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**Agenda item 3: Thematic Decisions**

**Pollution Assessment Criteria and Thresholds**

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### List of Acronyms and key definitions

<b>ADR</b>	Adriatic Sea eco-region
<b>AEL</b>	Aegean and Levantine Seas
<b>BAC</b>	Background Assessment Criteria
<b>BC</b>	Background Concentration
<b>CEN</b>	Central Mediterranean Sea
<b>EAC</b>	Environmental Assessment Criteria
<b>GES</b>	Good Environmental Status
<b>IQR</b>	Interquartile range as a measure of the data dispersion (non-parametric distributions)
<b>Mean</b>	Arithmetic mean as central tendency estimator
<b>Med BAC</b>	Mediterranean BAC (MEDPOL datasets)
<b>Median</b>	Midpoint value (50th percentile) of the datasets
<b>Med BC</b>	Mediterranean BC (MEDPOL datasets)
<b>N</b>	Number of individual data
<b>Percentile(s)</b>	Value(s) below a given % of data of the ordered datasets can be found
<b>WMS</b>	Western Mediterranean Sea eco-region
<b><math>\lambda</math> (%)</b>	% of data of the aggregated datasets in the primary normal component
<b>Cd</b>	Total cadmium
<b>Hg/HgT</b>	Total mercury
<b>Pb</b>	Total lead
<b>PAHs</b>	Polycyclic Aromatic Hydrocarbons (group of petroleum hydrocarbons compounds)
<b>N</b>	Naphthalene
<b>ACE</b>	Acenaphthene
<b>ACY</b>	Acenaphthylene
<b>FL</b>	Fluorene
<b>P</b>	Phenanthrene
<b>A</b>	Anthracene
<b>PY</b>	Pyrene
<b>BaA</b>	Benz(a)anthracene
<b>C</b>	Crysene
<b>BaPY</b>	Benzo(a)Pyrene
<b>GHI</b>	Benzo[g, h, i]perylene
<b>DA</b>	Dibenz[a,h]anthracene
<b>ID</b>	Indene[123-c,d]pyrene
<b>OCs</b>	Organochlorinated Compounds (group of compounds including PCBs and Pesticides)
<b>PCBs/CBs</b>	Polychlorinated biphenyl ethers
<b>PCBs/CBs</b>	Polychlorinated biphenyl ethers
<b>HCH</b>	Hexachlorocyclohexane
<b>DDE</b>	Dichlorodiphenyldichloroethylene
<b>HCB</b>	Hexachlorobenzene
<b>AChE</b>	Acetylcholinesterase biomarker
<b>MT</b>	Metallothioneins biomarker
<b>MN</b>	Micronuclei Frequency
<b>LMS</b>	Lysosomal Membrane Stability
<b>SoS</b>	Stress on Stress

## **Note by the Secretariat**

1. 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 (Oslo Paris Convention) methodology approach (UNEP(DEPI)/MED WG.365/Inf.8). Later in 2014, an informal online expert group on contaminants was established and delivered its first report on assessment criteria in March 2015 and it was discussed at the MEDPOL Focal Points Meeting in June 2015 (UNEP(DEPI)/MED WG.417/Inf. 15).
2. The online group made a preliminary proposal regarding the Mediterranean BACs for major chemical pollutants (in sediment and biota) and biomarkers and recommended as a first step the use of a number of BAC and EAC values both adopted by OSPAR and developed by scientific studies in the Mediterranean Sea. The group pointed out the need to undertake a data analysis of additional data sets from Reference Stations in order to adjust (or to develop) as appropriate threshold assessment criteria for the Mediterranean Sea region.
3. In February 2016, Decision IG. 22/7 at the 19th Meeting of Contracting Parties (COP19) agreed on the Integrated Monitoring and Assessment Programme (IMAP) of the Mediterranean Sea and Coast and Related Assessment Criteria. The main outputs during the initial phase of IMAP will include further update of GES definitions, refinement of assessment criteria and development of national level integrated monitoring and assessment programmes.
4. Therefore, this document with refined assessment criteria will contribute as an instrument to both assess and monitor the achievement of GES (2017-2021). This assessment criteria includes Background Concentrations (BCs), Background Assessment Criteria (BACs) and Environmental Assessment Criteria (EACs), for hazardous chemical substances and biomarkers for the Mediterranean Sea as a whole. For the first time, background concentrations (BCs) have been also calculated at regional and sub-regional scales. These values will be updated and refined in the future with new available data.
5. The methodology and results contained in this document are fully elaborated in the information document (UNEP(DEPI)/MED WG.427/Inf.3), which contains all the detailed data/metadata information, datasets characteristics, statistical results and scientific rationale of the performed analysis and assessment.
6. This proposal was presented at the Meeting of the Ecosystem Approach Correspondence Group on Pollution Monitoring held in Marseille, France, 19-21 October 2016. During the meeting it was noted that true natural background concentrations were difficult to assess and even reference stations might be contaminated to a certain extent. Also it was recommended to follow closely the recent and evolving work within the MSFD so as to be aligned if appropriate. Other suggestions included the need for more data on biomarkers, to consider how baselines may vary significantly even within one sub-region, and how to attain the most accurate baseline value for sediments. Among the revisions include the addition of AChE activity (nmol/min mg protein in gills) for France as well as Spain.
7. In addition and for future elaboration of assessment criteria the general proposed changes and considerations include:
  - a. Nine new values for Cd, Hg and Pb in mussel, fish and sediment are proposed as new Mediterranean Background Assessment Criteria (Med BACs), as well as the adjustment of the fish EACs for trace metals. Further, the Annex to Decision 22/7 (IMAP) should be considered for EACs already imported from EU Directives (EU/1881/2006 and EU/629/2008) and OSPAR, despite taking into account the refinements proposed in this document (see Annex I). It is worth to

mention, that for Cd and Pb in fish fillet tissue, datasets exhibits analytical issues, such as they are up to 100% below the detection limits (<BDLs). Contracting Parties may consider, whether to continue reporting on these metals in fish flesh tissue, or instead report on these metals in liver tissue.

b. The Med BACs for organic compounds have been proposed solely for PAHs in biota, thus no more datasets were available. Twelve Med BACs are proposed as a new assessment criteria (see Annex I). For PAHs in sediment and organochlorinated compounds (OCs) no data was available (either sufficiently available or quality assured) to perform data analysis to derive Med BACs. Nevertheless, a revision has been provided for OCs (see Annex I). It is suggested that the Annex to Decision 22/7 (IMAP) for EACs (imported from adopted OSPAR values) should be considered for reference, as revised and reported in the present document (see Annex I). Further, it is recommended that Contracting Parties consider regular sediment sampling and the determination of organic contaminants, as limited studies are available for the Mediterranean Sea basin and sub-regions to establish proper assessment criteria.

c. New three calculated Med BACs and one revised EAC are proposed for three biomarkers (see Annex I). Nevertheless, the assessment criteria (BACs and EACs) for biomarkers are based on limited geographical data (i.e. mainly from Croatia, Spain and Italy) and multiple methodologies and reference values have been used (in particular for Lysosomal Membrane Stability, LMS). It is therefore suggested to ensure more comparable and precise results and standard methodologies to be adopted for all Mediterranean laboratories. Further, the Annex to Decision 22/7 (IMAP) for biomarkers assessment criteria in the Mediterranean should consider the observations mentioned above and the proposed amendments in this document.

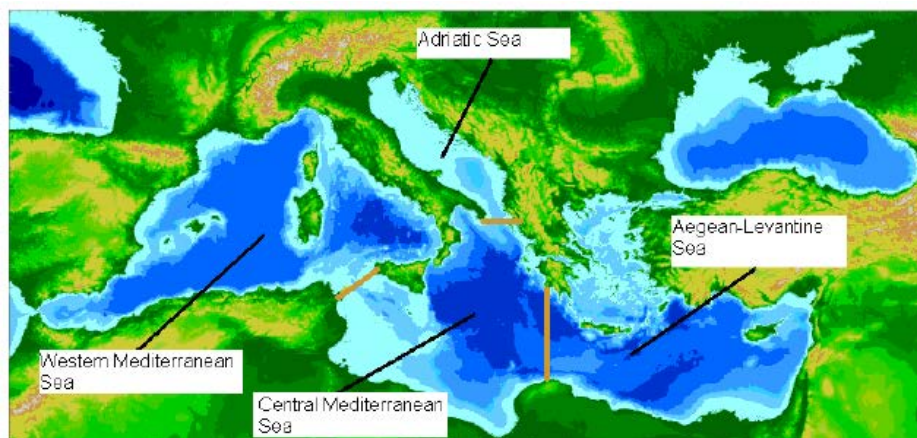
8. The present document was reviewed by the Meeting of the MED POL Focal Points, held in Rome, Italy, 29-31 May 2017, the 6<sup>th</sup> Meeting of the Ecosystem Approach Coordination Group, held in Athens, 11 September 2017 and the Meeting of the MAP Focal Points, held in Athens, 12-15 September 2017.

9. It reflects the comments expressed at the meetings, including the ones referring to the inclusion of a list of acronyms for PAHs and POPs, correction of units in French text and explanation that threshold values will be updated in the future due to new data.

## 1. Database Sources

1. This document uses the datasets for the MEDPOL Reference Stations updated until 2012, as well as the datasets submitted by Contracting Parties through the informal online group on contaminants during the period 2014-2015. The later, constitutes a selection of reference stations undertaken by national experts that has made available more recent data (including 2014) from national monitoring networks. This data was provided in the majority of cases for trace metals, petroleum hydrocarbons and biological markers (in the MEDPOL database format) for different biota species (fish and bivalves) and marine sediments all over the Mediterranean coastal environments. However, the online group data sets were not representative of Reference Stations/Areas for all the countries datasets submitted, and historical datasets, as well as from coastal and polluted areas were also submitted which would influence the results if computed straight. Therefore, a data selection process was found to be mandatory. Both databases needed a selection, aggregation and quality assurance processes (based on model-based clustering, see Section 3.6 in UNEP(DEPI)/MED WG.427/Inf.3) in order to select the reference stations before the statistical data analysis and assessments could be performed.

2. We have undertaken a synoptic approach to develop the calculations of the Mediterranean BCs (Med BCs) and BACs (MedBACs) at different spatial scales with the available merged datasets from both sources. Therefore, we assigned and grouped the selected stations by eco-regions as shown in the Figure 1.1 and further detailed in Table 1.1. It has been recognised that differences between regions and sub-regions within the Mediterranean Sea basins are likely to occur and should be taken into account for the environmental and pollution assessments as will be shown later in this document. The calculation of the Med BACs have been performed for the Mediterranean Sea as a whole with the calculated Med BCs (either corresponding to the median (50<sup>th</sup> percentile) for hazardous chemical substances or the 10<sup>th</sup> or 90<sup>th</sup> percentile for selected biomarkers), including BCs developed both for Mediterranean eco-regions and sub-regional seas. For, EACs, those adopted within OSPAR and the EU Directives (EU/1881/2006 and EU/629/2008) are further suggested.



**Figure 1.1.** The four Mediterranean MEDPOL eco-regions (WMS, Western Mediterranean Sea; ADR, Adriatic Sea; CEN, Central Mediterranean and AEL, Aegean and Levantine Seas)

**Table 1.1.** The Mediterranean eco-regions and sub-regions aggregation according the database sources and availability within this document.

Eco-regions	Sub-regional seas/basins*
Western Mediterranean Sea (WMS)	Alboran Sea (ALBS) North Western Mediterranean Sea (NWMS) Tyrrhenian Sea (TYRS) Western Mediterranean Islands and Archipelago (WMIA)
Adriatic Sea (ADR)	North Adriatic (NADR) Middle Adriatic (MADR) South Adriatic (SADR)
Central Mediterranean (CEN)	Central Mediterranean(CEN) Ionian Sea (IONS)
Aegean and Levantine Seas (AEL)	Aegean Sea (AEGS) Levantine (LEVS)

\*all available MED POL data was used (see UNEP(DEPI)/MED WG.427/Inf.3)

## 2. Scientific assessment of Background Concentrations (BCs) and Background Assessment Criteria (BACs)

### 2.1. Assessment criteria for trace metals (Cd, Hg, Pb) in mussels

3. For *Mytilusgalloprovincialis* species (MG), the tables below (Table 2.1 and Table 2.2) show the Med BCs and Med BACs calculated for the Mediterranean Sea basin, and the BCs for each eco-region except for the Central Mediterranean basin (no data available). The table also compares the determined Med BACs with the median value (50% of the data) of the MEDPOL Database for each eco-region earlier assessed (UNEP(DEPI)/MED WG.365/Inf.4), which included coastal and hotspot stations.

**Table 2.1.** Mediterranean BCs and BACs (Med BACs) for *Mytilusgalloprovincialis* ( $\mu\text{g}/\text{kg}$  d.w.)

Trace metal	Mediterranean Sea Basin		Western Mediterranean (WMS)		Adriatic Sea (ADR)		Central Mediterranean (CEN)		Aegean-Levantine Seas (AEL)	
	MedB Cs	Med BACs	WMS BCs	*50% MEDPOL Database	ADR BCs	*50% MEDPOL Database	CEN BCs	*50% MEDPOL Database	AEL BCs	*50% MEDPOL Database
Cd	730.0	1095.0	660.5	660 <MedBAC	782.0	800 <MedBAC	-	430 <MedBAC	942.0	750 <MedBAC
HgT	115.5	173.2	109.4	130 MedBAC	126.0	140 <MedBAC	-	160 <MedBAC	110.0	80 <MedBAC
Pb	1542	2313	1585	2000 <MedBAC	1381	1530 <MedBAC	-	810 <MedBAC	2300	2280 <MedBAC

\*median value of the MEDPOL Database from UNEP(DEPI)/MED WG.365/Inf.4 Report (2011); BAC=BCx1.5

4. It should be noticed that some calculated BCs for eco-regions (with solely reference stations datasets) are above the Med BCs, although below Med BACs. For example, cadmium and lead in the AEL eco-region (Aegean-Levantine Sea) in the Eastern Mediterranean Sea. Here, the explanation is that the major contributor to this eco-region (Turkey) presents quite higher background values for MG than the average for the reference stations in the Mediterranean Sea (see Information Document).



**Table 2.2.** Summary of statistical results of the Background Concentrations for trace metals (TM) in mussel by eco-regions in the Mediterranean Sea ( $\mu\text{g}/\text{kg dw}$ )

Trace metal	Eco-region	N <sup>1</sup>	Mean <sup>2</sup>	10 <sup>th</sup> 5	25 <sup>th</sup> 5	Median <sup>3</sup> (BCs)	IQR <sup>4</sup>	75 <sup>th</sup> 5	90 <sup>th</sup> 5
	ADR	151	1565.7	725.0	952.1	1381.0	1037.0	1989.1	2688.1
<b>Pb</b>	AEL	27	2174.8	944.4	1430.0	2300.0	1500.0	2930.0	3730.0
	WMS	157	1541.0	500.0	1075.4	1585.2	824.6	1900.0	2374.0
	ADR	154	196.2	85.6	104.6	126.0	71.4	176.0	409.0
<b>HgT</b>	AEL	31	123.4	74.2	80.0	110.0	79.0	159.0	177.8
	WMS	174	117.3	70.0	90.8	109.4	43.3	134.0	170.0
	ADR	151	753.8	413.0	510.5	782.0	470.5	981.0	1099.9
<b>Cd</b>	AEL	32	1269.1	297.1	737.7	942.0	1199.0	1936.7	3132.8
	WMS	174	731.5	364.0	520.0	660.5	370.0	890.0	1218.1

<sup>1</sup> N=number of individual data, <sup>2</sup> Mean=arithmetic mean as central tendency estimator <sup>3</sup>Median is the midpoint value (50<sup>th</sup> percentile) of the datasets, <sup>4</sup>IQR= interquartile range as a measure of the data dispersion (non-parametric distributions), <sup>5</sup>Percentile(s) = indicative value below a given % of the ordered datasets can be found.

## 2.2. Assessment criteria for trace metals (Cd, Hg, Pb) in sediment

5. The tables below (Table 2.3 and 2.4) shows the calculated Med BCs and BACs for the Mediterranean Sea and the BCs for each eco-region, except for the Central Mediterranean basin due to the shorten datasets in this eco-region. The calculated background concentrations (BCs) for each eco-region (aggregated datasets) exhibit values under the calculated Med BACs. The aggregated datasets considered solely the size fractions from <63 $\mu\text{m}$  to 2mm (see UNEP(DEPI)/MED WG.427/Inf.3).

**Table 2.3.** Mediterranean BCs and BACs (Med BCs and BACs) in surface sediments ( $\mu\text{g}/\text{Kg d.w.}$ )

Trace metal	<i>Mediterranean Sea Basin</i>		Western Mediterranean (WMS)		Adriatic Sea (ADR)		Central Mediterranean (CEN)		Aegean-Levantine Seas (AEL)	
	<i>MedB Cs</i>	<i>Med BACs</i>	<i>WMS BCs</i>	*50% MEDPOL Database	<i>ADR BCs</i>	*50% MEDPOL Database	<i>CEN BCs</i>	*50% MEDPOL Database	<i>AEL BCs</i>	*50% MEDPOL Database
Cd	85.0	127.5	91.2	1600 >MedBAC	92.3	210 >MedBAC	-	90 <MedBAC	56.0	100 <MedBAC
HgT	53.0	79.5	60.0	160 >MedBAC	106.8 >MedBAC	100 >MedBAC	-	50 <MedBAC	31.2	150 >MedBAC
Pb	16950	25425	20465	19400 <MedBAC	13932	9830 <MedBAC	-	4390 <MedBAC	4920	16890 <MedBAC >AELBAC**

\*median value of the MEDPOL Database from UNEP(DEPI)/MED WG.365/inf.4 Report (2011); BAC=BCx1.5;\*\*see text

6. Despite unbalanced datasets for each eco-region in the MEDPOL Database, this should be interpreted as preliminary information for the surface sediments contamination originated mostly from highly impacted coastal sites and known hotspots, particularly, in the Western Mediterranean Sea. Similarly as for mussels, it should be noticed that some calculated BCs for eco-regions with reference stations datasets are above the calculated Med BCs, although below Med BACs (except for HgT). Further, to correctly assess Pb in sediments in the AEL eco-region an AEL BAC should be considered.

**Table 2.4.** Summary of statistics for TM BCs in the Mediterranean eco-regions ( $\mu\text{g}/\text{kg}$  dw sediment).

Trace metal	Eco-region	N <sup>1</sup>	Mean <sup>2</sup>	10 <sup>th</sup> 5	25 <sup>th</sup> 5	Median <sup>3</sup> (BCs)	IQR <sup>4</sup>	75 <sup>th</sup> 5	90 <sup>th</sup> 5
<b>Pb</b>	ADR	37	16543	3429	7513	13932	19711	27223	32098
	AEL	85	13897	2123	3727	4920	13223	16950	41200
	CEN	2	2761	-	-	2761	-	-	-
	WMS	132	18792	6020	15935	20465	9135	25070	28845
<b>HgT</b>	ADR	27	119.6	28.9	37.8	106.8	149.9	187.7	240.8
	AEL	84	43.4	1.8	5.4	31.2	59.6	65.0	118.5
	CEN	1	58.0	-	-	58.0	-	-	-
<b>Cd</b>	WMS	122	70.3	24.8	40.0	60.0	40.0	80.0	138.4
	ADR	32	125.8	60.0	70.6	92.3	75.4	146.1	268.1
	AEL	20	114.5	35.4	45.7	56.0	42.8	88.5	387.1
	CEN	2	25.5	-	-	25.5	-	-	-
	WMS	85	94.4	50.0	71.5	91.2	32.5	104.0	128.4

<sup>1</sup> N=number of individual data, <sup>2</sup> Mean= arithmetic mean as central tendency estimator <sup>3</sup> Median is the midpoint value (50<sup>th</sup> percentile) of the datasets, <sup>4</sup> IQR= interquartile range as a measure of the data dispersion (non-parametric distributions), <sup>5</sup>Percentile(s) = indicative value below a given % of the ordered datasets can be found.

### 2.3. Assessment criteria for trace metals (Cd, Hg, Pb) in fish

7. Trace metals in fish are determined in several species in the Mediterranean Sea, such as *Mullus barbatus* (MB), *Boopsboops* (BB), *Mullussurmuletus* (MS) and *Upneusmollucensis* (UM). These species have been selected in the framework of MEDPOL according their geographical distribution within the national monitoring programs, with some countries monitoring more than one species. The majority of monitoring datasets are available for *Mullus barbatus* (MB), and therefore, this species has been chosen as the reference species to calculate the Mediterranean BCs and BACs for trace metals, although the statistical analysis has been undertaken for all (see UNEP(DEPI)/MED WG.427/Inf.3). The countries with available datasets for reference stations for MB were Cyprus, Greece, Italy, Spain, Turkey and Israel.

8. In terms of quality assurance, the datasets for Cd and Pb in fillet tissue of MB presents some analytical issues which impede to determine consistent Med BCs and BACs. Particularly, for Cd, the majority of the countries datasets reported over a 90% of the data as BDLs (below detections limit), if not a 100%. Similarly, for Pb, a majority of datasets are reported as either as BDLs or with large values pointing to sample contamination or reporting issues. This is valid for reference stations, but also for coastal and hotspot stations within the MEDPOL Database, and therefore, it should be concluded that the MB species is not a good proxy for the evaluation of Cd and Pb in Mediterranean fish (fillet tissue). More, the organic contaminant determinations in MB are almost all 100% reported as BDLs (eg. OCs). Obviously, this impedes to correlate any biological effects with the concentrations of hazardous chemical contaminants in MB sampled from the environment. The table below (Table 2.5) presents the calculated Med BCs and BACs for fish (fresh weight), despite the considerations explained above need to be observed.

**Table 2.5.** Mediterranean BCs and BACs (Med BACs) in fish ( $\mu\text{g}/\text{kg}$  f.w.)

Trace metal	<i>Mediterranean Sea Basin</i>		Western Mediterranean (WMS)	Adriatic Sea (ADR)	Aegean-Levantine Seas (AEL)
	<i>MedBCs</i>	<i>Med BACs</i>	<i>WMS BCs</i>	<i>ADR BCs</i>	<i>AEL BCs</i>
Cd	(3.7) <sup>a</sup>	(16.0) <sup>b</sup>	-	-	-

Hg	50.6	101.2	68.0	150.5 >MedBAC	44.6
Pb	(31) <sup>a</sup>	(40) <sup>b</sup>	38	-	20

<sup>a</sup>Cd value is below the detection limit (<BDL) and Pb presents a majority of non-detected values in monitoring datasets.

<sup>b</sup>estimated BACs from reliable limits of detection (BAC=1.5 x LOD) using both analytical data and certified reference material information (DORM-2). However, liver tissue matrix should be recommended for Cd and Pb as within OSPAR Convention.

;BAC=BCx2.0

#### 2.4. Assessment criteria for polycyclic aromatic hydrocarbons (PAHs) in mussels

9. In order to develop Mediterranean BC and BACs for PAHs in mussels a limited number of datasets from MEDPOL countries for reference stations were available (Table 2.6). The contributions were from France, Greece, Italy, Spain and Turkey. The polycyclic aromatic hydrocarbons were determined in mussel samples of similar length; despite each country followed different strategies to pool the samples (see UNEP(DEPI)/MED WG.427/Inf.3).

**Table 2.6.** Mediterranean BCs and BACs (Med BACs) in mussel samples (µg/kg d.w.)

PAH*	Mediterranean Sea Basin		Western Mediterranean (WMS)	Adriatic Sea (ADR)	Aegean-Levantine Seas (AEL)
	MedBCs	Med BACs	WMS BCs	ADR BCs	AEL BCs
F	1.0	2.5	0.96	1.07	0.60
P	7.1	17.8	4.93	9.04	7.55
A	0.5	1.2	0.52	0.38	0.30
FL	3.0	7.4	3.38	2.03	6.60
PY	2.0	5.0	3.02	0.85	5.90 >MedBAC
BaA	0.8	1.9	1.20	0.53	1.60
C	1.0	2.4	1.24	0.27	5.20 >MedBAC
BkF	0.6	1.4	1.27	0.29	1.50 >MedBAC
BaP	0.5	1.2	0.60	0.32	0.70
GHI	0.9	2.3	0.90	-	1.20
DA	0.5	1.3	0.53	-	-
ID	1.2	2.9	1.23	-	0.90

\*Not included: Naphthalene, Acenaphthylene, Acenaphthene are below detection limits (BDLs) or had a limited monitoring datasets, and therefore BACs are preliminary estimations. Benz(e)pyrene and Benzo(b)fluoranthene had not enough datasets.

10. In terms of quality assurance checks, the primary normal component ( $\lambda$ ) for PAHs ranged between values from 58% to 96% for the aggregated reference stations datasets indicating from sufficient to excellent aggregated datasets. In the Table 2.7 below, the medians (BCs) for individual

PAHs are shown for each Mediterranean eco-region. It should be noticed, that some eco-region medians (BCs) are above the calculated Med BACs. In this case, as mentioned before, this responds to the effect of grouped data by geographical areas with a scarce number of high data in reference stations. Further, when the medians (BCs) and confidence intervals are above the Med BACs for a sub-regional sea, the number and magnitude of the data should be further examined in detail and related to low  $\lambda$  values (see UNEP(DEPI)/MED WG.427/Inf.3).

**Table 2.7.** Summary of statistics for PAHs BCs in the Mediterranean eco-regions ( $\mu\text{g}/\text{kg}$  dw mussel).

PAHs	Eco-region	N <sup>1</sup>	Mean <sup>2</sup>	10 <sup>th</sup> 5	25 <sup>th</sup> 5	Median <sup>3</sup> (BCs)	IQR <sup>4</sup>	75 <sup>th</sup> 5	90 <sup>th</sup> 5
F	ADR	60	1.13	0.66	0.77	1.07	0.59	1.36	1.73
	AEL	3	0.93	0.50	0.50	0.60	-	-	-
	WMS	76	1.50	0.49	0.60	0.96	1.19	1.78	3.23
P	ADR	60	9.25	5.94	7.87	9.04	2.73	10.60	13.92
	AEL	2	7.55	5.60	5.60	7.55	-	-	-
	WMS	90	7.17	2.35	3.70	4.93	3.76	7.46	11.59
A	ADR	55	0.77	0.21	0.25	0.38	0.33	0.58	2.09
	AEL	3	0.43	0.30	0.30	0.30	-	-	-
	WMS	53	0.88	0.29	0.38	0.52	0.33	0.71	1.36
FL	ADR	60	2.66	1.23	1.41	2.03	2.32	3.73	4.65
	AEL	3	5.50	1.90	1.90	6.60	-	-	-
	WMS	90	5.42	1.71	2.03	3.38	3.91	5.94	12.51
PY	ADR	60	2.82	0.38	0.51	0.85	1.22	1.73	4.43
	AEL	3	6.53	3.40	3.40	5.90	-	-	-
	WMS	90	5.17	0.97	1.77	3.02	4.75	6.52	13.20
BaA	ADR	60	1.02	0.19	0.28	0.53	0.94	1.22	3.17
	AEL	3	1.70	1.40	1.40	1.60	-	-	-
	WMS	40	2.82	0.29	0.57	1.20	3.70	4.27	7.34
C	ADR	50	0.74	0.12	0.19	0.27	0.93	1.11	2.27
	AEL	3	4.63	2.70	2.70	5.20	-	-	-
	WMS	68	4.20	0.55	0.77	1.24	4.12	4.90	12.83
BeP	ADR	-	-	-	-	-	-	-	-
	AEL	3	2.63	1.90	1.90	2.80	-	-	-
	WMS	42	1.12	0.36	0.51	0.79	0.79	1.30	2.41
BbF	ADR	30	0.59	0.13	0.35	0.43	0.30	0.65	1.46
	AEL	3	3.93	1.20	1.20	5.30	-	-	-
	WMS	59	2.43	0.18	0.26	0.49	3.44	3.70	7.40
BkF	ADR	24	0.28	0.09	0.14	0.29	0.20	0.33	0.53
	AEL	3	1.10	0.30	0.30	1.50	-	-	-
	WMS	46	2.07	0.32	0.50	1.27	2.23	2.73	5.26
BaP	ADR	27	0.38	0.10	0.11	0.32	0.32	0.43	1.09
	AEL	3	0.60	0.30	0.30	0.70	-	-	-
	WMS	45	1.17	0.21	0.38	0.60	0.72	1.10	3.00
GHI	AEL	3	1.13	0.70	0.70	1.20	-	-	-
	WMS	50	1.37	0.31	0.40	0.90	1.23	1.63	2.73
DA	WMS	24	0.76	0.32	0.38	0.53	0.21	0.60	0.88
ID	AEL	3	0.80	0.40	0.40	0.90	-	-	-
	WMS	25	1.74	0.50	0.61	1.23	1.59	2.20	4.47

<sup>1</sup> N=number of individual data, <sup>2</sup> Mean= arithmetic mean as central tendency estimator <sup>3</sup> Median is the midpoint value (50<sup>th</sup> percentile) of the datasets, <sup>4</sup> IQR= interquartile range as a measure of the data dispersion (non-parametric distributions), <sup>5</sup>Percentile(s) = indicative value below a given % of the ordered datasets can be found.

## 2.5. Assessment criteria for biomarkers (AChE, MT, MN, LMS and SOS) in mussels

11. The developments of the assessment criteria using the information from the MEDPOL pilot biomonitoring programmes were too limited with datasets from Croatia, Italy and Spain. Some datasets from Greece were also available from 2005. Therefore, the Western Mediterranean Sea and the Adriatic Sea eco-regions were evaluated for the majority of biomarkers, whilst the Aegean-Levantine Sea eco-region was only evaluated for one biomarker (LMS-LP) but not finally used. The following table (Table 2.8) shows the calculated Mediterranean BCs and BACs for selected biomarkers (see UNEP(DEPI)/MED WG.427/Inf.3 for full details and plots) using the WMS region data, thus data from the Adriatic Sea should be further investigated and could be of use solely at local level. Therefore, it should be noted that the proposed assessment criteria (see Annex I in this document) is applicable to a sub-regional scale according the countries datasets origin (see further details in UNEP(DEPI)/MED WG.427/Inf.3 ) and previous information.

**Table 2.8.** Mediterranean BCs and BACs calculated for biomarkers in mussel samples.

Biomarker	<i>Mediterranean Sea Basin</i>		Western Mediterranean (WMS)	Adriatic Sea (ADR)
	<i>MedBCs (median)</i>	<i><sup>a</sup>Med BACs</i>	<i>WMS BCs</i>	<i>ADR BCs</i>
AChE activity (nmol/min mg protein in gills) <sup>b</sup>	21	15	20.86	12.20 <MedBAC
Metallothioneins (µg/g digestive gland (DG))	192	247	191.3	200.5
Lysosomal membrane stability (LMS-Neutral red retention (NRR), minutes)	(45)	120*	45.0 <Standard	47.4 <Standard
Lysosomal membrane stability (LMS-Liabilisation period (LP), minutes)	(13)	20*	-	16.8 <Standard
Micronuclei frequency (per 1000 in haemocytes)	0.0	1.0	0.0	0.5
Stress on stress (days)	7	11	-	-

<sup>a</sup>either the 10<sup>th</sup> percentile or the 90<sup>th</sup> percentile are considered for biomarkers to establish the Background Assessment Criteria (BACs), see UNEP(DEPI)/MED WG.427/Inf.3;<sup>b</sup>from available datasets from Spain; \*adopted ICES/OSPAR standard

**Annex I**  
**Tables of the proposed assessment criteria**

**A. Table of the proposed assessment criteria for trace metals (TMs)**

The tables below (Tables A.1.1 and A.1.2) compare the new proposed/revised BCs, BACs and EACs in this document (using Reference Stations datasets) with the earlier proposed threshold values in the Mediterranean Sea. Further details can be found in the information document (UNEP(DEPI)/MED WG.427/Inf.3).

**Table A.1.1.** Mediterranean Sea: Background Concentrations (Med BCs), Med BACs and EACs; Calculation =>BC = 50th (median); BAC=1.5 x BC (mussel, sediment); BAC=2.0 x BC (fish)

Trace metal	Mussel (MG) µg/kg d.w.			Fish (MB) µg/kg f.w.			Sediment µg/kg d.w.		
	BC	Med BAC	EC*	BC	Med BAC	EC*	BC	Med BAC	ERL**
Cd	730.0	1095.0	5000	(3.7) <sup>a</sup>	(16.0) <sup>b</sup>	50	85.0	127.5	1200
Hg	115.5	173.2	2500	50.6	101.2	1000	53.0	79.5	150
Pb	1542	2313	7500	(31) <sup>a</sup>	(40) <sup>b</sup>	300	16950	25425	46700

<sup>a</sup>Cd value is below the detection limit (<BDL) and Pb presents a majority of non-detected values in monitoring datasets.

<sup>b</sup>estimated BACs from reliable limits of detection (BAC=1.5 x LOD) using analytical data and certified reference material information (DORM-2) (see also text). However, liver tissue matrix should be recommended in fish for Cd and Pb as within OSPAR Convention.

\*EC/EU 1881/2006 and 629/2008 Directives for maximum levels for certain contaminants in foodstuffs

\*\* Long et al. 1995 (idem OSPAR adopted values)

**Table A.1.2.** Earlier data (2011-2015) from UNEP(DEPI)/MED WG.365/Inf.8, UNEP(DEPI)/MED WG.417/inf.15 Part3 and Annex to UNEP(DEPI)/MED IG.22/7Decision.

Trace metal	<sup>a</sup> Mussel (MG) µg/kg d.w.			<sup>b</sup> Mussel µg/kg d.w.	<sup>c</sup> Fish (MB) µg/kg d.w. <sup>f</sup>			Sediment µg/kg d.w.		
	BC	Med BAC	EC	BAC	BC	Med BAC	(EC)	BC	<sup>e</sup> Med BAC	ERL
Cd	725	1088	5000	1000	4	8/16 <sup>d</sup>	207	-	150	1200
Hg	125	188	2500	170	296	600	4150	-	45	150
Pb	2500	3800	7500	1000	279	558	1245	-	30000	46700

<sup>a</sup> preliminary data for the NW Mediterranean (Spain);

<sup>b</sup> additional BAC data provided by Lebanon for *Brachidontes variabilis* species;

<sup>c</sup> preliminary data for the NW Mediterranean (Spain);

<sup>d</sup> earlier estimation wet weight;

<sup>e</sup> estimated from sediment cores (UNEP(DEPI)/MED WG.365/Inf.8, 2011);

<sup>f</sup> a dry/wet ratio of 20 should be used to convert units for MG (f.w. units = d.w. units / 5)

## B. Table of the proposed assessment criteria for polycyclic aromatic hydrocarbons (PAHs)

The tables below (Tables A.2.1 and A.2.2) compare the new proposed/revised BCs, BACs and EACs with the earlier proposed threshold values in the Mediterranean Sea. Further details can be found in the information document (UNEP(DEPI)/MED WG.427/Inf.3).

**Table A.2.1.** Mediterranean Sea Background Concentrations (BCs), Med BACs and EACs; Calculation =>BC = 50th (median); BAC=2.5 x BC (mussel); no data for sediment available

PAH compound	Mussel (MG) µg/kg d.w.			Sediment µg/kg d.w.		
	Med BC	Med BAC	<sup>a</sup> OSPAR EAC	<sup>a</sup> OSPAR BC	<sup>a</sup> OSPAR BAC	<sup>c</sup> ERL
F	1.0	2.5	-	-	-	-
P	7.1	17.8	1700	4.0	7.3	240
A	0.5	1.2	290	1.0	1.8	85
FL	3.0	7.4	110	7.5	14.4	600
PY	2.0	5.0	100	6.0	11.3	665
BaA	0.8	1.9	80	3.5	7.1	261
C	1.0	2.4	-	4.0	8.0	384
BkF	0.6	1.4	260	-	-	-
BaP	0.5	1.2	600	4.0	8.2	430
GHI	0.9	2.3	110	3.5	6.9	85
DA	0.5	1.3	-	-	-	-
ID	1.2	2.9	-	4.0	8.3	240

<sup>\*</sup>Naphthalene, Acenaphthylene, Acenaphthene, Benz(e)pyrene and Benzo(b)fluoranthene are below detection limits (BDLs) or have limited monitoring datasets, and therefore their BACs are preliminary estimations.

<sup>a</sup>OSPAR Commission, CEMP: 2008/2009 Assessment of trends and concentrations of selected hazardous substances in sediments and biota (OSPAR PAHs sediment datasets from Spain, not TOC corrected; <sup>c</sup>ERL: Effect Range Low

**Table A.2.2.** Earlier data (2011-2015) from UNEP(DEPI)/MED WG.365/Inf.8, UNEP(DEPI)/MED WG.417/inf.15 Part3 and Annex to UNEP(DEPI)/MED IG.22/7Decision.

PAH compound	Mussel (MG) µg/kg d.w.			Sediment µg/kg d.w.		
	Med BC	Med BAC	<sup>a</sup> OSPAR EAC	<sup>a</sup> OSPAR BC	<sup>a</sup> OSPAR BAC	<sup>a</sup> ERL
P		24.3	1700		7.3	240
A		4.1	290		1.8	85
FL		6.8	110		14.4	600
PY		6.1	100		11.3	665
BaA		1.3	80		7.1	261
C		2.4	-		8.0	384
BkF		1.8	260		-	-
BaP		1.3	600		8.2	430
GHI		1.3	110		6.9	85
ID		0.8	-		8.3	240



**C. Table of the proposed assessment criteria for organochlorinated compounds (OCs)**

(Summary of OSPAR values to be used in the Mediterranean Sea)

**Table A.3.1.** OSPAR Region (Background Concentrations (BCs), BACs and EACs)<sup>1</sup>

OCs compound	Mussel µg/kg d.w.			Fish µg/kg w.w.			Sediment µg/kg d.w.		
	BC/LC <sup>c</sup>	BA C	EAC	BC/LC <sup>c</sup>	BA C	EAC (lipid w.)	BC/LC <sup>c</sup>	BAC	EAC/ER L
CB28 <sup>a</sup>	0.25	0.75	3.2	0.05	0.10	64	0.05	0.22	1.7
CB52 <sup>a</sup>	0.25	0.75	5.4	0.05	0.08	108	0.05	0.12	2.7
CB101 <sup>a</sup>	0.25	0.70	6.0	0.05	0.08	120	0.05	0.14	3.0
CB105 <sup>a</sup>	0.25	0.75	-	0.05	0.08	-	0.05	-	-
CB118 <sup>a</sup>	0.25	0.60	1.2	0.05	0.10	24	0.05	0.17	0.6
CB138 <sup>a</sup>	0.25	0.60	15.8	0.05	0.09	316	0.05	0.15	7.9
CB153 <sup>a</sup>	0.25	0.60	80	0.05	0.10	1600	0.05	0.19	40
CB156 <sup>a</sup>	0.25	0.60	-	0.05	0.08	-	0.05	-	-
CB180 <sup>a</sup>	0.25	0.60	24	0.05	0.11	480	0.05	0.10	12
Σ7CBs ICES <sup>b</sup>	-	-	-	-	-	-	0.20	0.46	11.5*
Lindane <sup>a</sup>	0.25	0.97	1.45	-	-	11**	0.05	0.13 <sub>+</sub>	3.0*
α-HCH <sup>a</sup>	0.25	0.64	-	-	-	-	-	-	-
pp'DDE <sup>a</sup>	0.25	0.63	5-50***	0.05	0.10	-	0.05	0.09 <sub>+</sub>	2.2*
HCB <sup>a</sup>	0.25	0.63	-	0.05	0.09	-	0.05	0.16 <sub>+</sub>	20.0*
Dieldrin <sup>a</sup>	-	-	5-50***	-	-	-	0.05	0.19 <sub>+</sub>	2.0*

<sup>1</sup>OSPAR Commission, 2013.

<sup>a</sup>OSPAR Commission, CEMP: 2008/2009 Assessment of trends and concentrations of selected hazardous substances in sediments and biota, Monitoring and Assessment Series

<sup>b</sup>OSPAR Commission, Background document on CEMP assessment criteria for the QSR 2010, Monitoring and Assessment Series

<sup>c</sup>LC: Low concentrations calculated from QUASIMEME; However, BC values should be considered as zero for OCs

<sup>d</sup>Total organic carbon (TOC) corrected values; <sup>+</sup>LC from Spain (OSPAR, 2013)

\*ERLs values instead EACs: Effect Range Low (Long et al. 1995); ERL for ICES Σ7CB is total CB concentration/2

\*\*EAC for fish liver derived by applying a conversion factor of 10 on EAC for whole fish (CEMP 2008/2009)

\*\*\*Ecotoxicological assessment criteria (earlier data from the QSR2000 Report-Chapter 4)

It should be noted that at present, no quality assured or sufficient datasets exist in the MEDPOL Database to calculate the threshold values for the Mediterranean Sea.

#### D. Table of the proposed assessment criteria for biological markers in mussels

The tables below (Tables A.4.1 and A.4.2) compare the new proposed/revised BCs, BACs and EACs with the earlier proposed threshold values in the Mediterranean Sea. Further details can be found in the information document (UNEP(DEPI)/MED WG.427/Inf.3).

**Table A.4.1.** Mediterranean Sea and standard reference values; Calculation => BAC = 10<sup>th</sup> or 90<sup>th</sup> percentile depending on the parameter.

Biomarkers	Mussel ( <i>Mytilusgalloprovincialis</i> )	
	Med BAC	EAC
Stress on Stress (SOS, days)	11	5 <sup>a</sup>
Metallothioneins (µg/g digestive gland)	247	-
Lysosomal membrane stability (LMS-NNR, neutral red retention method, minutes)	120 <sup>a*</sup>	50 <sup>a*</sup>
Lysosomal membrane stability (LMS-LP, Cytochemical method, labilisation period minutes)	20 <sup>a*</sup>	10 <sup>a*</sup>
AChE activity (nmol/min mg protein in gills) <sup>b</sup> -France	29	20
AChE activity (nmol/min mg protein in gills) <sup>b</sup> -Spain	15	10 <sup>a</sup>
Micronuclei frequency (per 1000 in haemocytes)	1.0	-

<sup>a</sup>Technical annex: assessment criteria for biological effects measurements. Integrated monitoring of chemicals and their effects. ICES Cooperative Research Report No. 315. Davies, I.M. and Vethaak, A.D.Eds.

<sup>b</sup>subregional differences between assessment criteria are observed by countries

\*Moore et al., 2006 (Standard values adopted by ICES)

**Table A.4.2.** Earlier data (2015) from UNEP(DEPI)/MED WG.417/inf.15 Part3 and Annex to UNEP(DEPI)/MED IG.22/7Decision.

Biomarkers	Mussel ( <i>Mytilusgalloprovincialis</i> )	
	Med BAC	EAC <sup>a</sup>
Stress on Stress (SoS, days)	10	5
Lysosomal membrane stability (LMS-NNR, neutral red retention method, minutes)	120	50
Lysosomal membrane stability (LMS-LP, Cytochemical method, labilisation period minutes)	20	10
AChE activity (nmol/min mg protein in gills) - France	29	20
AChE activity (nmol/min mg protein in gills) - Spain	15	10
Micronuclei frequency (per 1000 in haemocytes)	3.9	-

**Annex II**  
**References**

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