



HarmoNIA

Harmonization and Networking for  
contaminant assessment in the Ionian and  
Adriatic Seas

## Deliverable T1.1.2 Methodological proposal for harmonized sampling procedure, analytical methodologies

# Work Package T1 - Sharing best practices for transnational harmonization for EQSD implementation and link with UNEP/MAP MEDPOL Program and Offshore Protocol of Barcelona Convention

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## 1. Introduction

In relation to the heterogeneity in monitoring procedures highlighted in the framework of Interreg ADRION HarmoNIA activities, deliverable T1.1.2 required defining the best practices of sampling procedures, analytical methodologies and QA/QC procedures regarding selected pollutants in water, sediment and biota, in line with the provision of EQSD (Environmental Quality Standards Directive) and of the indications of UNEP/MAP Working Groups on Pollution Monitoring. This Methodological Proposal addresses harmonization of monitoring procedures for the assessment of the *good environmental status* (GES) required by Marine Strategy Framework Directive 2008/56/EC and UNEP/MAP Integrated Monitoring and Assessment Programme (IMAP) for the implementation of the Ecosystem Approach (EcAp) environmental objectives. Actually, both MSFD and EcAP require harmonized and integrated approach for water quality assessment at the regional and subregional level but, until now, MSFD relies mainly on the results of Water Framework Directive which in turn is focused on inland water (EQSD threshold values) and EcAP process has just started with the IMAP indicator development still to be completed. As a consequence, the harmonization of the monitoring procedures for GES assessment is still an open issue in the Mediterranean Region and, in particular, in the Adriatic and Ionian sub region, with some obvious gaps such as the lack of common threshold values for sediment matrix which have not been included in EQSD or the absence of common procedures for basin level assessment in UNEP/MAP as most of the work done was based on MEDPOL monitoring programme mainly focused on hot spots.

## 2. Report on the comparison of QA/QC procedures on collected data from questionnaire and proposal of the best practice of selected pollutants

In order to define the common protocols and to propose the best QA/QC procedures for each contaminant, we compared the information obtained with the questionnaire by the 9 participants, for water, sediments and biota matrices (HarmoNIA Deliverable T1.1.1).

On the basis of the outcomes of Deliverable T1.1.1, the main targets of harmonization are:

- Matrix characterization
- Measurement units
- Sampling procedures
- Indications on sample storage and methods of analysis



- Indications on LOQ

The methodological proposals are presented in Annexes I to III.

Generally, about the QA/QC procedures we have **proposed the procedures reported in European Directives** (LOQ, method of analysis, etc.), when possible. For compounds not included in the Directives, we suggest methods that allow to achieve the comparability of the results among different laboratories and having LOQ that ensure a good environmental protection standard, considering their potential effects on biota matrix.

It must be taken into account that for some groups of contaminants and/or for some single pollutants, participants reported no QA/QC data. Furthermore, in some cases, even if participants reported that they have carried out the analyses, they have not given complete information about the requested QA/QC procedures.

We suggest to perform all activities, from sampling to chemical analyses, according to quality assurance prescriptions as indicated ISO/IEC 17025. Its indications on traceability of each of these steps could advantage the harmonization of the procedures and favor the comparability among different laboratories, even if it will not ensure *a priori* a better analytical performance.

## 2.1 Documents of reference

In the framework of the Barcelona Convention (under Mediterranean Action Plan (MAP) and Regional Seas Programme of the United Nations Environment Programme (UNEP)), the HarmoNIA project focuses on multilateral scientific and technological cooperation related to the stipulation of monitoring plans and the management of environmental contamination emergencies. In the context of the harmonization of the analytical methods, in particular in order to define the best reference LOQ to propose to the HarmoNIA participants for each contaminant in different matrices, the following documents were considered as references:

- European Directive (2013/39/EU)
- Commission Directive 2009/90/EC (on technical specifications for chemical analysis and monitoring of water status)
- Technical Report 2010 - 041 of the Common Implementation Strategy for the WFD
- UNEP (DEPI)/MED WG.427/inf.3 2016 “Agenda item 5. Review of Proposed Background Concentrations (BC)/Background Assessment Criteria (BACs)/Environmental Assessment Criteria (EACs) for Contaminants and Biomarkers at Mediterranean and Regional Scales Proposal of Assessment Criteria for Hazardous Substances and Biological Markers in the Mediterranean Sea Basin and its Regional Scales”



- JRC technical reports (2019) “Marine chemical contaminants - support to the harmonization of MSFD D8 methodological standards”.
- Commission Regulation (EC) No 1881/2006 and amendments.

## 2.2 LOQ proposal references

For marine chemical contaminants, the threshold values to consider for the scope of the harmonization proposal for LOQ are:

- EU-wide Environmental Quality Standard (EQS) laid down in part A of annex I to Directive 2008/105/EU as amended by Directive 2013/39/EU. The EQS is the concentration of a contaminant, which should not be exceeded to protect human health and environment. EQS are set as a maximum allowable concentration (MAC-EQS) or an annual average (AA-EQS), protecting aquatic organisms from acute and chronic effects, respectively. For water, the EQS is expressed as total concentration in the whole water, except in the case of cadmium, lead, mercury and nickel, where the water EQS refers to dissolved concentration (sample filtered at 0.45 µm filter or equivalent pre-treatment). For biota, depending on the bioaccumulation potential of pollutants and the protection goal, the EQS<sub>biota</sub> concern the concentration in mussel or fish at trophic level 4 or 4.5. Moreover, in order to account for differences in the lipid content, the concentration of pollutants should be normalized to lipid content (5% for fishes and 1% for mussels) before comparison with EQS<sub>biota</sub>.
- National EQS established according to the procedure described in the WFD guidance document no. 27 (European Commission, 2011 and 2018 revision). Alternatively, to water EQS, sediment or biota EQS can be set, as long as they provide at least the same level of protection.
- Background Concentrations (BCs) developed by Convention for the Protection of the Marine Environment of the North-East Atlantic (Oslo Paris Convention, OSPAR), defined as “the concentration of a contaminant at a ‘pristine’ or ‘remote’ site based on contemporary or historical data” (OSPAR Agreement 2005-6), considered close to zero for organic contaminants and at naturally occurring concentrations for metals. In order to facilitate precautionary assessments of data collected under the OSPAR Coordinated Environmental Monitoring Programme (CEMP) against BCs, OSPAR has developed Background Assessment Concentrations (BACs). Mean concentration below the BACs are said to be near background. However, given that BACs do



not consider ecotoxicological aspects, OSPAR has developed Environmental Assessment Criteria (EACs) which represent the contaminant concentration in the environment, below which no chronic effects are expected to occur in marine species, including the most sensitive ones. OSPAR BACs and EACs are often used in tandem in assessment approaches.

- Mediterranean BACs (Med BACs): as a preliminary proposal regarding the Mediterranean BACs for major chemical pollutants (in sediment and biota) and biomarkers, it was recommended to use (as a first step) a number of BAC and EAC values, either adopted by OSPAR and/or developed by scientific studies in the Mediterranean Sea (UNEP(DEPI)/MED WG.365/Inf.8, and the references cited therein). This was made in order to adjust (or to develop) an appropriate threshold assessment criteria for the Mediterranean Sea region. The first estimates of Mediterranean background concentrations (BCs) and both background and environmental assessment criteria (BACs/EACs) were made for trace metals in sediments and biota and PAHs in sediments in 2011, following the OSPAR methodology approach (UNEP(DEPI)/MED WG.365/Inf.8). Later in 2014, an informal online expert group on contaminants was established: its first report on assessment criteria was delivered in March 2015 and it was discussed at the MEDPOL Focal Points Meeting in June 2015 (UNEP(DEPI)/MED WG.417/7, 2015).

The calculation of the Med BACs has been performed for the Mediterranean Sea as a whole with the calculated Med BCs (either corresponding to the median (50th percentile) for hazardous chemical substances, or the 10th or 90th percentile for selected biomarkers), including BCs assessed for Mediterranean eco-regions and sub-regional seas (UNEP(DEPI)/MED WG.427/7, 2016; UNEP/MED WG.463/Inf.6, 2019). We refer to Med BACs threshold values referred to whole Mediterranean Sea, even if specific BACs for Adriatic, Aegean-Levantine and Western Mediterranean Seas were assessed.

In biota, we suggest to use the EAC values adopted within OSPAR and the European Commission Regulations (2006/1881/EC and 2008/629/EC ).

- Effects Range-Low (ERL) values developed by the United States Environmental Protection Agency (US EPA) for assessing the ecological significance of sediment concentrations (JRC technical reports, 2019). ERL is the lower tenth percentile of the data set of concentrations in sediments, which were associated with biological



effects. Adverse effects on organisms are rarely observed when concentrations fall below the ERL value. ERL values used as reference were 2.5 % total organic carbon normalized (UNEP(DEPI)/MED WG.427/7, 2016).

Both Water Framework Directive (2000/60/EC) and Marine Strategy Framework Directive (2008/56/EC), as well as relative amendments and implementing decrees, focused on good environmental status (GES), taking into account the features regarding **water** and **biota**. In this context, one of the new contributions of HarmoNIA project is to harmonize the threshold values for **sediment** contamination considering the legislation of participating partners.

Besides the threshold values and assessment criteria agreed at EU or regional level, MSFD have also indicated some national standards used for the assessment of some contaminants.

For Italy, the EQS in sediments were set in compliance with the WFD in marine-coastal and transitional waters for some substances (Legislative Decree 172/2015). These values have been derived based on direct risks for the benthic communities, but also for human health through seafood consumption. The ecotoxicological criterion used as a reference, agreed with the National Scientific Institutes, was the TEL (Threshold Effect Level) (Environment Canada and Ministère du Développement durable, de l'Environnement et des Parcs de Québec, 2007), the threshold value below which the adverse effects are expected to occur only rarely (Long and Mac Donald, 1998). For most substances, chemical and ecotoxicological data were collected along the Italian coast from 2001 to 2008.

The monitoring program of the state of coastal sea ecosystem of Montenegro in its structure and methodological approach is harmonized with the national regulation: Law on Environment (Official Gazette of Montenegro 48/08), Law on waters (Official Gazette of Montenegro 27/07), Decree on classification and categorization of surface and ground waters (Official Gazette of Montenegro 02/07), and with the requirements of relevant EU Directives, European Environment Agency Guide on transitional, coastal and marine waters (Eurowaternet technical guidelines, Cullingford et al., 2003), and related guidelines for reporting (WISE-SoE Reporting on Transitional, Coastal, and Marine Waters), as well as requirements of MEDPOL program.

There is no regulation for threshold values of contaminants in sediment in Montenegro. The international standards regulating impacts of sediments on human health and ecosystem are applied in the assessment of sediment quality (such as OSPAR Convention).

Water quality assessment, also in terms of marine chemical contaminants, is based on the criteria prescribed under the Decree on classification and categorization of surface and ground waters (Official Gazette of Montenegro 2/07).

The chemical monitoring in biota is regulated with:



- Law on Food Safety (Official Gazette of Montenegro 57/2015);
- Regulation on requirements for traceability of foodstuffs of 2016 (Official Gazette of Montenegro 48/16) which transposes the requirements of Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs;
- Rulebook on maximum level residua of pesticides in or on plants, plant products, food or feed (Official Gazette of Montenegro 21/15) which transposes Regulation (EC) No 396/2005 of the European Parliament and of the Council of 23 February 2005 on maximum residue levels of pesticides in or on food and feed of plant and animal origin and amending Council Directive 91/414/EEC.

Besides the national criteria, the decisions and recommendations of developed countries (such as OSPAR Convention criteria) are used.

The finalization of the Monitoring Programme of Marine Environment in accordance with the Barcelona Convention and IMAP recommendations is in progress. In line with harmonization with the EU legislation, it is expected that Government of Montenegro adopts the Law that will transpose MSFD, in the 2nd quarter of 2019 (Law on the Establishment of a Framework for the Protection of the Marine Environment in line with EU legislation in this area as well as with the Montenegrin legislation standards), but the detailed technical rulebooks in compliance with this regulation will be elaborated in the next 2-3 years (subordinate legislation related to MSFD parameters).

For Greece, threshold values for priority substances in waters comply with those set by the Directive 2013/39/EU. National standards exist for a list of 'specific pollutants' for surface inland waters (not included in the priority substances list) and are set by the Joint Ministerial Decree, Gov. Gazette no 1909/8-12-2010. These include 59 organic compounds and 9 metals (As, Sn, Co, Mo, Se, Cu, Cr<sup>+6</sup>, Cr tot, Zn). For sediments and biota no national standards are in force.

In Slovenia, threshold values for priority substances in water and biota comply with those set by the Directive 2013/39/EU. National standards also exist for a list of about 30 'specific pollutants' for marine waters (not included in the priority substances list). All the standards are set by the Decree on surface water status, Gov. Gazette no. 14/09, 98/2010, 96/2013, 24/2016. For sediments no national standards are in force.

According to JRC Technical report (2019), in Croatia, in compliance with the WFD, the substances monitored in the scope of the coastal and transitional waters monitoring programmes (in 2015 and 2017), will most likely be the basis for the 2018 MSFD reporting. However, no official decision has been made yet at the time of preparation of the 2019 report. Regarding sediments, any decision was made on the thresholds that will be used. The OSPAR sediment thresholds were applied in the previous GES assessment in 2012, but it was agreed that those values were not applicable for the specific regional conditions. For the purposes of the general assessment of the state of the environment in the scope of the WFD monitoring, the



sediment quality criteria from the Norwegian system for classification of sediments (Bakke et al., 2010) were used for sediment categorization.

## 2.1 Marine waters and biota

In order to normalize the concentration of pollutants, favoring the comparability of chemical data, we suggest to indicate the biometric measurements of the organisms used for chemical analyses. At this regard it would be also advisable to include in the procedure of analysis of the condition index of mussels and isotopic analysis aimed at the definition of trophic level on the base of what reported in the Common Implementation Strategy for the WFD (European Commission, 2010).

Regarding certified reference materials, we observed that, in particular for waters, all laboratories used in-house or spiked internal materials, except for heavy metals. We suggest the use of certified reference materials for all compounds in order to achieve the best methodological accuracy and data comparability. At this regard, given the difficulty to provide certified reference materials for water matrix, we suggest that all the laboratories should attend an intercalibration round (e.g. QUASIMEME, Aquacheck, IAEA etc.) for all the considered compounds, also in order to improve the analytical accuracy and reproducibility.

Then, a further proposal could be the implementation of analytical panels of pollutants, considering also that the European legislation is still developing, with the addition of other contaminants (such as the emerging ones) reaching the concentrations of environmental relevance.

As the basis for the proposal of LOQ for marine **waters** and **biota** we suggest to use Environmental Quality Standard (EQS) values reported in the 2013/39/EU Directive (as annual average values - AA-EQS for “other surface waters” column for waters, and EQS biota for biota).

It must be pointed out, that the Comm. Dir. 2009/90/EC (Art. 4), requires a LOQ value equal or below 30% of the EQS values reported in the 2013/39/EU Directive, considering the minimum performance criteria for analytical methods. These values should be considered in order to provide a more precautionary approach.

In the tables reported in annexes I, II, we indicate as the reference values EQS (2013/39/EU Directive, Italian Legislative Decree 172/2015 and, only for water, Greek Decree 1909/8-12-2010) and the LOQ proposal according to 2009/90/EC (in red color). For the compounds not included in the legislative references, we propose LOQ on the base of the results obtained by the questionnaire (values in black color) (Annexes I, II), considering the laboratories that gave the lowest values in order to obtain the best performance in terms of QA/QC and precautionary approach and for enhance the harmonization of threshold values.



Regarding the LOQ proposed for biota it must be considered that the EQS-biota values reported in the 2013/39/EU Directive are relative to fish species. The exceptions are dioxins and dioxin-like compounds where EQS refers to fish, crustaceans and molluscs, while for fluoranthene and PAH the EQS values refer to molluscs and crustaceans.

For total hydrocarbons, aliphatic hydrocarbons C<sub>6</sub>-C<sub>12</sub> and aliphatic hydrocarbons C<sub>10</sub>-C<sub>40</sub> in biota, it was not possible to indicate a LOQ proposal, as these contaminants are not mentioned in 2013/39/EU Directive and the range of the values reported by different participants was too wide or the number of data was insufficient.

In the table reported in Annex II, we reported as reference values also the OSPAR BAC, OSPAR EAC, MED BAC and values from Reg. 2006/1881/EC and its amendment 2008/629/EC.

None of the laboratories reported data about radionuclides and pharmaceuticals compounds. For the latter, we propose to provide data for these parameters on the base of the Commission Implementing Decision 2018/840/EU that indicate as maximum acceptable detection limit for macrolides (0.019 µg/l), amoxicillin (0.078 µg/l) ciprofloxacin (0.089 µg/l). Inventory of laboratories monitoring radionuclides was carried out in order to investigate data availability in the Adriatic - Ionian region.

## 2.2 Sediment

Sediment monitoring generally addresses the top layer of the sediment because this layer indicates the recently deposited material and the actual status of pollution. The main criterion for choosing the correct sediment sampling depth (the thickness of the sediment layer sampled) in a water body is the knowledge of the deposition rate of the sampling site. In theory, the lower the deposition rate, the thinner the layer that one might be interested to sample. In fact, if the sedimentation rate is 0.2 mm/year (in the offshore zones), 1 cm is deposited in 50 years. Close to coast, 1-2 mm/year of sedimentation rate is very likely, thus 1 cm means 5 years. The depletion of oxygen occurs in 1-2 cm layer, which is the most active sediment layer. Regarding the redox conditions, the upper 1-2 cm are optimal for sediment finer than coarse sands. In coarser sediments like pebbles, it is not relevant, because there is not fine fraction to adsorb the pollutants. In practice, it is recommended to sample the top layer of the sediment, from 0 to 2 cm depth, depending on the deposition rate in order to appreciate the recent temporal trends. It is also recommended to measure sedimentation rate to select the more appropriate sediment depths, eventually thinner than 2 cm. Box corer was chosen as the most appropriated tool for the soft sediment sampling, as it is designed to minimize the sample disturbance. To improve the comparability, it is better to consider the grain size fraction < 2 mm, because in sandy sediments the



fraction < 63  $\mu\text{m}$  would represent a negligible part, not representative of the bulk sediments.

We propose LOQ on the base of the results obtained by the HarmoNIA partnership, considering the laboratories that reported the lowest values in order to obtain the best performance in terms of QA/QC and precautionary approach.

With the aim to provide a common indication on LOQ values to be adopted in sediment, in Annex III the following reference values were reported: Med BACs, US-ERL, Threshold Effect Level (TEL) from Canadian Environmental Quality Standard (Environment Canada and Ministère du Développement durable, de l'Environnement et des Parcs di Quebec, 2007) and EQS reported in the Italian Decree 172/2015. In addition, the recent review carried out at EU scale (JRC Technical Report, 2019) was taken in consideration. The legislative references of different partners were not included in the Annex III because they declared, as described above, that there are no national standards for sediments.

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