentioning the national MSFD assessments and reports that Member States deliver periodically to the European Commission[7].

48. The EU-funded DEVOTES project[8] produced a vast list of GES indicators for D4 (Teixeira et al. 2014, Heiskanen et al. 2016) that has been updated up to some point.

49. Overall, D4 is maybe the most challenging of all MSFD descriptors, since it is very hard to identify simple indicators able to assess the health of highly dynamic and complex interactions like those occurring in marine food webs, considering the differences in climate and habitats across European seas (Rombouts et al. 2013). Piroddi et al. (2015) compared the output of 44 ecological models implemented in European case studies, showing that food web indicators were often addressed by models (86 out of 200 indicators derived).

50. Machado et al. (2021b) discussed four food web indicators in the light of their application to the assessment of D4.

51. Korpinen et al. (2022) assessed the application of food web indicators and models in the Baltic Sea, and provided advice for an enhancement of their use.

OSPAR

52. Among OSPAR's goals, one is to advise about the fulfilling of MSFD objectives in the OSPAR area (OSPAR ICG COBAM 2012, Elliott et al. 2017, Padegimas et al. 2017).

53. The OSPAR Commission carries out thematic assessments of a number of environmental themes, one of which is food webs. In this framework three food web common indicators are currently used[9]:

- Size composition in fish communities (FW3)
- Change in average trophic level of marine predators in the Bay of Biscay (FW4)
- Proportion of large fish (Large Fish Index) (FC2).

54. OSPAR has implemented a pilot assessment of food webs in the Baltic area using Ecological Network Analysis (Safi et al. 2019, Schückel et al. 2023), pilot assessment of feeding guilds (Thompson et al. 2023), and assessment of changes in average trophic level of marine consumers (Preciado et al. 2023). An assessment of the ecological status of the OSPAR area was carried out by McQuatters-Gollop et al. (2022) as an expansion of the OSPAR 2017 assessment of marine biodiversity.

55. Through the EcApRHA project (Applying an Ecosystem Approach to (sub) Regional Habitat Assessment) OSPAR aims at addressing gaps in the development of biodiversity indicators for the OSPAR Regions and at overcoming the challenges in the development of indicators relating to MSFD descriptors such as D4 (Elliott et al. 2017).

- HELCOM

56. The Baltic Marine Environment Protection Commission (also known as the Helsinki Commission, HELCOM) among its goals adopts Recommendations related to the protection of the marine environment in the Baltic Sea. In the framework of the working group on biodiversity (WG BioDiv) an expert group on food webs (EG FOODWEB) has been established to develop quantitative, indicator-based assessments to support the implementation of the MSFD objectives. The expert group has supported the 2016-2021 HELCOM Holistic Assessment (HOLAS 3)[10] (HELCOM 2023). Annex A1.6 of HELCOM (2023) describes the methodology used to assess indicators D4C1 and D4C2 of the MSFD and the Ecopath with Ecosim model (EwE: see chapter 5 below).

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[7] https://water.europa.eu/marine/policy-and-reporting/msfd-reports-and-assessments
[8] https://mcc.jrc.ec.europa.eu/main/dev.py?N=simple&O=309&titre_page=DEVOTES
[9] https://www.ospar.org/work-areas/cross-cutting-issues/ospar-common-indicators
[10] https://helcom.fi/baltic-sea-trends/holistic-assessments/state-of-the-baltic-sea-2023/

57. The holistic assessment acknowledged that a quantitative evaluation of food web status in the Baltic Sea is currently not possible due to a lack of harmonized data and regionally agreed-upon indicators (Nordström et al. 2021, HELCOM 2023).
58. Eero et al. (2021) addressed the contribution of food web knowledge to the management of living resources in the Baltic Sea.

5. Projects and initiatives

59. A certain number of research projects and other initiatives exist in Europe with regards to the assessment and monitoring of marine food webs. The Mediterranean seems far less active in this field as regards project-leading institutions, although a number of scientific papers on Mediterranean case studies have been produced (see references in Chapter 4 above). A list of recently concluded and ongoing European projects and initiatives that develop new approaches or use existing methodologies to investigate marine food webs is given hereafter.

- *PROJECTS*

60. DEVOTES: Development of innovative tools for understanding marine biodiversity and assessing good environmental status (EC FP7, 2012-2016). The project has developed tools to understand and describe biodiversity, food webs and seafloor integrity status at a European scale, including as many components of the ecosystem as possible, providing the scientific knowledge, upon which appropriate monitoring and management strategies under MSFD can be designed and made available for managers. The activities included support the development and testing of indicators and analytical tools for MSFD Descriptor 4.
61. OCEAN-CERTAIN: Ocean Food-web Patrol – Climate Effects: Reducing Targeted Uncertainties with an Interactive Network (EC FP7, 2013-2017). The project identifies and quantifies multi-stressor impacts and feedbacks and how these alter the functionality and structure of the food web and the efficiency of the biological pump in different biogeographical regions. This is done by utilizing existing ecosystem models employing existing data, in addition to mesocosm, lab-scale experiments and field studies.
62. Assessment of marine and coastal ecosystems and ecosystem services (EFESE project, 2015-2017). This project has carried out three specific research operations in addition to the synthesis of available knowledge: (i) consultation with the stakeholders interested in marine habitats and their uses; (ii) studying the forms of demand to which ecosystems and ecosystem services are subjected; (iii) analyzing processes of patrimonializing ecosystems and ecosystem services. Marine food webs are among the ecosystem features that were included in the study.
63. IMMERSE: Integrating Macroecology and Modelling to Elucidate Regulation of Services from Ecosystems (NERC, 2014-2018). The project targets key knowledge gaps by applying the latest method developments in understanding food webs. Isotopic methods are used to trace the relative input of seaweed and planktonic algae into the base of the food web; these isotopic tracers are followed in the lab and in the wild to understand exactly how these plants are incorporated into the rest of food web; new image analysis technology is used to quantify the full size range of organisms in the sea; and the latest molecular techniques to trace who eats whom are employed.
64. FaCE-It: Functional biodiversity in a Changing sedimentary Environment: implications for biogeochemistry and food webs in a managerial setting (BRAIN-be, Belgian Research Action through Interdisciplinary Networks, 2015-2020). The project aims at understanding the impact of fining and hardening on the benthic ecosystem functioning (i.e. biogeochemical cycling and food webs) from the local scale to those larger scales in which marine managers are interested. Focus is on the effect of sediment fining on nutrient cycling, and on the effect of hardening on food-web structure and carbon flow.

65. CERES: Climate change and European aquatic resources (EC Horizon 2020, 2016-2020). Among other objectives, the project integrates the knowledge on changes in productivity, biology and ecology of wild and cultured animals (including key indirect / food web interactions), and ‘scale up’ to consequences for shellfish and fish populations, assemblages as well as their ecosystems and economic sectors

66. CoDINA: Cod, DIet and food web dyNAmics (Marinforsk, Research Council of Norway, 2016-2021). The project aims to increase the understanding of pelagic food web dynamics in sub-Arctic ecosystems by studying the interaction between Barents Sea cod and its prey. The mechanisms generating predator-prey interactions will be examined to gain a deeper understanding of trophic connections in the Baltic Sea.

67. MicroPolar: Processes and Players in Arctic Marine Pelagic Food Webs (Marinforsk, Research Council of Norway, 2016-2021). The project (i) quantifies carbon flow in Arctic microbial food webs and the factors regulating production and consumption of dissolved organic carbon and CO_2; (ii) refines current microbial food web models experimentally in Arctic systems by exploring combined effects of altered microbial community structure and nutrient conditions as foreseen caused by progressing climate changes.

68. MISTIC SEAS III: Developing a coordinated approach for assessing Descriptor 4 via its linkages with Descriptor 1 and other relevant MSFD descriptors in the Macaronesian sub-region (EC DG ENV/MSFD 2018, 2019-2021). The project addresses the assessment of the environmental status of the marine environment, based on D4 in the Macaronesia sub-region, following the criteria set by the European Commission. Methodologies that enable to identify trophic guilds are defined and tested. The results of this work are integrated with the criteria established for MSFD descriptors D1 and D3.

69. Progetto 3 Golfi (Regional Government of Sicily, 2022-2023). The project aims at assisting in the development of a new management plan for Sicilian fisheries. Among its tasks, an Ecopath with Ecosim model including data on the food web of soft bottom communities is developed.

70. MARAT: Marine Arctic Resilience, Adaptations and Transformations (multiple funders, 2020-ongoing?). This project integrates models, local knowledge, and comparative case studies to assess the resilience of Arctic marine food webs to climate and fishing pressures, and how communities adapt or transform to such changes.

71. ECOTIP: Investigating Ecological Tipping Cascades in the Arctic seas (EC Horizon 2020, 2020-ongoing). The project maps Arctic biodiversity and investigates the effects of human activities and other external drivers on ecosystem components and processes, including food webs.

72. Multiple States and Transition Paths: Applications to Marine Ecosystems (University of Bergen, 2023-ongoing). The project will describe the interlink among multiple predator-prey groups. This description is a parsimonious representation of a marine ecosystem. By extending the deterministic to a stochastic framework, they aim to mimic the nature of empirical marine ecosystems.

73. IMBER: Integrated Marine Biogeochemistry and Ecosystem Research (International Science Council, Scientific Committee on Oceanic Research, 2005-ongoing). This large-scale project is focused on in-depth regional and topical analyses and comprehensive comparisons of diverse marine ecosystems. The results from these activities provide new understanding about the potential effects of global environmental changes on biogeochemical cycling, food web dynamics, and impacts and linkages to human systems at multiple scales.

74. ACTNOW: Advancing understanding of cumulative impacts on European marine biodiversity, ecosystem functions and services for human wellbeing (EC Horizon 2020, 2023-ongoing). Among the other goals, the project employs state-of-the-art bio-logging technology and molecular methods, in combination with knowledge on oceanographic processes to understand the effects of agents of change on the ecology and population dynamics through marine food webs.

75. Project Breathless (U.S. National Science Foundation). The project focuses on how low-oxygen conditions influence fish, their habitats and the food webs that support them, as well as ecosystem services, including fisheries production, in the Baltic Sea, Gulf of Mexico and Lake Erie.

- OTHER ACTIVITIES

76. ZooTrait: Insight into the structure and function of marine pelagic food webs: traits and trade-offs in zooplankton feeding behaviour (EC FP7, 2015-2017). The project aims to increase the ability to understand the factors that govern the structure and function of pelagic food webs, and predict their changes under different environmental conditions, including global climate change scenarios.

77. FIMAF: Isotopic evidence for the impacts of fishing on marine food web structure (EC Marie Skłodowska-Curie Actions, 2016-2018). Using stable isotope analysis techniques for tackling the challenging question of the impacts of fishing activities on fish diet and trophic level, the project investigates temporal shifts in otolith signatures occurring in response to variability in fishing pressure.

78. BLUE2: Assistance for better policy-making on freshwater and marine environment. BLUE2 is an activity of the EC's JRC (Joint Research Center) in support of the evaluation, implementation and possible review of the EU water and marine legislation. The related publications and reports produced by the MEME (Network of Experts for ReDeveloping Models of the European Marine Environment) focus, among many other subjects, also on food web modelling (e.g.: Corrales et al., 2020; Macias Moy et al., 2018a, 2018b, 2020; Piroddi et al., 2020, 2021).

79. Follow the food! Using food webs as indicators of ecosystem functioning (MARE, Marine and Environmental Sciences Centre, 17-18/1/2023). This workshop brought together specialists working on different aspects of food web ecology and discussed the current and future state of food webs in the context of the indicators of GES.

80. NSSIA: Isotope ecology network in the Baltic Sea region (Stockholm University, 2014-20??). The project aims to facilitate mapping of trophic structure of the Baltic ecosystems through the analysis of bulk C, N, and S isotopes. The usefulness of isotope analyses of essential and non-essential fatty acids and amino acids as indicators of trophic position and base of the food chain is investigated. Ultimately, these data will be used to explain biological responses, trophic magnification factors of contaminants in biota, as well as effects of nutrient and contaminant sources on the spatial differences in marine food webs and in sea-atmospheric transfer.

81. Fish species occurrence and food web dynamics in a coastal lagoon (Irish Marine Institute, 2020-2021). This project supports the MFSD requirement for data and reporting under Descriptors 1, 4 and 6, and provides data into Ireland's food-web contributions to the 2023 OSPAR Quality Status Report (QSR) for the NE Atlantic.

82. Plankton-fish interactions: an understudied link in Baltic Sea food webs and fisheries management (Stockholm University, 2020-2023). This activity investigates prey preference of small pelagic fish including the entire prey spectrum using novel molecular tools that amplify and sequence low levels of DNA combined with network models, to project trophic coupling under changing climate, nutrient and fisheries scenarios.

83. BalticCAT: Cumulative effect Assessment Tools for the Baltic Sea (Stockholm University, 2020-ongoing). This activity applies spatial-temporal food web models to test the cumulative effects of natural and anthropogenic stressors on the state of the Baltic Sea ecosystem, and to predict the effects of management actions such as fisheries regulations, reduced nutrients, contaminant emissions and marine protected areas on ecosystem state.

84. ECOBASE: A repository solution to gather and communicate information from Ecopath with Ecosim (EwE) models (UMR DECOD, France). This activity makes available to scientists an up-to-date and comprehensive list of published ECOPATH models. It is managed and

supported by the members of the Model Repository working group of the Ecopath Research and Development Consortium (ERDC).

85. Med QSR: Mediterranean Quality Status Reports. The Mediterranean Quality Status Reports 2017 and 2023 proved a useful tool to assess the status of the Mediterranean marine ecosystem and to achieve the GES, but they do not still include EO4 (i.e., food web assessment) due to lack of shared consolidated protocols for data collection and analysis at regional level.

6. Research teams

86. Several research teams dedicated to the assessment and monitoring of marine food webs exist in Europe. Only a minor number of them is based in Mediterranean countries. Here follows a list of the currently active research teams.

87. MEME, the Network of Experts for ReDeveloping Models of the European Marine Environment (EC's JRC) carries out the modelling efforts of JRC in support of the EU marine legislation.

88. EcApRHA Food Web working group is one of the WGs of the EcApRHA project developed inside OSPAR.

89. ICES Working Group on Ecosystem Assessment of Western European Shelf Seas (WGEA-WESS) aims at providing high-quality science in support to holistic, adaptive, evidence-based management in the Celtic seas, Bay of Biscay and Iberian coast regions.

90. ICES WKFOODWEB: Workshop on the operational use of Food Web indicators and information (ICES, Denmark) reviews the latest advances in marine food web ecology and supports the ICES advice to fishery science. The next meeting is scheduled on 19-23 February 2024.

91. Group AML Karlson (Stockholm University, Sweden) studies food-web and ecosystem ecology, ecotoxicology and stress ecology, through environmental monitoring and stable isotope techniques.

92. Group Winder (Stockholm University, Sweden) studies drivers of food web interactions and community dynamics to better understand the ecological impacts of environmental change on ecosystem functioning.

93. Fish in food-webs (Swedish University of Agricultural Sciences, Sweden) links variation in body size and community composition to ecological and evolutionary dynamics in changing environments.

94. Regime Shifts DataBase, Theme: Marine food webs (Stockholm Resilience Centre, Sweden) analyzes regime shifts in aquatic systems that involve an abrupt increase in the dominance of lower trophic level groups within aquatic food webs.

95. Pelagic food-webs (SDU, University of Southern Denmark) belongs to the Ecology Group of SDU and studies trophic interactions in the pelagic domain in the sea and freshwaters.

96. Centre for Ocean Life (DTU, Technical University of Denmark) carries out studies on marine food web ecology.

97. EccoWebs (CCMAR, Algarve University, Portugal) investigates the effects of environmental change on marine organisms, populations and food webs, from the sub-cellular to the whole-animal level, and complex trophic networks.

98. Ecosystem Modelling Working Group (GEOMAR, Germany) aims at investigating the relationships between biodiversity and ecosystem functioning under the effect of global warming, and at quantifying the impact of multi-stressors on food web structure and functioning.

99. Ecosystem Understanding Team (CEFAS, United Kingdom) explores the connectivity between nutrient flows, food webs and the impact of human activities on these ecosystem components, and how marine ecosystem dynamics are driven by linkages between climate forcing, hydrography, benthic & pelagic food webs and higher trophic levels or species higher up the food chain.

100. EII, Ecopath International Initiative (Barcelona, Spain) is focused on the contribution to the sustainability of living resources and ecosystems through the development, implementation, education and promotion of analytical tools and ecological modeling, with special emphasis on the use of modeling approach Ecopath with Ecosim.
101. Dynamics of Ecosystems and Computational Oceanography team (OGS, National Institute of Oceanography and Applied Geophysics, Italy) analyzes trophic networks and spatial distribution of marine organisms in relation to environmental variables to identify ecological niches, explore population dynamics, ecosystem health and biodiversity, and to explain and predict possible variations at different spatio-temporal scales.

7. Knowledge gaps

102. A good knowledge of biodiversity and food web functioning – considered closely related, see *inter alia* the Commission Decision (EU) 2017/848, and EC (2020) - is paramount to any holistic approach. The assessment and monitoring of marine food webs is still one of the most challenging research topics. In the frame of MSFD for example, D4 is considered the least well-developed of the descriptors, as metrics and indicators are generally not well-established, thus the definition of GES for food webs is particularly difficult (Crise et al., 2015; ICES, 2015). According to a recent report the GES for marine food webs in EU waters is largely not assessed or unknown, with some data available only from coastal and shelf systems (EC, 2020). The main difficulties lie in data collection, reliable indicators, and thresholds setting, which all lead to the lack of common, shared targets and of harmonized monitoring initiatives. Such gaps occur at global (or European) level and even more so at the Mediterranean level, where the history of food web studies is more recent and relatively less developed.
103. Here follows a tentative list of the main gaps as drawn from the scientific and grey literature and from EC documents (Rombouts et al., 2013; Crise et al., 2015; ICES, 2015; EC, 2020; Machado et al., 2021a):

- high uncertainty related to top predators productivity as a measure of food web functioning, and thus difficulty in estimating the energy flux to lower trophic levels;
- some proposed size-based food web indicators (like e.g., LFI) are not well suited to this role since they depend on the relationship between fishing pressure and size structure of target and non-target populations, which is still not fully quantitatively known;
- poor reliability of abundance trends in single species or species groups because of high inter-annual variability due to natural and anthropogenic pressures;
- need of extensive datasets on feeding habits, especially for species at lower trophic levels;
- need to extend or adapt locally established operational indicators to the larger scale of EU waters;
- lack of more integrative, ecosystem-based indicators vs. indicators based on single species or species groups;
- unknown long-term effects of global change (warming, acidification) on food web components and functioning;
- role and impact of invasive alien species and their use of resources shared with indigenous species;
- poorly known effect of changes in nutrients on carbon fluxes;
- poorly known effects of habitat loss due to anthropogenic activities and consequent change in the benthic communities composition and structure;
- general lack of baseline data, especially from hard bottoms, coastal areas and deep seas, despite the existence of national/regional monitoring programs;
- very few available assessments of D4 under MSFD, and hardly comparable;
- lack of appropriate metrics;
- few appropriate datasets (that need to address an extensive number of ecosystem components);

- lack of knowledge on direct cause-effect relationships between anthropogenic pressures and their effects on food-web indicators;
- lack of comparable data between taxonomic groups to develop fully operational indicators that can integrate trophic structure and functions;
- need to focus on well studied pelagic and benthic species;
- lack of thresholds to describe and identify the ecological status of food webs;
- only few indicators have been fully operationalized, i.e. quantitatively defined, assessed in relation to a defined threshold, and responding clearly to anthropogenic activities;
- lack of regionally coordinated monitoring of indicators;
- unreliability of biomass data used in models due to unreported catches and other uncertainties in mortality estimates;
- trophic level estimations are difficult for omnivorous species;
- lack of data on invertebrates, non-commercial fishes and non-indigenous species;
- limited temporal and spatial coverage for data on trophic links and organisms abundance, which are sometimes drawn from decades-old data sets collected in communities that may have undergone major changes and shifts;
- the assessment scale should be related to the entire scale at which each drive of change (e.g., fishing) is exerted homogeneously;
- lack of knowledge on the potential impact of future changes in the abundance and/or distribution of key species on food web structure and functions;
- lack of knowledge on trophic interactions in coastal rock and biogenic reef habitats.

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