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Agenda Item 6: Technical Guiding Elements on IMAP Implementation: Assessment Criteria and Scales, Thresholds, Baseline Values

Assessment Criteria Methodology for IMAP Common Indicator 13: Pilot Application in Adriatic Sub-region

UNEP/MAP
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Note by the Secretariat

In line with Decision IG.23/6 related to 2017 Mediterranean Quality Status Report (MED QSR) adopted at COP 20 (Tirana, Albania, December 2017), the Contracting Parties and the Secretariat are encouraged to test the assessment criteria including the coastal water types reference conditions and boundaries in the Mediterranean as endorsed by the Decisions IG.22/7 (Athens, Greece, February 2016). Furthermore, it was requested to develop region-wide harmonized criteria for reference conditions and threshold/boundaries values for key nutrients in water column, taking account of available standards for coastal waters and use of data stored in other databases were some of the Mediterranean countries regularly contribute.

To that effect the Meeting of the Ecosystem Approach Correspondence Group on Pollution Monitoring (Podgorica, Montenegro, 2 - 3 April 2019), considered data availability for setting the assessment criteria for nutrients and consequently recommended to the Secretariat to undertake actions to set the reference conditions not only for chlorophyll *a*, but also for nutrients, transparency and oxygen, as minimum requirements. That work needs to build on the substantial knowledge acquired from marine environment monitoring.

In line with the Programme of Work 2020-2021 adopted by COP21 (Naples, Italy, December 2019), the MED POL Programme has undertaken further actions aimed at harmonization and standardization of the monitoring and assessment methods related to IMAP Pollution and Marine Litter Cluster (Activity 2.4.1.4), including present work aimed at proposing reference conditions and boundary values for nutrients.

Due to nitrogen/phosphorus limitations present in the Mediterranean (i.e. restricted measurements of Dissolved Inorganic Phosphorous - DIP), as well as due to limited data availability and related demanding statistics, the present document paves the way for setting the reference conditions and boundary values for Dissolved Inorganic Nitrogen (DIN) and Total Phosphorous (TP). In that respect, it elaborates possible use of the following tools and methods: methodological approach developed for Adriatic Sea; Best Practice Guide for nutrients toolkit (JRC) and FAN/FLU index (Spain), in relevant sub-areas. In addition, the practical application of the methodological approach is showcased for Adriatic Sea. Setting of the assessment criteria for transparency and dissolved oxygen is found premature at this stage.

In order to ensure use of the new assessment criteria within preparation of the inputs for 2023 MED QSR, the present proposal of the process for setting reference conditions and boundary values for DIN and TP within implementation of IMAP CI 13 was submitted to the Meeting of the Ecosystem Approach Correspondence Group on Pollution – CorMon Pollution that was held from 26 - 28 April 2021.

Considering the evolving nature of this document the Meeting of CorMon Pollution agreed to recommend its use as a basis for progressing towards setting the assessment criteria for DIN and TP, and recommended its submission to the Meeting of the MED POL Focal Points for its consideration. It was emphasised that further elaboration will continue, including within the Online Working Group (OWG) on Eutrophication. To this aim, the Contracting Parties were invited to support this work through: i) analysis of available/ new monitoring data; and ii) elaboration and testing of proposed methodological approaches for setting boundary values, including relevant statistical approaches, as suitable for specific areas in Mediterranean sub-regions.

Further to the discussion that took place during resuming session of the Meeting of MEDPOL Focal Points that was held on 9 July, the present document was recommended for submission to the 8th EcAp Coordination Group Meeting. Given a short time period between the Meeting of CorMon on Pollution Monitoring and the Meeting of MEDPOL Focal Points that was held from 27 to 28 May 2020, the present document does not contain changes undertaken after the Meeting of CorMon Pollution in order to address written comments received during or immediately after this meeting.

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List of acronyms / abbreviations

BQE	Biological Quality Element
CI	Continental Influence
CIs	Common Indicators
CIS	Common Implementation Strategy
CPs	Contracting Parties
CWs	Coastal Waters
DIN	Dissolved Inorganic Nitrogen
DIP	Dissolved Inorganic Phosphorous
EMODnet	European Marine Observation and Data Network
EQR	Ecological Quality Ratio
EU	European Union
F_dil	Dilution factor
FAN	Phosphate-Ammonium-Nitrate Index
FLU	FLUviality Index
G_mean	Geometric mean
GES	Good Environmental Status
G/M	Good/Moderate (threshold)
HELCOM	Baltic Marine Environment Protection Commission - Helsinki Commission
IMAP	Integrated Monitoring and Assessment Programme
JRC	Joint Research Centre - the European Commission's science and knowledge service
LMs	Linear Models
MED GIG	Mediterranean Geographical Intercalibration Group
MEDPOL	The Programme for the Assessment and Control of Marine Pollution in the Mediterranean
MSFD	Marine Strategy Framework Directive
MSs	Member States
Nm	Nautical Mile
OWG	Online Working Group
QSR	Quality Status Report
RC	Reference Conditions
RSCs	Regional Seas Conventions
SD	standard deviation
TN	Total Nitrogen
TP	Total Phosphorous
TRIX	Trophic Index
UNEP/MAP	United Nations Environment Programme / Mediterranean Action Plan
WFD	Water Framework Directive
WISE	Water Information System for Europe

1 Introduction

1. Eutrophication is a process driven by enrichment of water by nutrients, especially compounds of nitrogen and/or phosphorus, leading to: increased growth, primary production and biomass of algae; changes in the balance of nutrients causing changes to the balance of organisms; and water quality degradation (IMAP, 2017[1]). Seawaters depending on nutrients loading and phytoplankton growth are classified according to their level of eutrophication. Low nutrient/phytoplankton levels characterize oligotrophic areas, whilst water enriched in nutrients is characterized as mesotrophic and water rich in nutrients and algal biomass is characterized as eutrophic.

2. Good environmental status (GES) with regard to eutrophication is achieved when the biological community remains well-balanced and retains all necessary functions in the absence of undesirable disturbance associated with eutrophication (e.g. excessive algal blooms, low dissolved oxygen, declines in sea-grasses, kills of benthic organisms and/or fish) and/or where there are no nutrient-related impacts on sustainable use of ecosystem goods and services. Specifically, for IMAP Common Indicator 13 related to key nutrients in water column GES is achieved when concentrations of nutrients in the euphotic layer are in line with prevailing physiographic, geographic and climate conditions.

3. Coastal waters (CWs) are among the most highly impacted ecosystems in the world presenting inherently high variability over both spatial and temporal scales (Reyjol et al., 2014[2]). In those environments, the greatest impacts of increasing nutrient concentrations have been observed at sites with restricted water exchange, resulting in phytoplankton and macroalgal blooms (Tett et al., 2003[3]; Teichberg et al., 2010[4]).

4. A significant amount of research has been done in developing and intercalibrating biological indicators to assess impact of eutrophication in coastal waters (Borja et al., 2013[5]). Phytoplankton is the most suitable for assessing eutrophication due to direct response to nutrient conditions (Devlin et al., 2007[6]). However, less attention has been directed to linking ecological status to management actions and establishing meaningful and consistent nutrient criteria to support achievement of GES (Hering et al., 2015[7]).

5. The European experience is relevant in the field. A comparison of nutrient boundaries set for the Water Framework Directive (WFD) and the Marine Strategy Framework Directive (MSFD) in transitional, coastal and marine waters across EU Member States (Dworak et al., 2016[8]) revealed a huge variability in nutrient concentrations boundaries, but also in other relevant aspects such as the nutrient parameters and metrics used, the time of year assessed, the reference conditions established. It also revealed that often MSs` boundary values of nutrient concentrations do not follow relevant Regional Seas Conventions (RSCs) nutrient standards.

6. The possible implications of the wide variations in the nutrient concentration boundaries need to be understood in the context of establishing appropriate nutrient boundaries to achieve GES. A Best

[1] IMAP (2017). Integrated Monitoring and Assessment Programme of the Mediterranean Sea and Coast and Related Assessment Criteria UNEP, Athens, 52 pp.
[2] Reyjol, Y., Argillier, C., Bonne, W., Borja, A., Buijse, A. D., Cardoso, A. C., et al. (2014). Assessing the ecological status in the context of the European water framework directive: where do we go now?. Sci. Total Environ. 497, 332–344.
[3] Tett, P., Gilpin, L., Svendsen, H., Erlandsson, C. P., Larsson, U., Kratzer, S., et al. (2003). Eutrophication and some European waters of restricted exchange. Cont. Shelf Res. 23, 1635–1671.
[4] Teichberg, M., Fox, S. E., Olsen, Y. S., Valiela, I., Martinetto, P., and Iribarne, O. (2010). Eutrophication and macroalgal blooms in temperate and tropical coastal waters: nutrient enrichment experiments with Ulva spp. Global Change Biol. 16, 2624–2637.
[5] Borja, A., Elliott, M., Henriksen, P., and Marb, N. (2013). Transitional and coastal waters ecological status assessment: advances and challenges resulting from implementing the European water framework directive. Hydrobiologia 704, 213–229.
[6] Devlin, M., Best, M., Coates, D., Bresnan, E., O'Boyle, S., Park, R., et al. (2007). Establishing boundary classes for the classification of UK marine waters using phytoplankton communities. Mar. Pollut. Bull. 55, 91–103.
[7] Hering, D., Borja, A., Carstensen, J., Carvalho, L., Elliott, M., and Feld, C. K. (2010). The European water framework directive at the age of 10: a critical review of the achievements with recommendations for the future. Sci. Total Environ. 408, 4007–4019.
[8] Dworak, T., Berglund, M., Haider, S., Leujak, W. and Claussen, U. (2016). A comparison of European nutrient boundaries for transitional, coastal and marine waters. Working Group on ecological Status ECOSTAT.

Practice Guide (BPG, Phillips et al., 2018[9]) has been elaborated in this context. Its purpose is to help in achieving GES in surface waters. It complements previous guidance on eutrophication assessment (EC, 2009[10]) by providing more targeted advice on how to link nutrient concentrations in surface waters to specific policy objectives. The new guide includes a tool kit to facilitate the application of the different statistical approaches proposed to establish the nutrients` targets.

7. The statistical approaches proposed in the BPG in coastal and transitional waters focus on the pressure-response relationships found between the nutrients and phytoplankton. Most of the EU MSs use the concentration of Chlorophyll a (Chl *a*) as a proxy measure for phytoplankton biomass within intercalibration, whereas most of the indicators of other sub-elements (phytoplankton composition and blooms) have not yet been intercalibrated (Garmendia et al., 2013[11]). This corresponds to monitoring of IMAP Common Indicator 13 (i.e. concentration of key nutrients in water column) and Common Indicator 14 (i.e. concentration of Chl *a*) that does not define the criteria/sub-indicators related to the harmful algal bloom; photic limit (transparency) of the water column; relative abundance or depth distribution of macrophyte communities; as well as the species composition and relative abundance of macrofaunal communities, as mandatory parameters within IMAP implementation.

1.1 Nitrogen or phosphorus limitation

8. All sub-indicators/parameters related to IMAP CIs 13 and 14 are monitored or have been defined for regular monitoring within implementation of recently prepared national IMAP Pollution Cluster monitoring programmes. The temporal scales are well harmonized among Parties. Only two Parties have already defined the scales according to the type of waters and the rest will do so during the 1st implementation phase.

9. However, in the Mediterranean region there are many differences in the nutrients` parameters assessed, the assessment period (summer, year-round, i.e. annual), and in the statistic used (mean, median or 90th percentile) within assessment of the conditions of saline waters. These differences can be also observed within the four marine ecoregions defined by the MSFD and even between transitional, coastal and marine waters at national levels (Dworak et al., 2016).

10. In general nitrogen, rather than phosphorus, is considered to be the most likely limiting nutrient in many temperate coastal waters (Tsirtsis, 1995[12]). However, for the Mediterranean area it is the opposite and can be summarised as presented here-below.

11. In the Mediterranean phosphorus is often the limiting nutrient (Lazzari et al., 2016[13]; Thingstad et al., 2005[14]), although it is closely followed by nitrogen in this limiting role (Estrada, 1996[15]). The dissolved nitrogen to phosphorus ratio in the Mediterranean has been reported at about 21 to 23 in the

[9] Phillips G, Kelly M, Teixeira H, Salas F, Free G, Leujak W, Pitt Ja, Lyche Solheim A, Varbiro G, Poikane S, (2018) Best practice for establishing nutrient concentrations to support good ecological status, EUR 29329 EN, Publications Office of the European Union, Luxembourg
[10] European Commission [EC] (2009). Guidance Document on Eutrophication Assessment in the Context of European Water Policies. Common Implementation Strategy Guidance Document No. 23. Luxembourg: Office for Official Publications of the European Union.
[11] Garmendia, M., Borja, Á, Franco, J., and Revilla, M. (2013). Phytoplankton composition indicators for the assessment of eutrophication in marine waters: present state and challenges within the European directives. Mar. Pollut. Bull. 66, 7–16.
[12] Tsirtis, G.E. (1995). A simulation model for the description of a eutrophic system with emphasis on the microbial processes. Water Science and Technology 32: 189 -196.
[13] Lazzari, P., Solidoro, C., Salon, S. and Bolon, G. (2016). Spatial variability of phosphate and nitrate in the Mediterranean Sea: a modeling approach. Deep Sea Research Part 1: Oceanographic Research Papers 108: 39-52.
[14] Thingstad, T.F., Krom, M.D., Mantoura, R.F.C., Flaten, G.A.F., Groom, S., Herut, B., Kress, N., Law, C.S., Pasternak, A., Pitta, P., Psarra, S., Rassoulzadegan, F., Tanaka, T., Tselepides, A., Wassmann, P., Woodward, E. M. S., Wexels Riser, C., Zodiatis, G. and Zohary T. (2005). Nature of phosphorus limitation in the ultraoligotrophic Eastern Mediterranean. Science (New York) 309: 1 068-1 071.
[15] Estrada, M. (1996). Primary production in the Northwestern Mediterranean. Sci. Mar. 60, 55–64.

western part (Bethoux et al., 1992[16]), and even higher in the eastern basin (Krom et al., 1991[17]), which is quite different from the ratio of 16 found in the global ocean (Tyrrell, 1999[18]).

12. This has been proved by the model-based reconstruction of inorganic phosphate and nitrate distributions presented by Lazzari et al. (2016). The model demonstrated that when nutrient limitation occurs, in the vast majority of cases, phosphorus is the limiting nutrient, with the notable exception of the Alboran Sea, which is mainly nitrogen limited, and the southwest basin, in which both nitrogen and phosphorus can limit plankton growth. Ramirez et al. (2005[19]) showed nitrogen-limitation in the upper layers (first 20 m) of the north-west Alboran Sea during the winter, summer and autumn while Dafner et al. (2003[20]) suggested phosphorus limitation in the Strait of Gibraltar area. However, phosphorus limitation in the upper layers of this area is not due to very low phosphorus concentrations but rather to a very high Nitrogen:Phosphorus (N:P) ratio in the area east of Gibraltar, caused by the upwelling of deep Mediterranean waters. The Adriatic Sea is mostly phosphorus-limited (Rinaldi, 2014[21]). Along the coast of the northern and central Adriatic Sea, 90 % of the overall chlorophyll *a* variability is explained by Total Phosphorous (TP)(Giovanardi et al., 2018[22]). The high N:P ratios in the Adriatic Sea (>50) demonstrate that nitrogen does not limit algal growth. The Po River has a major effect on the whole Adriatic basin, determining patterns of both spatial and temporal variation.

1.2 Total or inorganic dissolved fraction, assessment period and statistics

13. During phytoplankton blooms, dissolved inorganic nutrients in surface layers may be almost completely consumed, leading to nutrient limitation at periods of peak of biological activity. This results in large seasonal variability of nutrients` concentrations. For this reason, Dissolved Inorganic Nitrogen (DIN) and Dissolved Inorganic Phosphorus (DIP) are usually measured and assessed when biological activity is lowest.

14. Total Nitrogen (TN) and Total Phosphorus (TP), which include all forms of nitrogen and phosphorus compounds, are also important parameters that should be assessed in addition to the dissolved nutrients, as it is already common practice for example by HELCOM and in Swedish, Finnish and Estonian coastal waters (HELCOM, 2009[23]). Adding total nutrients alongside inorganic nutrients, as core indicators, strengthens the link from nutrient concentrations in the sea to nutrient enrichment. These parameters may also allow consideration of climate change in the eutrophication assessment since higher temperatures will lead to year-round phytoplankton proliferation and/or possible changes in zooplankton communities.

15. In addition, there are other considerations that are not directly linked to setting of nutrients` thresholds but that are nevertheless important. Total nutrients are essential for determining nutrient budgets (i.e. an estimation of how much nutrient enters and leaves an area). Such budgets have particular importance in coastal and marine waters that are influenced by transboundary nutrients` transport and receive nutrients` inputs from other countries.

16. Furthermore, total nutrients are also essential parameters for establishing nutrient reduction targets. This means that monitoring and assessing both total and dissolved nutrients forms is necessary

[16] Bethoux, J.P., Morin, P., Madec, C. and Gentili, B. (1992). Phosphorus and nitrogen behaviour in the Mediterranean sea. Deep-Sea Res 39: 1 641-1 654.
[17] Krom, M. D., Kress, N., and Benner, S. (1991). Phosphorus limitation of primary productivity in the eastern mediterranean sea. Limnol. Oceanogr. 36, 424–432.
[18] Tyrrell, T. (1999). The relative influences of nitrogen and phosphorus on oceanic primary production. Nature (London) 400: 525-531.
[19] Ramírez. T., Cortés, D., Mercado, J.M., Vargas-Yáñez, M., Sebastián, M.,. Liger, E. (2005) .,Seasonal dynamics of inorganic nutrients and phytoplankton biomass in the NW Alboran Sea Estuar. Coast. Shelf Sci., 65 (4), pp. 654-670.
[20] Dafner, E.V., Boscolo, R. and Bryden, H.L. (2003). The N:Si:P molar ratio in the Strait of Gibraltar. Geophysical Research Letters 30: 1 506-1 509.
[21] Rinaldi, A. (2014). Fioriturі algali In Adriatico. Il bacino padano-adriatico tra sviluppo e scienza (Algal blooms in the Adriatic. The Padano-Adriatic basin between development and science) Editrice La Mandragora.
[22] Giovanardi, F., Francé, J., Mozetič, P., Precali, R. (2018). Development of ecological classification criteria for the Biological Quality Element phytoplankton for Adriatic and Tyrrhenian coastal waters by means of chlorophyll a (2000/60/EC WFD). Ecological Indicators. 93. 316-332.
[23] Helcom (2009). Eutrophication in the Baltic Sea — An integrated thematic assessment of the effects of nutrient enrichment and eutrophication in the Baltic Sea region. Baltic Sea Environment Proceedings No 115B.

for good understanding of the trend in nutrients` concentrations in the marine environment. However, within present monitoring of eutrophication, both within implementation of IMAP and MSFD, the measurement of all total (TN, TP) and dissolved (ammonia, nitrite, nitrate, orthophosphate, orthosilicate) forms are requested, in order to allow the calculation of all aggregated form as DIN and DIP.

17. To enable a consistent management approach, it is important to ensure a consistency between transitional, coastal and marine waters, at least within a region or subregion, in relation to monitoring and assessment of nutrients` parameters.

18. A final consideration is the choice of statistical measures used to aggregate nutrients` samples from a chosen assessment period in order to determine the concentrations of monitored parameter/indicator. Most of the Regional Seas Conventions use mean concentrations to ensure cross-comparisons. However, there might be cases where using the median is more robust, since it is less influenced by outliers. The choice of the appropriate statistics depends very much upon sampling size and quality of monitoring.

19. Since statistical distributions of chlorophyll *a* and nutrients tend towards log-normality, the parameter that better estimates the value around which central clustering occurs, is represented by the geometric mean, i.e. the arithmetic mean of log-data reconverted into numbers. The normalization of the data distributions by means of log transformation stabilizes the variance, with a standard deviation (sd) practically constant in the case of decimal log-transformation (Giovanardi and Tromellini, 1992[24]). These statistical properties indicate that the use of the annual geometric mean of data as the metric for setting the assessment criteria in Mediterranean is appropriate statistical measure.

20. Further to above considerations and given limited data availability as presented here-below, present document paves the way for calculation of the reference conditions and boundary values for Dissolved Inorganic Nitrogen (DIN) and Total Phosphorous (TP).

1.3 Data availability

21. The elaboration of data availability for calculation of the assessment criteria for DIN and TP includes the following 3 sources:

1) New data from IMAP Pilot Info System that include national monitoring data reported during its testing phase, and in particular after launching formal call for data reporting in June 2020;
2) All monitoring data from MEDPOL Database (i.e. data reported before 2012 that were uploaded into MEDPOL Database along with data reported to MEDPOL outside MEDPOL Database in the format of old metadata templates in period 2013-2019) that are in the process of their migration into IMAP Pilot Info System;
3) The EU data center (European Marine Observation and Data Network - EMODnet).

IMAP Pilot Info System and MEDPOL Database

22. A summary of both data reported both to IMAP Pilot Info System and MEDPOL Database are presented in Table 1.

Table 1: Datasets from IMAP Pilot Info System and MEDPOL Database available for calculation of the assessment criteria for DIN and TP. The datasets used in the 2017 and 2019 assessments are given for comparison.

Country	Data reported to MEDPOL Database	Data reported to IMAP Pilot Info system*	
		Validated	Not validated
Albania	2005-2006	-	
Algeria	2012	-	

[24] Giovanardi, F., Tromellini, E., 1992. An empirical dispersion model for total phosphorus in a coastal area: the Po River-Adriatic system. Sci. Total Environ. Supplement 201–210.

Bosnia and Hercegovina	2006-2008	2013-2020	
Croatia	2009, 2011-2014	-	
Cyprus	1999-2015	-	2016-2019
Egypt	2009-2010; 2012;2015	-	
France	2009-2012;2013; 2016		
Greece	1999-2000, 2004-2006	-	
Israel	2001-2013; 2015	2018-2019	
Italy	-	-	
Libya	-	-	
Malta	-	-	
Monaco	-	-	
Montenegro	2008-2012; 2014-2015; 2016-2017	-	2018-2019
Morocco	2006-2008; 2013-2015	-	
Syria	2007	-	
Slovenia	1999-2013, 2015-2016	2017-2019	
Spain	-	2019	
Tunisia	2002-2014	-	
Turkey	2005-2009, 2011, 2013-2015	-	

*Both validated and not validated data have been used for assessing sources for calculation of the assessment criteria for DIN and TP, given temporary not validated status may be assigned to data due to certain technical issues in IMAP Info System

23. It can be concluded that data available for calculation of the assessment criteria (i.e. reference conditions (RCs) and boundary values) for both DIN and TP are insufficient. Namely, for calculation of the RCs and boundary values as a minimum the following datasets need to be provided: three continuous years of monitoring with a minimum monthly frequency for Water types I and II and bimonthly to seasonal for Type III. It should also be noted that other supporting parameters (i.e. temperature, salinity and dissolved oxygen) need to be available for defining the water typology,

Data available in the EU data center (European Marine Observation and Data Network - EMODnet)

24. Given scarcity of data reported into IMAP Pilot Info System and MEDPOL Database, data availability in EMODnet has also been explored (Table 2). However, it must be noted that EMODnet data are limited only to Croatia, France, Greece, Israel, Italy, Montenegro, Spain, Tunisia and Turkey. There is also different format of EMODnet data compared to data reported into IMAP Pilot Info System. Therefore, a significant further work is needed to correlate and aggregate two data sources.

Table 2: Datasets for Chlorophyll a and nutrients available at EMODnet, for period 2015-2020.

Country	Total available data	Unrestricted
Croatia	429	-
France	2344	493
Greece	229	-
Israel	29	29
Italy	2156	1247
Montenegro	146	-
Spain	244	-
Tunisia	29	-
Turkey	726	180

25. In line with above elaborated data availability, the following Mediterranean sub-regions/sub-areas may be indicated for calculation of the reference conditions and boundary values for DIN and TP: Adriatic, North Western Mediterranean Sea, Tyrrhenian Sea, Aegean Sea and Levantine Sea. They are proposed also considering the areas that were initially proposed for calculation of the assessment

criteria for contaminants[25]. This can be considered the initial phase for establishing reference conditions and boundary values for nutrients, whereas the values can be proposed for entire Mediterranean upon data reporting from CPs that will fill present data gaps.

26. Online Working Group for EO5 needs to substantively contribute to increased data availability and statistical calculation of data in above indicated areas.

2 The calculation of the assessment criteria for DIN and TP in Adriatic Sub-region

27. The scientific experience related to eutrophication in Adriatic Sea is huge and relay on the problems derived from the eutrophic pressure connected with the Po River watershed where live around 16 000 000 inhabitants. Near the scientific experience also a huge data set exists that enabled development of TRIX (Volenweider et all., 1998[26]), an index for the assessment of the eutrophication, and a regional approach for development of classification criteria based on Chlorophyll *a* within IMAP (Giovanardi et al, 2018).

28. This ensures further development of a harmonized approach to the definition of reference conditions and boundary values for DIN and TP based on the relationship between pressure and responses. To that effect the necessary steps are presented here below, whilst detail elaboration of this approach is provided in UNEP/MED WG.492/Inf.12 submitted for information of present Meeting.

2.1 Water typology

29. The Water typology is very important for further development of classification schemes of a certain area. The major coastal water types and related criteria in the Mediterranean were defined following on their inter calibration, that was applicable for phytoplankton only, as provided in Decision IG.22/7 on IMAP (COP 19, 2016[27]).

30. The water typology, a parameter having a robust numerical basis, can describe the dynamic behaviour of a coastal system. The assessment criteria are built per Water types that are mainly focused on hydrological parameters and characterization of water bodies' dynamics and circulation/ They are based on the introduction of density, the static stability parameter (derived from temperature and salinity values in the water column) for characterising of water bodies.

31. The first step in setting reference conditions and boundary values for an area i.e. Adriatic Sea sub-region is to identify present Water types and to attribute the data related to the density or salinity boundaries (Table 3).

Table 3. Major coastal water types relevant for Adriatic Sea with density and salinity boundaries

	Type I	Type IIA, IIA Adriatic	Type IIIW
σ_t (density)	<25	25<d<27	>27
S (salinity)	<34.5	34.5<S<37.5	>37.5

2.2 Reference condition

32. Reference Conditions (RCs) represent "a description of the biological quality elements that exist, or would exist, at high status". That is, with no, or very minor disturbance from human activities.

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[25] The Mediterranean sub-regions and subareas are initially proposed according to availability of database sources for calculation of the assessment criteria for contaminants (UNEP(DEPI)/MED WG.427/Inf.3; UNEP/MED WG.463/8; UNEP/MED WG.467/7).

[26] Vollenweider, R.A., F. Giovanardi, G. Montanari, A. Rinaldi, (1998). Characterization of the Trophic Conditions of Marine Coastal Waters. Environmetrics, 9, 329-357.

[27] COP 19. (2016). Decision IG.22/7 - Integrated Monitoring and Assessment Programme (IMAP) of the Mediterranean Sea and Coast and Related Assessment Criteria. COP19, Athens, Greece. United Nations Environment Programme, Mediterranean Action Plan, Athens.

The objective of setting reference conditions` standards is to enable the assessment of ecological quality against these standards (WFD CIS Guidance Document No. 5 (2003[28])).

33. An acceptable approach is to use a comprehensive pressure indicator that is able to address the potential transport of nutrients (natural loads plus anthropogenic loads) from the mainland to the sea, and that also measure, albeit roughly, this transport verifying the eventual absence of pressures of some importance exerted by human activities. For this purpose, use of dilution factor is considered as it was the case when the RCs for the Adriatic and Tyrrhenian Sea were developed (Giovanardi *et al.*, 2018).

34. The dilution factor is formulated as follows: F_dil=[(S-s)/S]*100, where S = open sea salinity, s = measured salinity at a given coastal sampling point (Giovanardi and Vollenweider, 2004[29]).

35. The role of the F_dil factor in assigning the chlorophyll *a* RCs is depicted in Figure 1.

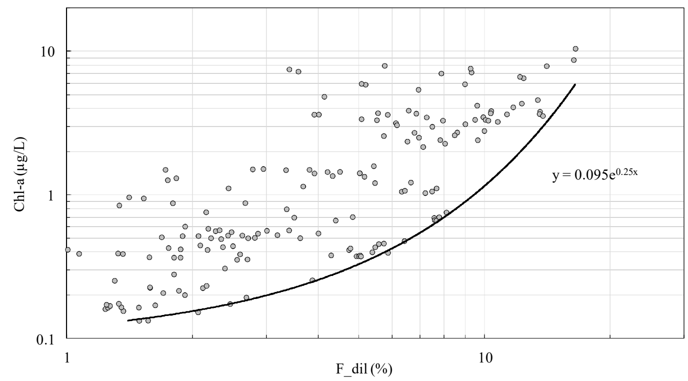

Figure 1. Scatter plot of annual *G_means* of chlorophyll *a* (Chl-a) against the dilution factor (F_dil) for Types I and II A. The curve marks the boundary of the lower limit of chlorophyll *a* reference conditions values (RCs). Original Figure from Giovanardi *et al,* 2018.

36. This separation line can be interpreted as the threshold between natural and anthropogenic pressures. It is assumed that the nutrient loads, either natural or generated by minor human activities, determine a response of the coastal systems that is well-represented by concentrations of chlorophyll *a* lying on the curve (Figure 1). Thus, the assessment of RCs does not derive from theoretical considerations or expert judgments, but refers to real situations occurring along the Adriatic coast.

37. The same approach cannot be used for the nutrients, given the dilution factor represents an integrated measure of the nutrient's pressures to the ecosystem. However, defining the reference conditions for chlorophyll *a* for different water types, precedes to setting of the reference conditions for nutrients, whilst the nutrients RCs will be derived from the pressure to effects relationship as presented here-below.

38. In order to define more accurately chlorophyll *a* RCs for each water type, the data corresponding to individual Adriatic types were considered separately. Then it was possible to plot the curves separately for all types (Figure 2), which represent the RCs for each type.

[28] WFD CIS Guidance Document No. 5 (2003) Transitional and Coastal Waters Typology, Reference Conditions and Classification Systems.
[29] Giovanardi, F., Vollenweider, R.A., (2004). Trophic conditions of marine coastal waters: experience in applying the Trophic Index TRIX to two areas of the Adriatic and Tyrrhenian seas. J. Limnology 63, 199–218.

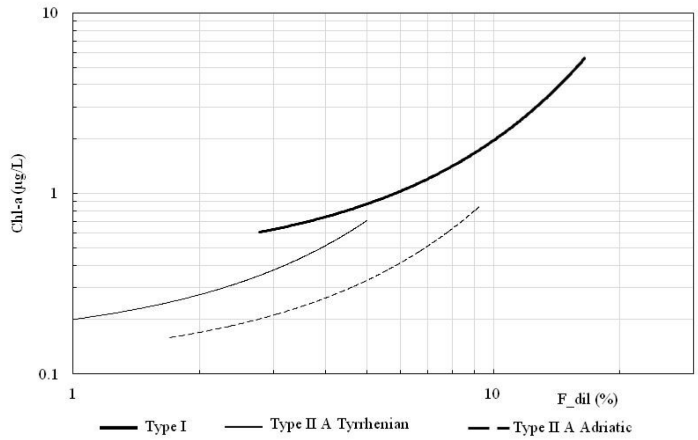

Figure 2. Reference conditions for chlorophyll *a* (Chl a) corresponding to different water types, depending on the gradient of the dilution factor (F_dil). Original Figure from Giovanardi *et al,* 2018.

39. The best functional relationships between chlorophyll *a* RC and F_dil were always exponential. The equations describing these relationships have been used to derive a unique chlorophyll *a* RCs per water types corresponding to the mean value of F_dil. Table 4 summarizes the results.

Table 4. Summary table for BQE phytoplankton reference conditions (RC) based on chlorophyll *a*.

Type	Functional relationships	F_dil (%) *Mean value*	RC - Chl-a (µg/L) as *G_Mean*
Type I	$y = 0.388\ e^{0.162x}$	7.9	1.40
Type II A Adriatic	$y = 0.109\ e^{0.221x}$	4.96	0.33

2.3 Pressure to effect relationship

40. Defining pressure to effect relationship is critical for nutrients RCs setting. Furthermore, a complete understanding of the functional relationship which links pressures to ecological effects result at the end with the programmes and measure as the final goal of the assessment process. To define the pressure to effect relationship, there is a need to apply relevant statistical analyses.

41. First the sensitivity of the selected metrics to different pressure indicators, with multiple regression analysis with linear models (LMs), was performed. By means of this stepwise regression technique, the variations of chlorophyll *a* concentration were tested against the pressure indicators available in dataset for Adriatic Sea (i.e. nutrient concentrations, oxygen saturation, dilution factor and Secchi depth). Annual geometric means of the parameters were used for this analysis.

42. The statistical analyses were performed using statistical packages offered by the program R and the exact statistical procedure is explained in UNEP/MED WG.492/Inf.12[30] submitted for information of present meeting.

43. Data processing involved the use of techniques of regression analysis provided by the package *stats*. For Type I among all the possible combinations, the stepwise regression technique provided the following linear model:

lm (formula = Chl-a ~ F_dil + aD_O + TP + DIN, data = Type_I)

44. The fitted linear model explains 89% of the total chlorophyll *a* variability and the maximum weight in determining this variability accounts to TP.

[30] UNEP/MED WG.492/Inf.12. Analysis of the Methodologies Available for Establishment of the Assessment Criteria for IMAP Common Indicator 13.

45. For Type II A coastal water the linear model provided by the stepwise regression technique was:

lm (formula = Chl-a ~ F_dil + TP, data = Type_II A)

46. The linear model is quite simple. Only two regressors were chosen with a largely dominant weight of TP over the weight of F_dil and the amount of chlorophyll *a* variability explained by this model is 78%. As TP accounts for the maximum weight in determining the variability of chlorophyll *a*, for both Type I and Type II A Adriatic, this parameter can be considered as the most eligible indicator of the pressure gradient. In this case the phosphorus pool in the water column (TP) can be considered as an internal measure of external phosphorus enrichment.

47. The above calculated relationships showed that chlorophyll *a* sensitivity, considered as the response of coastal systems to the availability of nutrients in terms of phytoplankton biomass production, is largely controlled by total phosphorus, which can therefore assume the role of the main pressure indicator.

48. The important regression equations used subsequently for the construction of the ecological classification criteria are summarized in Table 5.

Table 5. List of functional relationships of interest per water types. For each regression equation, the sample size N and the R-squared values are provided.

Functional link	Type I	Type II A Adriatic
1.TP vs TRIX	[TP] = exp [(TRIX – 6.064)/1.349] N = 15	[TP] = exp [(TRIX – 6.148)/1.583] N = 52
2. Chl-a *vs* TP	[Chl-a] = 10.591 $[TP]^{1.237}$ N = 15; $R^2 = 0.835$; $P = 4.45\ 10^{-6}$	[Chl-a] = 3.978 $[TP]^{1.347}$ N = 52; $R^2 = 0.896$; $P = 2.2\ 10^{-16}$

49. The nature of these relationships is almost always *log-log* type, which provides the highest degree of correlation. The equations in row 1 were obtained from the inverse relationship between the TRIX index and its component TP. For Type I and II A Adriatic these equations were prepared separately per water type, using the same data as those used to assess the functional relationships between TP and chlorophyll *a*. Finally, equations in row 2 exploit the relationship between TP and chlorophyll *a*, with the aim of fixing the limits among the ecological quality classes of the classification criterion, both for RCs and boundaries values.

50. The DIN was not elaborated further as the stepwise regression (i.e. the linear models) showed that it is not explaining the variability of the chlorophyll *a* and precise boundaries for DIN cannot be set.

2.4 Boundaries setting

51. With the definition of nutrients` RCs for Type I and Type II A coastal waters and the unveiling of their pressure-impact relationships, all the necessary tools are provided for defining the classification criteria for Biological Quality Element (BQE) phytoplankton in Adriatic coastal waters. Given the Trophic Index (TRIX, Vollenveider et al, 1998) was developed first for the northern Adriatic and it ecological use is well known, it was used as an internal scale in setting the boundaries.

52. The first step was to calculate the RCs for type I and Type II Adriatic from the functional relationship between Chla and TP (Table 5, row 2) and resulting in 0,19 μmol/L and 0,16 μmol/L, respectively.

53. The next in setting the boundaries was the definition of the most important boundary i.e. the Good/Moderate (G/M) boundary, which delimits the need for taking measures in case of good ecological status failure. Firstly, the boundary was set for TP, as it appeared to be the best pressure indicator for phytoplankton as explained above. The G/M boundary for TP was calculated using the equations in row 1 of Table 5, at the corresponding TRIX boundary between Good and Mediocre Trophic Status (TRIX = 5; Giovanardi et al, 2018), which matches the transition from mesotrophic to eutrophic conditions in the coastal ecosystem.

54. This boundary was used for Type II A Adriatic Sea giving the values of 0.48 µmol/L. For Type I, the value of TRIX for deriving the G/M boundary was increased to 5.25, in order to take into account the nutrient loads originating from natural sources carried by the Po River into the Adriatic Sea, presumably in not negligible amounts. In this way, the G/M boundary for TP was set at 0.55 µmol/L for Type I. In the same manner all boundaries` values for Types I and II A Adriatic were calculated (Tables 6 and 7).

55. The identified P/B boundaries refer to "virtual" conditions, since it was not possible to detect real situations related to ecological class "Bad" in any of the datasets analysed in this work. TP concentrations characterizing "Bad" ecological class have been extrapolated from the functional relationships extended to the area of the diagrams not actually covered by observations. It is impossible to predict how coastal systems would behave with such high concentrations of phosphorus, especially since annual averages need to be determined. Therefore, this class is considered as indicative, but not strictly necessary for proper ecological classification of the BQE phytoplankton based on TP concentration.

Table 6. Reference conditions and boundaries of ecological quality classes for BQE phytoplankton expressed by different parameters for Type I coastal waters.

Boundaries	TRIX	Chl-a annual *G_Mean*	TP annual *G_Mean*
		µg/L	µmol/L
Reference Conditions	-	1.40	0.19
H/G	4.25	2.0	0.26
G/M	5.25	5.0	0.55
M/P	6.25	12.6	1.15
P/B	7	25.0	2.00

Table 7. Reference conditions and boundaries of ecological quality classes for BQE phytoplankton expressed by different parameters for Type II A Adriatic coastal waters.

Boundaries	TRIX	Chl-a annual *G_Mean*	TP annual *G_Mean*
		µg/L	µmol/L
Reference Conditions	-	0.33	0,16
H/G	4	0.64	0.26
G/M	5	1.5	0.48
M/P	6	3.5	0.91
P/B	7	8.2	1.71

Type III W Adriatic

56. Following the same approach used for Type I and II A waters, overall G_means of nutrients` concentrations were related to the dilution factor for Type III W. No correlation was found for DIN (R2=0.05; P=0.303), while for the TP the relationship was even inverse to the one expected (Giovanardi et al, 2018). Additionally, overall values of G_mean of chlorophyll *a* range from around 0.1 to around 0.4 µg/L. Since the ecological classification scheme consists of 5 ecological quality classes, the discrimination limit between two contiguous chlorophyll *a* annual G_mean values would not be suitable for proper and safe classification (Giovanardi et al, 2018). For that reason, a single threshold value is therefore proposed for Type III W coastal waters that is the H/G value for Type IIA Adriatic of 0,26 µmol/L.

3 Other suggested methodologies for boundaries setting

3.1 The Best Practice Guide (BPG, Nutrient boundaries definition toolkit, JRC)

57. The document "*Best practice for establishing nutrient concentrations to support good ecological status*" is developed by the Joint Research Centre (JRC), the European Commission's science and knowledge service (Phillips et al, 2018). The purpose of the document is to help EU MSs achieve good ecological status (GES) in surface waters. It complements the Common Implementation

Strategy (CIS) Guidance document on eutrophication assessment in the context of European water policies (EC, 2009) by providing advice on how to link nutrient concentrations in surface waters to specific policy objectives. It can be used to check existing boundaries` values or to develop new ones. The guidance is supported by a toolkit in the form of an Excel workbook and a series of scripts which can be run using R, an open-source language widely used for statistical analysis and graphical presentation (R Development Core Team, 2016[31]). The toolkit provides the full R code, together with a series of examples which can be used to explore the methods.

58. This toolkit includes different statistical approaches to derive nutrients` boundaries, as elaborated here-below.

59. *Univariate linear regression:* Assuming a linear relationship between the ecological quality ratio (EQR) and nutrients, three regression types are implemented: two ordinary least squares OLS linear regressions between EQR and log nutrients concentration, where each variable is alternatively treated like the independent variable (because none of our two variables in practice can be considered to be free of error); and a third, type II regression, the ranged major axis (RMA) regression. The predicted range of nutrients` threshold values are then determined from the range of results obtained from these regressions' parameters.

60. *Logistic regression:* This approach treats ecological status as a categorical variable where a logistic model is fitted between categorical data using a binary response, "biology moderate or worse" = 1 or "biology good or better" = 0 and log of nutrient. Nutrient concentrations are determined where the probability of being moderate or worse was 0.5. In the case that additional pressures, other than nutrients, are suspected, a nutrient concentration value was determined at a probability of 0.75 instead of 0.5.

61. *Categorical methods:* Nutrient concentrations associated with a particular ecological status class could also be expressed as a distribution from which an upper quantile might be chosen to indicate a nutrient concentration above which good status was very unlikely to be achieved, or a lower quantile below which good status was very likely to be achieved (average of upper and lower quartiles of adjacent classes), so long as nutrients are the main driver of status. The average of the median of adjacent classes and the upper 75th percentile distribution are two additional categorical approaches tested.

62. *Minimisation of mismatch of classification:* Estimates the nutrient threshold value that minimizes the mismatch between status (good or better and moderate or worse) for the ecological and the supporting element.

63. *Linear quantile regression:* Useful alternative when the nutrient-biology interactions are confounded by other stressors, or environmental factors, leading to wedge-shape, or inverted- wedge, type of distributions. In such cases, the quantile regression allows different rates of change in the response variable to be predicted along the upper (in the presence of stressors) or lower (in the presence of mitigating environmental factors) quantiles of the distribution of the data (Cade and Noon, 2003[32]).Detailed information about the methods included in the toolkit is provided in the Guidance (Phillips et al., 2018).

3.2 Experience of Spain in establishment of nutrient boundary values for coastal waters of Catalonia

64. The FAN (Phosphate-Ammonium-Nitrite) and FLU (FLUviality) indices method assesses the physicochemical state of coastal waters and allows nutrient boundary values to support GES to be established. This method is based on a distinctly different process to establish these values than those described in this document. Rather than using nutrient and BQE data simultaneously, it assesses the

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[31] R Development Core Team (2006). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

[32] Cade, B.S. and Noon, B.R. (2003). A gentle introduction to quantile regression for ecologists. Frontiers in Ecology and the Environment 1: 412-420.

physicochemical state of coastal waters and then it relates this to the BQE. Nutrients` boundary values are then established from this relationship. This approach considers several dissolved inorganic nutrients concentrations and their stoichiometry at the same time rather than focusing on a single nutrient, as is the case when applying the toolkit.

65. The FAN and FLU indices method was developed using the physicochemical database of the Catalan Coastal Water Monitoring Programme. The data are representative of the north-west Mediterranean and comprise 20,102 records from 268 sampling stations collected between 1994 and 2014. A factorial analysis performed with this database revealed that the main pressures impacting coastal waters are *continental influences* (CI), which are related to gradients of dissolved inorganic nutrients, and freshwater content (inverse of salinity). An assessment of the physicochemical state of coastal waters based on the CI yielded results nearly equivalent (correlation of 0.93) to those obtained with the Trophic Index (TRIX) of Vollenweider et al. (1998). A further rotation applied to the factorial analysis revealed that CI is divided into two distinct gradients: levels of dissolved inorganic ammonium, phosphate, and nitrite define a gradient of urban influences while levels of dissolved inorganic silicate, and nitrate as well as the freshwater content, represent a gradient of freshwater influences or fluviality. The former is considered to reflect urban influences and the latter natural continental pressures on coastal waters (although freshwater influences are partly related to nitrate enrichment from agricultural sources).

66. These gradients of urban and freshwater influences were the basis for development of the FAN and FLU indices. The FAN index is scaled into five categories of water quality (high, good, moderate, poor and bad) and the FLU index into five categories of fluviality (very low, low, medium, high, and very high). The combined results provide a final assessment of the CI reaching coastal waters (urban, fluvial, mixed, or none) and, therefore, an assessment of their physicochemical state. The indices can be applied using data from inshore (0-200 m from the shore) or offshore (> 200 m from the shore) waters or both. The procedure, equations, and boundaries to apply the FAN and FLU indices together with detailed information on the method are available in Flo (2017[33]).

4 Elaboration of the assessment criteria i.e. reference conditions and boundary values for DIN and TP

67. Although setting of the assessment criteria for nutrients, transparency and oxygen was recognized as a minimum to address needs identified in 2017 Mediterranean Quality Status Report, data availability indicates that setting of RCs and boundary values is possible for DIN and TP only, and even for them in several sub-areas, but not at regional Mediterranean level.

68. To that effect the following datasets need to be provided as a minimum requirement: three continuous year of monitoring with a minimum monthly frequency for Water Type I; bimonthly frequency for Water Type II although preferable monthly; and seasonal frequency for Water Type III.

69. Further to analysis of monitoring data availability, the most suitable methods for setting the assessment criteria are recognized for each sub-area (Table 8). As explained above, data availability survey also indicates that data with unrestricted use from EMODnet may be used to supplement data sets available in IMAP Pilot Info System/MEDPOL.

Table 8. The Mediterranean sub-regions/subareas[34], where setting of the assessment criteria for DIN and TP is found possible in line with availability of monitoring data, along with relevant methods for setting of the boundary values.

Sub-regions	Sub-division (e.g. subareas/seas)	Availability of data (IMAP database + EMODNet)	Boundary setting method considered
Western Mediterranean Sea (WMS)	Alboran Sea (ALBS)	CPs contribution needed; data availability is restricted only to the northern part (Spain)	FAN/FLU, BPG toolkit
	North Western	Probably sufficient; CPs should	Methodology for Adriatic

[33] Flo, E. (2017). Opening the black box of coastal inshore waters in the NW Mediterranean Sea: environmental quality tools and assessment. PhD. 372 pages.
[34] (UNEP(DEPI)/MED WG.427/Inf.3)

	Mediterranean Sea (NWMS)	further improve data availability	sub-region, BPG toolkit
	Tyrrhenian Sea (TYRS)		Methodology for Adriatic sub-region
	Western Mediterranean Islands and Archipelago (WMIA)	Insufficient data	-
Adriatic Sea (ADR)	North Adriatic (NADR) Middle Adriatic (MADR) South Adriatic (SADR)	Probably sufficient data available; Adriatic Sea needs to be considered as a whole; CPs should further improve data availabilit;	Methodology for Adriatic sub-region
Central Mediterranean (CEN)	Central Mediterranean (CEN) Ionian Sea (IONS)	Insufficient data	-
Aegean and Levantine Seas (AEL)	Aegean Sea (AEGS) Levantine (LEVS)	Probably sufficient data available for the northern part, only; CPs should improve data availability	BPG Toolkit, Methodology for Adriatic sub-region

70. The Online Working Group (OWG) on eutrophication needs to support present work through data collection, elaboration and application of the methods for setting boundary values including relevant statistical approaches.

71. The setting of RCs and boundary values for DIN and TP is the essential step in supporting application of present assessment criteria for chlorophyll *a,* as established by Decisions IG.22/7. The G/M threshold for DIN and TP is the most important boundary which delimits the need for taking measures in case of GES failure. The development of the boundary values for the full scale of the relationship between pressures and effects will also allow monitoring of mitigation measures.

72. Within the scope of present IMAP implementation, development of the assessment criteria for transparency and dissolved oxygen is premature mainly due to the fact that elements of the measurements presently unrelated to primary production (Fleming-Lehtinen, 2016[35]) cannot be clearly identified. That is particularly true for shallow and coastal areas that are the most of our interest.

73. This proposal for setting the assessment criteria for DIN and TP within implementation of IMAP CI 13 corresponds well with the most recent findings related to EU WFD and MSFD implementation (Salas Herrero et al, 2020[36]). These findings refer to comparison between the information reported by EU MSs to WISE on the standards for general physico-chemical quality elements, including nutrients and the information on methodologies used to assess eutrophication in coastal waters in accordance with MSFD, with the methodological standards agreed and used for the assessment of the elements for eutrophication criteria (i.e. nutrient conditions, transparency, and dissolved oxygen) at regional sea level.

[35]Fleming-Lehtinen, V. (2016). Secchi depth in the Baltic Sea – an indicator of eutrophication. University of Helsinki, Faculty of Biological and Environmental Sciences, Helsinki. 42 pages.
[36] Salas Herrero, F., Aráujo, R., Claussen, U., Leujak, W., Boughaba, J., Dellsaea, J., Somma, F., Poikane, S,. (2020). Physico-chemical supporting elements in coastal waters: Links between Water and Marine Strategy Framework Directives and Regional Sea Conventions. EUR 30383 EN, Publications Office of the European Union, Luxembourg.

Annex I
References

Bethoux, J.P., Morin, P., Madec, C. and Gentili, B. (1992). Phosphorus and nitrogen behaviour in the Mediterranean sea. Deep-Sea Res 39: 1 641-1 654.

Borja, A., Elliott, M., Henriksen, P., and Marb, N. (2013). Transitional and coastal waters ecological status assessment: advances and challenges resulting from implementing the European water framework directive. Hydrobiologia 704, 213–229. doi: 10.1007/s10750-012-1276-9

Cade, B.S. and Noon, B.R. (2003). A gentle introduction to quantile regression for ecologists. Frontiers in Ecology and the Environment 1: 412-420.

Dafner, E.V., Boscolo, R. and Bryden, H.L. (2003). The N:Si:P molar ratio in the Strait of Gibraltar. Geophysical Research Letters 30: 1 506-1 509.

COP 19. (2016). Decision IG.22/7 - Integrated Monitoring and Assessment Programme (IMAP) of the Mediterranean Sea and Coast and Related Assessment Criteria. COP19, Athens, Greece. United Nations Environment Programme, Mediterranean Action Plan, Athens.

Devlin, M., Best, M., Coates, D., Bresnan, E., O'Boyle, S., Park, R., et al. (2007). Establishing boundary classes for the classification of UK marine waters using phytoplankton communities. Mar. Pollut. Bull. 55, 91–103. doi: 10.1016/j. marpolbul.2006.09.018

Dworak, T., Berglund, M., Haider, S., Leujak, W. and Claussen, U. (2016). A comparison of European nutrient boundaries for transitional, coastal and marine waters. Working Group on ecological Status ECOSTAT.

European Commission [EC] (2009). Guidance Document on Eutrophication Assessment in the Context of European Water Policies. Common Implementation Strategy Guidance Document No. 23. Luxembourg: Office for Official Publications of the European Union.

Estrada, M. (1996). Primary production in the Northwestern Mediterranean. Sci. Mar. 60, 55–64.

Fleming-Lehtinen, V. (2016). Secchi depth in the Baltic Sea – an indicator of eutrophication. University of Helsinki, Faculty of Biological and Environmental Sciences, Helsinki. 42 pages.

Flo, E. (2017). Opening the black box of coastal inshore waters in the NW Mediterranean Sea: environmental quality tools and assessment. PhD. 372 pages. https://www.tdx.cat/handle/10803/461378 (date accessed: 2.10.2018)

Garmendia, M., Borja, Á, Franco, J., and Revilla, M. (2013). Phytoplankton composition indicators for the assessment of eutrophication in marine waters: present state and challenges within the European directives. Mar. Pollut. Bull. 66, 7–16. doi: 10.1016/j.marpolbul.2012.10.005

Giovanardi, F., Francé, J., Mozetič, P., Precali, R. (2018). Development of ecological classification criteria for the Biological Quality Element phytoplankton for Adriatic and Tyrrhenian coastal waters by means of chlorophyll a (2000/60/EC WFD). Ecological Indicators. 93. 316-332. doi: 10.1016/j.ecolind.2018.05.015.

Giovanardi, F., Tromellini, E., (1992). An empirical dispersion model for total phosphorus in a coastal area: the Po River-Adriatic system. Sci. Total Environ. Supplement 201–210. doi:10.1016/B978-0444-9990-3.50022-5.

Giovanardi, F., Vollenweider, R.A., (2004). Trophic conditions of marine coastal waters: experience in applying the Trophic Index TRIX to two areas of the Adriatic and Tyrrhenian seas. J. Limnology 63, 199–218. doi: 10.4081/jlimnol.2004.199.

Helcom (2009). Eutrophication in the Baltic Sea — An integrated thematic assessment of the effects of nutrient enrichment and eutrophication in the Baltic Sea region. Baltic Sea Environment Proceedings No 115B.

Hering, D., Borja, A., Carstensen, J., Carvalho, L., Elliott, M., and Feld, C. K. (2010). The European water framework directive at the age of 10: a critical review of the achievements with recommendations for the future. Sci. Total Environ. 408, 4007–4019.

IMAP (2017). Integrated Monitoring and Assessment Programme of the Mediterranean Sea and Coast and Related Assessment Criteria UNEP, Athens, 52 pp.

Krom, M. D., Kress, N., and Benner, S. (1991). Phosphorus limitation of primary productivity in the eastern mediterranean sea. Limnol. Oceanogr. 36, 424–432. doi: 10.4319/lo.1991.36.3.0424

Lazzari, P., Solidoro, C., Salon, S. and Bolon, G. (2016). Spatial variability of phosphate and nitrate in the Mediterranean Sea: a modeling approach. Deep Sea Research Part 1: Oceanographic Research Papers 108: 39-52. doi: 10.1016/j.dsr.2015.12.006

Phillips G, Kelly M, Teixeira H, Salas F, Free G, Leujak W, Pitt Ja, Lyche Solheim A, Varbiro G, Poikane S, (2018) Best practice for establishing nutrient concentrations to support good ecological status, EUR 29329 EN, Publications Office of the European Union, Luxembourg, ISBN 978-92-79-92906-9, doi:10.2760/84425, JRC112667.

R Development Core Team (2006). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0. http://www.R-project.org

Ramírez. T., Cortés, D., Mercado, J.M., Vargas-Yáñez, M., Sebastián, M.,. Liger, E. (2005) .,Seasonal dynamics of inorganic nutrients and phytoplankton biomass in the NW Alboran Sea Estuar. Coast. Shelf Sci., 65 (4), pp. 654-670, 10.1016/j.ecss.2005.07.012

Reyjol, Y., Argillier, C., Bonne, W., Borja, A., Buijse, A. D., Cardoso, A. C., et al. (2014). Assessing the ecological status in the context of the European water framework directive: where do we go now?. Sci. Total Environ. 497, 332–344. doi: 10.1016/j.scitotenv.2014.07.119

Rinaldi, A. (2014). Fioriturі algali In Adriatico. Il bacino padano-adriatico tra sviluppo e scienza (Algal blooms in the Adriatic. The Padano-Adriatic basin between development and science) Editrice La Mandragora.

Salas Herrero, F., Aráujo, R., Claussen, U., Leujak, W., Boughaba, J., Dellsaea, J., Somma, F., Poikane, S,. (2020). Physico-chemical supporting elements in coastal waters: Links between Water and Marine Strategy Framework Directives and Regional Sea Conventions. EUR 30383 EN, Publications Office of the European Union, Luxembourg. ISBN 978-92-76-22418-1, doi:10.2760/444388, JRC121759.

Tett, P., Gilpin, L., Svendsen, H., Erlandsson, C. P., Larsson, U., Kratzer, S., et al. (2003). Eutrophication and some European waters of restricted exchange. Cont. Shelf Res. 23, 1635–1671. doi: 10.1016/j.csr.2003.06.013

Teichberg, M., Fox, S. E., Olsen, Y. S., Valiela, I., Martinetto, P., and Iribarne, O. (2010). Eutrophication and macroalgal blooms in temperate and tropical coastal waters: nutrient enrichment experiments with Ulva spp. Global Change Biol. 16, 2624–2637.

Thingstad, T.F., Krom, M.D., Mantoura, R.F.C., Flaten, G.A.F., Groom, S., Herut, B., Kress, N., Law, C.S., Pasternak, A., Pitta, P., Psarra, S., Rassoulzadegan, F., Tanaka, T., Tselepides, A., Wassmann, P., Woodward, E. M. S., Wexels Riser, C., Zodiatis, G. and Zohary T. (2005). Nature of phosphorus limitation in the ultraoligotrophic Eastern Mediterranean. Science (New York) 309: 1 068-1 071.

Tsirtis, G.E. (1995). A simulation model for the description of a eutrophic system with emphasis on the microbial processes. Water Science and Technology 32: 189 -196.

Tyrrell, T. (1999). The relative influences of nitrogen and phosphorus on oceanic primary production. Nature (London) 400: 525-531.

UNEP/MED WG.492/Inf.12. Analysis of the Methodologies Available for Establishment of the Assessment Criteria for IMAP Common Indicator 13.

Vollenweider, R.A., F. Giovanardi, G. Montanari, A. Rinaldi, (1998). Characterization of the Trophic Conditions of Marine Coastal Waters. Environmetrics, 9, 329-357.COP

WFD CIS Guidance Document No. 5 (2003) Transitional and Coastal Waters Typology, Reference Conditions and Classification Systems.