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Integrated indicator framework and methodology for monitoring and assessment of hazardous substances and their effects in the marine environment

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Abstract:

Many maritime countries in Europe have implemented marine environmental monitoring programmes which include the measurement of chemical contaminants and related biological effects. How best to integrate data obtained in these two types of monitoring into meaningful assessments has been the subject of recent efforts by the International Council for Exploration of the Sea (ICES) Expert Groups. Work within these groups has concentrated on defining a core set of chemical and biological endpoints that can be used across maritime areas, defining confounding factors, supporting parameters and protocols for measurement. The framework comprised markers for concentrations of, exposure to and effects from, contaminants. Most importantly, assessment criteria for biological effect measurements have been set and the framework suggests how these measurements can be used in an integrated manner alongside contaminant measurements in biota, sediments and potentially water. Output from this process resulted in OSPAR Commission (www.ospar.org) guidelines that were adopted in 2012 on a trial basis for a period of 3 years. The developed assessment framework can furthermore provide a suitable approach for the assessment of Good Environmental Status (GES) for Descriptor 8 of the European Union (EU) Marine Strategy Framework Directive (MSFD).

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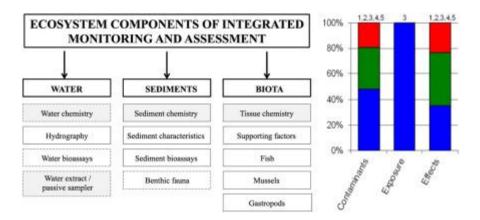
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Graphical abstract



Highlights

▶ An integrated framework for marine contaminants and their effects is developed. ▶ Determinants for sediment, fish, and shellfish with assessment criteria are provided. ▶ A multistep traffic light data aggregation tool is proposed and demonstrated. ▶ It resulted in OSPAR guidelines for integrated chemical-biological effect monitoring. ▶ The approach could be useful for determination of GES for Descriptor 8 of MSFD.

Keywords: chemical measurements, biomarker, bioassay, pollution effects, biomonitoring, environmental, impact, MSFD, ICES, OSPAR

1. Introduction

Our seas and oceans are dynamic and variable. They represent a fundamental component of global ecosystems and, as such, we need to be able to assess the health status of the marine environment. Furthermore, we need to be able to detect anthropogenic induced changes in seas and oceans and to identify the reasons for these changes. It is only through such understanding that we can advise on necessary and appropriate remedial responses, such as regulatory action, as well as report on any improvements resulting from management measures. There is a need to express clearly what is meant by the "health" of the marine environment, and for that purpose, we require

indicators of the health of ecosystem components, including indicator measurements for assessing the impacts of anthropogenic contaminants.

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The marine environment receives inputs of hazardous substances through riverine discharges, direct (end of pipe) inputs, and atmospheric deposition and is the ultimate repository for complex mixtures of persistent chemicals. Consequently, organisms are exposed to a range of substances, many of which can cause metabolic disorders, an increase in disease prevalence, and, potentially, effects on populations through changes in growth, reproduction, or survival (e.g. Matthiessen and Gibbs, 1998; Hylland et al., 2006a; Moore et al., 2006). Through much of the history of marine pollution research and monitoring, chemical and biological field studies have often remained largely independent of each other. There are many publications describing the distribution of hazardous substances in the marine environment and, equally, many describing the perturbations of species or communities as a consequence of exposure to hazardous substances (e.g. Muir et al., 1999; Vos et al., 2000; Hylland et al., 2006b). However, it is now generally agreed that the assessment of environmental quality, and the design and monitoring of measures to improve environmental quality, are best undertaken on the basis of combinations of appropriate sets of chemical and biological measurements (Hylland, 2006; Thain et al., 2009; Lyon et al., 2010; Piva et al., 2011; Roose et al., 2011; Benedetti et al., 2012; Lehtonen et al., 2014). In the past, monitoring to assess the potential negative impact of hazardous substances has been based primarily on measurements of substance concentration. This was because the questions being asked concerned concentrations of such substances in water, sediment, and biota, and such measurements were possible for a specific set of relevant substances. However, in order to more fully assess the health of our maritime area, questions about the bioavailability of hazardous substances and their impact on marine organisms or processes are now being posed. Biological effect techniques have become increasingly important in the past few decades. Sometimes a biological response can be observed when the causative substance is below current chemical analytical detection limits; the development of imposex in gastropod molluscs as a result of low concentrations of tributyltin (TBT) being a point in case (e.g.

Matthiessen and Gibbs, 1998; Antizar-Ladislao, 2008). However, biological responses may also occur as a result of low concentrations of several substances causing an additive or synergistic joint effect (e.g. McDowell et al., 1999; Silva et al., 2002; Pojana et al., 2006) or in the absence of identified causative compound (s) (e.g. Lyons et al., 2006).

Many strategies and approaches have been proposed to assess (marine) ecosystem health using ecological indicators (e.g. Rapport et al., 1998; EEA, 2001; Jorgensen et al., 2005; OSPAR 2010a). Among them, there are different tools for biological effect (biomarkers and bioassays) data integration and interpretation with the aim to develop integrated effect-based indices for the quantification of effects of hazardous substances at several levels of biological organization (e.g. Moore et al., 2004; Broeg and Lehtonen, 2005; Dagnino et al., 2007; Viarengo et al., 2007; Piva et al., 2011; Marigómez et al., 2013). Consequently, biological-effect methods are important elements in environmental monitoring programmes, because they can indicate links between contaminants and ecological responses. Biological effect monitoring can thus be used to indicate the presence of substances, or combinations of substances, that had not been identified previously as being of concern, but also to identify regions of decreased environmental quality or reduced ecosystem health.

The pressure to clarify an integrated approach to assessing the impact of contaminants through both biological effects and chemical monitoring increased as a result of the requirement to achieve Good Environmental Status under Descriptor 8 (Concentrations of contaminants are at levels not giving rise to pollution effects) of the European Union Marine Strategy Framework Directive (MSFD, Directive 2008/56/EC). The Regional Sea Conventions (RSCs) in Europe have largely agreed on an ecosystem approach to manage the marine environment, under which the Conventions have committed themselves to monitor marine ecosystems in order to understand and assess interactions between, and impact of, human activities on marine organisms. Integrated monitoring and assessment of contaminants in the marine environment and their effects will

contribute effectively to the integrated assessment of the full range of human impacts on the quality status of the marine environment, as part of the ecosystem approach.

This paper describes the integrated indicator framework and methodology for hazardous substances and their effects developed by the International Council for Exploration of the Sea (ICES) and OSPAR Commission. In addition, this paper serves as a background to the practical application of the framework for the ICON (Integrated assessment of contaminant impacts on the North Sea) project and other baseline studies in North East Atlantic waters (e.g. Giltrap et al., 2014; Lyon et al., this volume) and the West Mediterranean Sea (Martinez-Gomez et al., this volume). The guidelines are supported by associated background documents (OSPAR, 2013a), which provide information on the scientific background and assessment criteria to the contaminants and biological effects measurements included in the programme.

2. Current European strategies

The European Union (EU) has, over the last twenty years, developed its water policies so that now there is significant European legislation covering marine waters and the lakes and rivers that ultimately flow into our coastal ecosystems. The EU Water Framework Directive (WFD) (Directive 2000/06/EC) establishes a framework for community action in the field of water policy, central to which is a good ecological status for defined water bodies. This is described on the basis of biological quality, hydromorphological quality, and physico-chemical quality. More recently, the European Union has implemented the Marine Strategy Framework Directive (MSFD) (Directive 2008/56/EC). At its heart is the concept of "Good Environmental Status" (GES) for all European waters and the provision of a framework for the protection and preservation of the marine environment, the prevention of its deterioration, and, where practicable, the restoration of that environment in areas where it has been adversely affected. GES will be assessed on a regional basis. The Regional Sea Conventions (OSPAR Commission, Helsinki Commission (HELCOM), Barcelona

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Convention and the Black Sea Commission) which aim to protect the marine environment are required to support the implementation of the MSFD since the Directive requires that, in developing their marine strategies, Member States use existing regional cooperation structures to co-ordinate among themselves and to make every effort to coordinate their actions with those of third countries in the same region or sub-region. The programmes of the various Regional Sea Conventions, including OSPAR, provide a valuable source of data for the assessments that have been completed so far and will be required in the future. The MSFD specifies that GES will be assessed against 11 qualitative descriptors. The Commission Decision (2010 / 477 / EU) further described three criteria to be used in assessing GES for Descriptor 8 (Concentrations of contaminants are at levels not giving rise to pollution effects): contaminant concentrations (8.1), biological effects of chronic exposure (8.2.1) and the impact of acute pollution events (8.2.2); therefore, D8 has been interpreted as requiring assessments of contaminant concentrations and their biological effects. A task group established by the European Commission Joint Research Centre (JRC) and ICES interpreted this as meaning that the concentrations of contaminants should not exceed established quality standards (e.g. EQS, environmental assessment criteria (EAC)) and that the intensity of biological effects attributable to contaminants should not indicate harm at organism level or higher levels of organization (Law et al., 2010).

3. Integrated monitoring of contaminants and their effects

The contribution made by an integrated programme involving both chemical and biological effects measurements is primarily that the combination of the different measurements increases the interpretive value of the individual measurements and thus delivers an improved assessment of

¹ In 1975, 16 Mediterranean countries and the European Community adopted the Mediterranean Action Plan (MAP), the first-ever Regional Seas Programme under UNEP's umbrella.

status. For example, biological effects measurements assist the assessment of the significance of measured concentrations of contaminants in biota or sediments, and can include an assessment of the impact of concurrent exposure to multiple contaminants. When biological effects measurements are carried out in combination with chemical measurements (or additional effects measurements), allowing, in some cases, the identification of the substance/group of substances contributing to the observed effects. By bringing together these monitoring (and assessment) disciplines that have tended to be conducted separately, an integrated assessment can improve our ability to describe the reasons for areas with decreased or poor environmental status detected during monitoring programmes. The economic benefit of an integrated approach comes from the fact that the samples and data are gathered during the same surveys and that the data can be directly compared/used with holistic assessment tools to provide truly integrated (with respect to contaminant concentrations and their effects) assessments.

Fundamental aspects of the design of an integrated monitoring programme include key environmental matrices (water, sediment and biota), the selection of appropriate combinations of biological effects and chemicals to be measured, and the design of sampling programmes to allow the chemical concentrations, the biological effects data, and other supporting parameters to be combined to provide a more robust assessment of the impact of contaminants on the marine environment.

Chemical analyses in the different environmental matrices to be included in an integrated programme should cover the priority hazardous substances or chemicals listed by European legislation and Regional Sea Conventions. Analytical methods (including the sampling frequency and spatial distribution) should be sufficiently sensitive to detect variation in environmental quality and should be supported by appropriate quality management. Biological effects methods to be included in an integrated programme to assess the impact of contaminants on the marine environment require the following characteristics (ICES, 2007; adapted):

- the ability to separate contaminant-related effects from influences caused by other
 factors (e.g. natural variability, food availability);
 - sensitivity to a specific contaminant or group of contaminants (i.e. providing "early warning" of an impact through the identification of an effect);
 - a broad enough suite of methods that ensures coverage of a range of mechanisms of toxic
 action (e.g. oestrogenicity / androgenicity, neurotoxicity, carcinogenicity, genotoxicity,
 and mutagenicity); and
 - the inclusion of at least one method that measures the general health status of a test organism (whole-organism response).

Some matrices/determinands are considered fundamental to the integrated assessment of contaminants and are described as "core methods". Where additional matrices/ determinands have been found to add value to the integrated assessment, these have been described as "additional methods" and are not considered essential. The basic structure of an integrated monitoring and assessment programme is illustrated in Figure 1.

Biological effects measurements and chemical methods have been selected for the biota matrix (separated as fish, mussels and gastropods) using these criteria. In addition, some physiological characteristics of the specific fish and mussel populations are required. For example, in fish gonadosomatic index (GSI), liver somatic index (LSI), and condition factor, as described in supporting technical annexes (see OSPAR, 2013b). Similarly, spawning status in all species is relevant to the biological effect assessment. General designs for integrated monitoring of fish are presented in Figure 2 and of mussels in Figure 3. Designs for water, sediment, and gastropod monitoring are included as Figures 4, 5, and 6, respectively.

The integration of contaminant and biological effects monitoring, and thereafter assessment, requires a strategy for simultaneous sampling and subsequent analysis. Examples of sampling strategies for the integrated fish and shellfish schemes are shown in Figures S1 and S2. In order to integrate sediment, water chemistry, and associated bioassay components with the fish and bivalve schemes, sediment and water samples should be collected at the same time as fish / bivalve samples and from a site or sites that are representative of the defined station/sampling area. Additional integrated sampling opportunities may arise from trawl/grab contents, for example, gastropods for imposex or benthos, and these should be exploited where possible/practicable.

4. Integrated assessment of contaminants and their effects

4.1. The need for assessment criteria

It is not sufficient simply to coordinate sampling; integration must also involve a combined assessment of the monitored parameters, which must themselves be selected with the assessment aim in mind. Such a combined assessment may involve using environmental and biological parameters as covariates in statistical analyses or they may be used to standardize effect variables (e.g. temperature, seasonal, gender or size/age effects on biomarker responses). Similarly, normalization procedures for the expression of contaminant concentrations in biota and sediment have been established. For example, defined bases (e.g. dry weight or lipid weight) are used for biota analyses, and sediment data is, on occasions, normalized to organic carbon or aluminium to minimize the influence of differences in bulk sediment properties.

Ultimately, the purpose of an integrated monitoring programme is to provide the necessary data to facilitate integrated assessments to enable the status of the marine environment in relation to hazardous substances to be described as a contribution to general assessments of the quality status of the maritime area (e.g. OSPAR Quality Status Reports (QSRs; e.g. OSPAR (2010a), HELCOM Initial Holistic Assessment (HELCOM, 2010)); ICES Integrated Ecosystem Assessments).

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An important and essential step to integrate information from chemical contaminants, biomarkers and biological data is the establishment of assessment criteria for all parameters measured. For chemical contaminant concentrations, OSPAR has developed two types of assessment criteria to be assessed and presented in directly comparable "traffic light" formats (Figure 7) (OSPAR 2010a): those reflecting levels above Background Concentrations (BCs) referred to as Background Assessment Concentrations (BACs), and Environmental Assessment Criteria (EACs) representing concentrations below which unacceptable biological effects were unlikely to occur.

In the same way, OSPAR, with assistance from ICES, has more recently developed coherent sets of analogous assessment criteria for biological effects measurements, most of them specifically derived from field data of North Atlantic species in European waters (Table 1). However, unlike contaminant concentrations in environmental matrices, assessment criteria for certain biological responses have been developed taking into consideration factors such as species, gender, maturation status, season and temperature of ambient water. For other marine regions or species, outside the OSPAR maritime area, a regional-validated approach should be used to derive specific assessment criteria for environmental matrices and biological responses, such as those developed for the western Mediterranean (Martínez-Gómez et al, this issue). The concept of a background level of response (residual noise of the measurement found from responses of animals in relatively clean waters) is applicable to all effects measurements. Assessment criteria analogous to the EAC (i.e. representing levels of response below which unacceptable responses at higher, e.g. organism or population, levels of biological organisation would not be expected) are applicable for some biological effects measurements, and these have been termed "biomarkers of effect". In other cases, the link to higher level effects is less clear, and these measurements have been termed "biomarkers of exposure", in that they indicate that exposure to hazardous substances has occurred. Importantly, the processes used to derive both the BAC and their biological analogues and the EAC and their analogues have been applied consistently to all chemical and effects measurements. This

coherence across the broad range of assessment criteria forms the basis for integrated assessment schemes.

Furthermore, the coherence of assessment criteria across both chemical and biological effects measurements allows these two types of data to be brought together into a single integrated assessment scheme. The "traffic light" presentation is equally applicable to biological effects data and can be used to present data integrated over a range of geographical scales from the single sampling site to the sub-regional scale, as required under the MSFD. The application of this approach is described below.

4.2. Multi-step assessment framework

A multistep traffic light data aggregation tool to assess contaminants and biological effects data together is proposed which follows on from experience of the assessment of contaminants data for sediment, fish, and shellfish in an OSPAR context. The development of BAC and EAC equivalent assessment criteria for biological effects, which represent the same degree of environmental risk as indicated by BAC and EAC values for contaminants, allows the representation of these monitoring data alongside contaminant data using the same approach to the graphical representation

The process is informed initially by the individual assessment of determinands (contaminant concentrations or effect levels) in specific matrices at individual sites against the defined assessment criteria (BAC and EAC). Initial comparisons determine whether the determinand and site combinations are < BAC (blue), between the BAC and EAC (green), or > EAC (red). This summarized indicator of status for each determinand can then be integrated over a number of levels: matrix (sediment, water, fish, mussel, gastropod), site, and region and expressed with varying levels of aggregation to graphically represent the proportion of different types of determinands (or for each determinand, sites within a region) exceeding either level of assessment criteria.

Such an approach has several advantages. The integration of data can be simply performed on multiple levels depending on the type of assessment required and the monitoring data available. The

representation of the assessment maintains all of the supporting information, and it is easy to identify the causative determinands that may be responsible for exceeding EAC. In addition, any stage of the assessment can be readily unpacked to a previous stage to identify either contaminant or effects measurements of potential concern or sites with a poor outcome in terms of environmental status The inclusion of biological effects data to the system adds considerable value to the interpretation of assessments. Where sufficient effects monitoring data are available, confidence can be gained that contaminants are not (or are) having significant effects even where contaminant monitoring data are lacking. In instances where contaminant concentrations in water/sediment are > EAC, a lack of EAC threshold breach in appropriate effects data can provide some confidence that contaminant concentrations are not giving rise to pollution effects (due, for example, to lack of availability to marine biota). Similarly, the inclusion of effects data in the assessment framework can indicate instances where contaminants are having significant effects on biota, but have not been detected or covered in a contaminant-specific chemical monitoring programme.

The multistep assessment framework described in detail below provides an appropriate tool for assessment of environmental monitoring data to determine whether or not "Good Environmental Status" is being achieved for Descriptor 8 of the MSFD. Determinands with EAC or EAC equivalent assessment criteria provide appropriate indicators with quantitative targets. The assessment of contaminant and effects monitoring data against these EAC level assessment criteria provides information both on concentrations of contaminants likely to give rise to effects and the presence/absence of significant effects in marine biota.

Owing to the relatively large number of determinands monitored under the integrated approach, it is inappropriate to adopt an approach whereby EAC level failure of a single determinand results in failure of GES for a site or region ("one-out all-out" approach). A more appropriate approach would involve the setting of a threshold (%) of proportion of determinands that should be < EAC to achieve GES. Such an approach would avoid the failure of sites or regions as a result of

occasional outlying or erroneous results for particular determinands. The setting of an appropriate threshold for overall regional assessment for MSFD will require consideration and revision in the light of testing the framework described here with real monitoring data. However, an initial threshold of 95% < EAC (to ensure that the vast majority, but not all, of contaminants/effects measurements should be < EAC) is proposed here for the purposes of testing the system.

In order to best demonstrate how monitoring data (assessed against BAC and EAC) can be integrated for matrices, sites, and regions, and ultimately provide an assessment that could be useful for determination of GES for Descriptor 8, a worked example following a five-step process is provided in Table 1 and Figure 8.

5. Applicability of integrated indicator framework for OSPAR maritime areas

Among the Regional Sea Convention programmes, OSPAR has a well-established monitoring framework with agreed monitoring programmes and associated chemical and biological assessment criteria to focus on those determinands which will complement relevant activities made in other frameworks (EU WFD (Directive 2000/60/EC; EU MSFD (Directive 2008/56/EC) (OSPAR, 2010). The OSPAR Hazardous Substances Strategy (OSPAR Agreement 2003–2021; OSPAR, 2010, 2014) declares that the Commission will implement this Strategy progressively by making every endeavour to move towards the target of the cessation of discharges, emissions, and losses of hazardous substances by the year 2020. In association with this, OSPAR has developed the JAMP/CEMP (Joint Assessment and Monitoring Programme/Co-ordinated Environmental Monitoring Programme)) (OSPAR, 1998, 2008a,b, 2014. This provides the basis for the monitoring activities undertaken by contracting parties to assess progress towards achieving OSPAR objectives. In relation to hazardous substances, the JAMP/CEMP seeks to address the following questions:

What are the concentrations of hazardous substances in the marine environment? Are the
monitored hazardous substances at, or approaching, background levels for naturally occurring
substances and close to zero for synthetic substances? How are the concentrations changing

over time? Are the concentrations of either individual substances or mixtures of substances such that they are not giving rise to pollution effects.

How can OSPAR's monitoring framework be improved and extended and better linked with the understanding of biological effects and ecological impacts of individual substances and the cumulative impacts of mixtures of substances?

Therefore there is a need to adopt an integrated approach to the monitoring of contaminants in the marine environment and the biological responses to the presence of hazardous substances. In order to assess progress towards the objectives of the OSPAR Hazardous Substances Strategy, OSPAR has already developed assessment criteria for contaminant concentration and biological effects data (see Table S1). The work described above has resulted in the development of OSPAR JAMP guidelines for integrated chemical and biological monitoring of contaminants, at length described in this paper, and was adopted by OSPAR in 2012 to run on a 3-year trial basis (2012-2015). The ICON project, presented elsewhere in this volume, represents the first large scale integrated assessment of the status of a marine region for contaminants and their effects.

6. Conclusions and perspectives

This paper provides the scientific basis for a framework for integrated chemical and biological effects monitoring and assessment in the marine environment. The framework comprises a core set of biological effect techniques developed by ICES and included or recommended in the OSPAR monitoring programmes that can be used in an integrated manner together with chemical contaminant measurements in biota, sediments and water across OSPAR maritime areas (OSPAR Agreement 2012-09). It further comprises an assessment framework that integrates contaminant and biological effects monitoring data and that allows assessments to be made across matrices, sites, and regions. The assessment framework is simple and transparent and allows for multiple levels of aggregation for different assessment requirements. The presented integrated framework and methodology can generally be applied to other marine regions including the Baltic Sea and

Mediterranean Sea. However this may require some development and application of region-specific
methods in key species and associated assessment criteria.

- The key components of the integrated monitoring and assessment framework are:
- a) Defined combinations of chemical and biological effects measurements;
- 378 b) Carefully managed sampling programmes;
- 379 c) Measurement methods and determinands that are understood and well supported by
 380 background documents, technical annexes, standard protocols, quality control, etc.;
- d) A coherent set of assessment criteria that represent similar levels of environmental risk acrossdeterminands;
 - e) Data integration methods that enable combination of data over a range of geographical scales and which can accommodate limited or incomplete data sets.

The ICES/OSPAR framework links chemical contaminants with the health of the ecosystem and can provide a suitable approach for the assessment of GES for Descriptor 8 of the MSFD. In order to give some stability to assessments, it is important that future revisions of techniques and assessment criteria are harmonized with the MSFD cycle. Currently, the background documents and assessment criteria are available for all biological effect techniques relevant to the ecosystem components for integrated monitoring of contaminants and their effects, apart from benthic fauna and passive samplers. These are important elements of the integrated scheme, and work to prepare background documents and assessment criteria needs to be undertaken as soon as possible. However, it should be noted that our knowledge regarding integrated monitoring and assessment will continue to evolve and new emerging contaminants and new techniques should be added or replace old ones.

397	The ICES/OSPAR framework has been validated through the international ICON project and a set
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Legends to Figures and Table

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532	Figure 1. Overview of components in a framework for an integrated monitoring programme for
533	chemical contaminants and their biological effects developed by ICES and OSPAR. Solid lines, core
534	methods; broken lines, additional methods.
535	Figure 2. Determinands and measurements included in the fish component of the ICES/OSPAR
536	integrated monitoring framework. Solid lines, core methods; broken lines, additional methods. PCBs,
537	polychlorinated biphenyls; BFRs, brominated flame retardants; AChE, acetylcholinesterase. WFD,
538	Water Framework Directive. WFD priority substances are required in biota under Directive
539	39/2013/EU. Supportive factors for biota are not shown (details can be found in OSPAR et al.,
540	2013b).
541	Figure 3. Determinands and measurements included in the mussel component of the ICES/OSPAR
542	integrated monitoring framework. Solid lines, core methods; broken lines, additional methods. PCBs,
543	polychlorinated biphenyls; PAH, polycyclic aromatic hydrocarbon; BFRs, brominated flame
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547	Figure 4. Determinands and measurements included in the water component of the ICES/OSPAR
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550	ordinated Environmental Monitoring Programme of OSPAR. #CEMP and pre-CEMP determinants are
551	listed in OSPAR agreement 2010-01, as amended in 2014. WFD Priority Substances are listed in
552	Directive 2013/39/EU and have to be assessed for WFD in coastal and transitional waters.
553	Figure 5. Determinands and measurements included in the sediment component of the ICES/OSPAR
554	integrated monitoring framework. Solid lines, core methods; broken lines, additional methods.

Figure 6. Determinands and measurements included in the gastropod component of the integrated monitoring framework. Solid lines, core methods; broken lines, additional methods.

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Figure 7. OSPAR regional-level integration of the concentrations of priority contaminants in fish, shellfish, and sediment based on results from the OSPAR Coordinated Environmental Monitoring Programme (CEMP). As can be seen from the figure, the concentrations of Region II (Greater North Sea) are still widely above background values for mercury, cadmium, lead and PAHs and above zero for PCBs and are unacceptable in many, mostly coastal, areas. Overall, contamination is lowest in Region I (Arctic) where many of the sites monitored meet the OSPAR objective of background values for heavy metals; however concentrations of PAHs and PCBs are still unacceptable at a third of the sites monitored. Overall, the situation is better for heavy metals, although more than 40% of sites monitored show unacceptable levels of lead in Region II (Greater North Sea) and of mercury in Region IV (Bay of Biscay and Iberian Coast). Red status: concentrations are at levels such that they there is an unacceptable risk of chronic effects occurring in marine species, or are greater than EU dietary limits for fish or shellfish but the extent of risks of pollution effects is uncertain. Green status: concentrations of contaminants are at levels where it can be assumed that little or no risks are posed to the environment and its living resources at the population or community level. Blue status: concentrations are near background for naturally occurring substances or close to zero for man-made substances (reprinted with permission from OSPAR (2010c).

- Concentrations are at levels such that they there is an unacceptable risk of chronic effects occurring in marine species, or are greater than EU dietary limits for fish or shellfish but the extent of risks of pollution effects is uncertain.
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579	man-	-made substances.

Figure 8.1-5. Integrated assessment framework: integration of three colour (blue, green and red) classifications of measurements of contaminant concentrations and their effects. A red classification indicates that the Environmental Assessment Criteria (EAC) is exceeded, blue indicates compliance with the Background Assessment Concentration (BAC), whereas green indicates concentrations or levels of effects are between the BAC and EAC. 8.1 (Step 1): Illustration of classification of measurements of contaminants and their effects by matrix for a specific site; 8.2 (Step2): Integration across determinands within matrices for a given site; 8.3 (Step3): Integration of matrices by determinand category for a given site; 8.4A (Step 4): Integration of determinands across sampling sites within an assessment region; 8.4B (Step 4): Integration of matrices across sampling sites by determinand category within an assessment region. 8.5 (Step 5): Integration of determinands across sampling sites, matrices, and determinands within an assessment region.

Table 1. A worked example following a five-step process to demonstrate how monitoring data (assessed against BAC and EAC) can be integrated for matrices, sites, and regions and ultimately provide an overall assessment that could be useful for determination of GES for Descriptor 8 of the EU MSFD. Determinands and their status are provided for illustrative purposes only to show how subsequent integration can be performed.

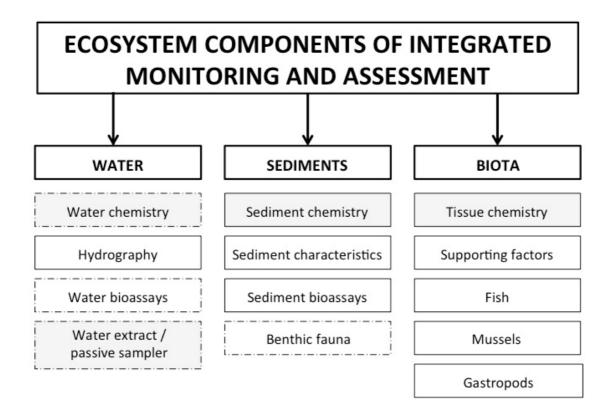


Figure 1. Overview of components in a framework for an integrated monitoring programme for chemical contaminants and their biological effects developed by ICES and OSPAR. Solid lines, core methods; broken lines, additional methods.

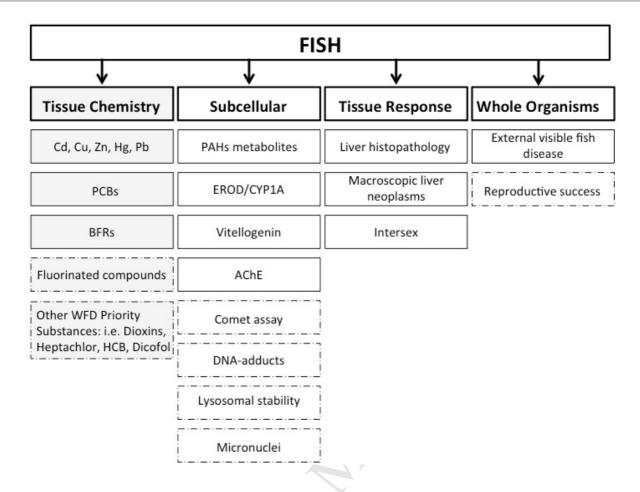


Figure 2. Determinands and measurements included in the fish component of the ICES/OSPAR integrated monitoring framework. Solid lines, core methods; broken lines, additional methods. PCBs, polychlorinated biphenyls; BFRs, brominated flame retardants; AChE, acetylcholinesterase. WFD, Water Framework Directive. WFD priority substances are required in biota under Directive 39/2013/EU. Supportive factors for biota are not shown (details can be found in OSPAR et al., 2013b).

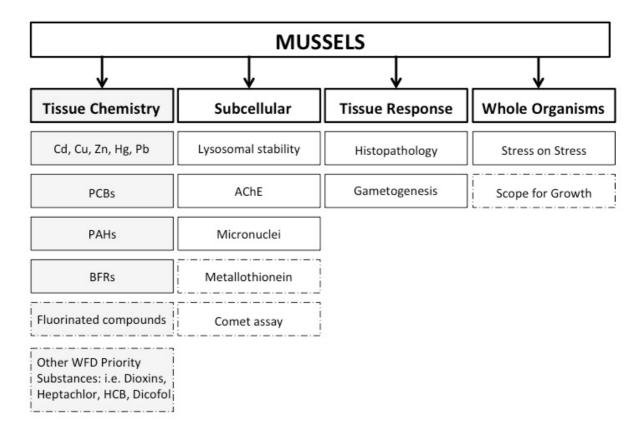


Figure 3. Determinands and measurements included in the mussel component of the ICES/OSPAR integrated monitoring framework. Solid lines, core methods; broken lines, additional methods. PCBs, polychlorinated biphenyls; PAH, polycyclic aromatic hydrocarbon; BFRs, brominated flame retardants; AChE, acetylcholinesterase. WFD, Water Framework Directive. WFD priority substances are required in biota under Directive 39/2013/EU. Supportive factors for biota are not shown (details can be found in OSPAR et al., 2013b).

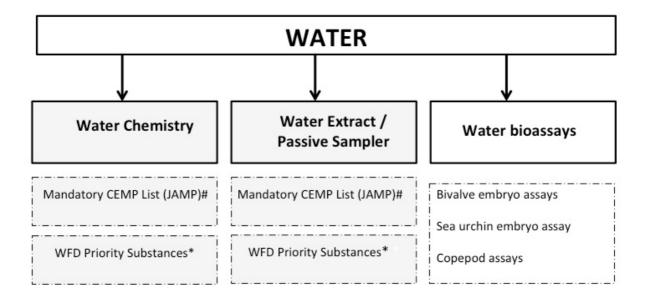


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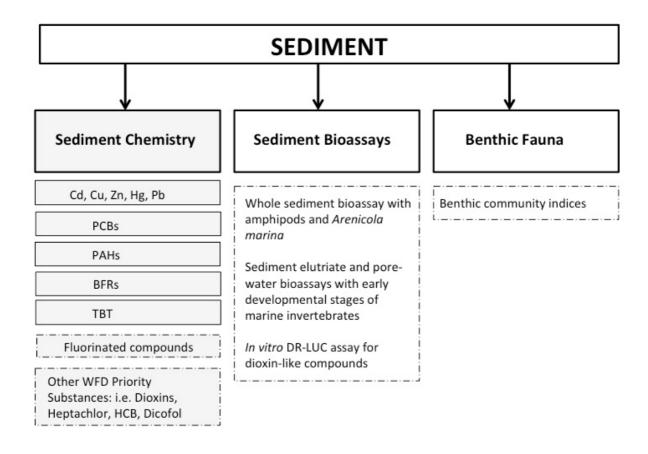


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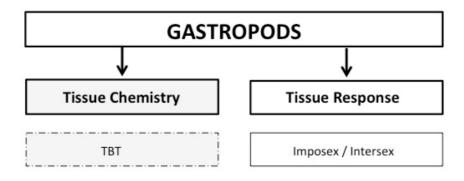


Figure 6. Determinands and measurements included in the gastropod component of the ICES/OSPAR integrated monitoring framework. Solid lines, core methods; broken lines, additional methods.

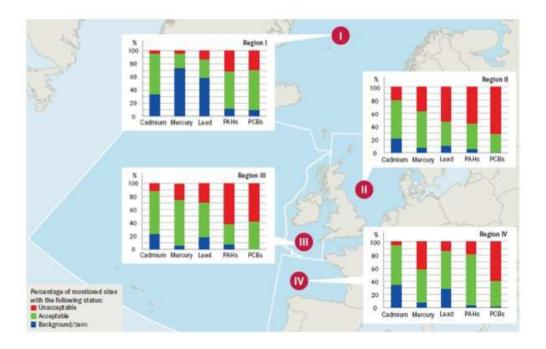


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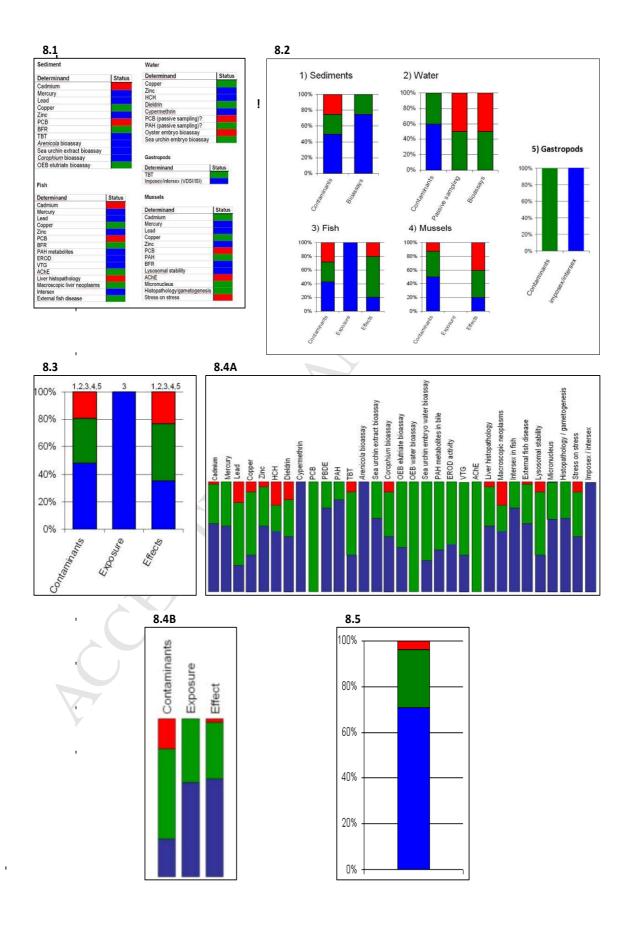


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Table 1. A worked example following a five-step process to demonstrate how monitoring data (assessed against BAC and EAC) can be integrated for matrices, sites, and regions and ultimately provide an overall assessment that could be useful for determination of GES for Descriptor 8.

Determinands and their status are provided for illustrative purposes only to show how subsequent integration can be performed.

Step	Level of integration	Description
1	Assessment of	All determinands available for a specific site assessment as shown
	monitoring data by	in Figure 8.1 are compiled with results presented by monitoring
	matrix against BAC	matrix and expressed as a colour depending on whether or not the
	and EAC	value exceeds BAC or EAC, following the "traffic light" system
		(OSPAR, 2008). Briefly, a red classification indicates that upper
		confident limit of the mean determinand values exceeded the EAC,
		blue indicates upper confident limit of the mean determinand
		values does not exceed the BAC, whereas green indicates upper
		and lower confident limits of concentrations or levels of effects are
		between the BAC and EAC.
2	Integration of	For each of the five matrices, the results of the individual
	determinands by	determinand assessments are aggregated into categories:
	matrix for a given	contaminants, exposure indicators, effects indicators, and, for
	site	sediment/water matrices, also passive sampling and bioassay
		categories. The integration by matrix and category of determinand
		can be expressed by three-coloured bars showing the proportions
		of determinands that exceed the BAC and EAC as shown in Figure
		8.2. It is necessary, however, to separate the biological effects
		measurements into different categories depending on whether or
		not an EAC-equivalent assessment criterion (AC) has been set.
		Otherwise, aggregated information on the proportion of
		determinands exceeding the separate AC will be incorrect. For
		simplicity, these categories have been termed "exposure
		indicators" (where an EAC has not been set) and "effects

indicators" where an EAC (equivalent to significant pollution effect) has been set for the measurement. On subsequent aggregation/integration of these indicators across matrices for a specific site, bioassays are considered "effects indicators" as EAC are available. It should be possible to include data from passive sampling in both the water and sediment schemes when assessment criteria have become available. They are nominally included in the example here to show how they could be included. Each method for contaminant, effect, or exposure assessment carries the same weight, within the matrix, in the integration shown in Figure 8.2. Note that for mussels in this instance, no exposure indicators are used, because all of the biological effects measurements have EAC available.

3 Integration of matrices for a site assessment

In order to simply express the results of assessment for a particular site, information can be aggregated across matrices and expressed by determinand category, as shown below (Figure 8.3). In order to achieve this, results from passive sampling from sediment and water categories could be integrated into the contaminant indicator graphic and bioassays and gastropod intersex/intersex integrated into "effects indicators". Thus, the outcome of assessment of all determinands from all matrices can be expressed for a whole site. In practice, the process adopted is to sum the percentages of each colour in, say, the "contaminants" columns for each matrix in Figure 8.2, and then to scale the sums to a total of 100%. The results for each matrix, therefore, carry equal weight in the integration shown in Figure 8.3.

For some assessments, this will be the highest level of aggregation required. However, for assessments covering larger geographical areas (subregional, regional, national, regional seas for the MSFD, etc.) where assessments need to be undertaken across multiple sites, a further level of integration is required (Steps 4 and 5). For transparency, each determinand grouping is labelled with the matrices from which it is comprised. Thus, it can quickly be determined whether the site assessment is composed of all or just

a subset of the monitoring matrices. In the example below (Figure 8.3), all five matrices have been used to determine the overall site assessment. However, only for fish (matrix 3) were there any effects measurements that did not have EAC available for assessment. Therefore, the exposure indicators graphic is labelled to show that only matrix 3 contributed to the site assessment of indicators of exposure.

 4 Regional assessment across multiple sites This can be done at multiple levels (aggregation of data at the subregional, regional, and national levels) in different ways to express both the overall assessment of proportion of determinands (across all matrices) exceeding both assessment thresholds (BAC/EAC; approach A) and by determinand for the region showing the proportion of sites assessed in the region that exceed the thresholds (approach B). Both approaches show the overall proportion of determinand/site incidences of threshold exceedance. However, approach A shows most clearly which determinands are responsible for any EAC exceedance, whereas approach B shows a more aggregated, summarized representation of the same information by determinand category. Both can be constructed directly from the output of Step 1.

4A Regional assessment of sites by determinand

This shows a graphical representation (Figure 8.4A) of the proportion of sites falling into each status class for each determinand across all relevant matrices (many determinands are only relevant to one or some of the matrices).

4B Regional assessment of sites by determinand category

The above regional assessment can be summarized by determinand category as was demonstrated in Step 3 for the site assessment and shown below (Figure 8.4B).

5 Overall assessment

The assessment by region can be aggregated further into a single schematic showing the proportion of all determinands across all sites that exceed BAC and EAC (Figure 8.5). This can be used for the purposes of an overall assessment, and it is proposed that a simple threshold figure (e.g. 95% <EAC) is used to determine whether or not "Good Environmental Status" for Descriptor 8 is met in this

assessment. The overall assessment can be easily unpacked through the steps above to determine which sites and determinands (effects types or contaminants) are contributing to, for example, the proportion of red (>EAC) data, and thereby potentially leading to failure to achieve GES for a region

Supporting Information

Integrated indicator framework and methodology for monitoring and assessment of hazardous substances and their effects in the marine environment

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- Figure S1. Sampling strategy for integrated fish monitoring.
- Figure S2. Sampling strategy for integrated bivalve monitoring.
- Table S1. Assessment criteria for biological effects measurements. Values are given for both background assessment criteria (BAC) and environmental assessment criteria (EAC), as available.

Integrated site 'fish scheme' Target species (e.g. dab, flounder, whiting, dragonet, or Sediment & water taken from sites representative suggested alternative). Sampling will consist of fish from one haul or more combined hauls at one station* Other integrated sampling opportunities: TBT-specific biological effects monitoring (e.g. gastropods) benthic biodiversity External disease assessment and macroscopic liver nodules. For guidance follow ICES published protocols (Bucke et Sediment chemistry (JAMP guidelines for monitoring al., 19961) and BEQUALM fish disease contaminants in sediment OSPAR 2002-16) programme² Chemical determinants (as appropriate) Contaminant analysis 25 fish in a Physical characteristics (e.g. sediment PSA 3cm length range (within size range Bioassays (e.g. CALUX, invertebrate embryo Sub sample 50 fish liver sampled for biomarkers) divided into Water Chemistry for histopathology 5 batches of 5. Follow JAMP Salinity, nutrients, oxygen Guidelines for contaminants in biota Chemical determinants (as appropriate) (OSPAR 1999-2) and Figure 2.2 to Liver histopathology assessment Passive samplers prioritise determinants Follow ICES TIMES protocols for liver histopathology³ FISH** Sub sample 20 fish (10 tissue chemistry subcellular tissue response whole organism males: 10 females) for biomarkers studies Externally visible Cd, Cu, Zn, Hg, Pb PAH metabolites Liver histopathology fish disease -----Macroscopic liver CBs EROD/CYP1A Reproductive success Biomarkers: Endpoints driven by site specific neoplasms factors and relevant integrated package(s) **BFRs** vitellogenin intersex Follow JAMP Guidelines for contaminant specific biological effects monitoring (OSPAR-2003-10) AChE compounds and Figure 2.2 to prioritise components Comet assay **DNA-adducts** Lysosomal stability

Figure or a larger defined area (from: Davis and Vethaak, 2012, see also OSPAR, 2013). (Solid lines **S1**. Overview methods, broken of methods ţ be included additional methods). 1 ⊒. integrated programme . Note: \triangleright station may be site specific for selected fish species.

Figure

Overview

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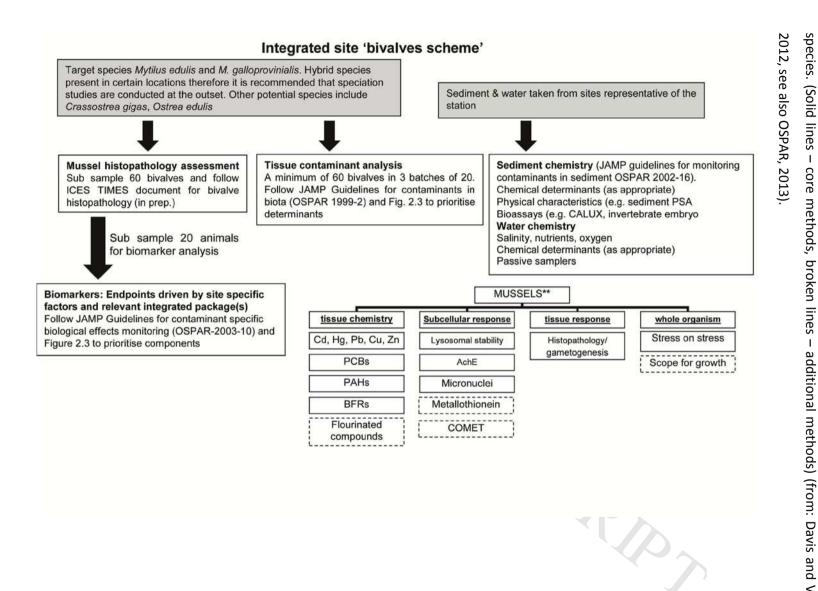


Table S1. Assessment criteria for biological effects measurements. Values are given for both background assessment criteria (BAC) and environmental assessment criteria (EAC), as available. (F) female, (M) male. Full details of the assessment criteria and how they were derived can be found in the OSPAR background documents for individual biological effects methods and reports from OSPAR (OSPAR, 2013b), ICES (Davis et al., 2012), updated in 2013 by ICES (ICESWGBEC report, 2013).

Biological effect	Applicable to:	BAC	EAC
Vtg in plasma; μg ml⁻¹	Cod	0.23	
	Flounder	0.13	
Reproduction in eelpout (Zoarces viviparus); mean frequency (%)	Malformed fry	1	2
	Late dead fry	2	4
	Early dead fry	2.5	5
	Total abnormal fry	5	10
EDOD: pmol ma ⁻¹ protein	Dab (F)	178	
EROD; pmol mg ⁻¹ protein pmol min ⁻¹ mg ⁻¹ protein S9	Dab (M)	147	
pmol min ⁻¹ mg ⁻¹ microsomal protein	Dab (M/F)	680	
First time and	Flounder (M)	24	
	Plaice (M)	9.5	
	Cod (M/F)	145*	
	Plaice (M/F)	255*	
	Four spotted megrim (M/F)	13*	
	Dragonet (M/F)	202*	
	Red mullet (M)- April	208	
	Red mullet (M/F)-October	115 ⁺	
	12-18 cm; GSI <1		
	Bottom temperature 16-20°C		
	Eelpout (F)	10	
	Dab	16 ¹ *	
PAHs bile metabolites;	Dab		
¹ ng ml⁻¹; HPLC-F		3.7 1 **	
² pyrene-type μg mΓ¹; synchronous scan fluorescence 341/383 nm		0.15 ²	22 ²
³ ng g ⁻¹ GC-MS	Cod	21 ¹ *	483 ³ *
*1-OH pyrene		2.7 1 **	528 ³ **
**1-OH phenanthrene		1.1 ²	35 ²
	Flounder	16 ¹ *	
		3.7 1 **	
		1.3 2	29 ²
	Haddock	13 1 *	
		0.8 1 **	- 2
BB 1 -1 1		1.9 ²	35 ²
DR-Luc; ng TEQ kg ⁻¹ dry wt, silica clean-up	Sediment (extracts)	10.0	40.0
DNA adducts; nm adducts mol DNA	Dab	1.0	4.0
	Flounder Long Rough Dab	1.0	4.0 4.0
	Halibut		4.0 5.8
	Herring and sprat		0.4
	Cod	1.6	6.7
	Haddock	3.0	6.7
Bioassays; % mortality	Sediment, Corophium	20	60
	Sediment, Arenicola	10	50
	Water, copepod	10	50

Biological effect	Applicable to:	BAC	EAC
Bioassays; % abnormality	Water, oyster embryo	20	50
	Water, mussel embryo	30	50
	Water, sea urchin embryo	10	50
Bioassay; % growth	Water, sea urchin embryo	30	50
Disassay, is grown	Tracol, ood dioriii oliibiyo	00	00
Lysosomal stability; min	Cytochemical; all species	20	10
	Neutral red retention: all species	120	50
Micronuclei; 0/00 (frequency of micronucleated cells)	Mytilus edulis	2.5 ¹	
¹ Gill cells		2.5 ²	
² Haemocytes	Mytilus galloprovincialis	3.9 ²	
³ Erythrocytes	Mytilus trossulus	4.5 ²	
	Flounder	0.0-0.3 3	
	Dab	0.5 ³	
	Eelpout	0.3-0.4 ³	
	Cod	0.4 3	
	Red mullet (M/F)-October	0.3 3	
	, ,	0.3	
	12-18 cm; GSI<1		
	Bottom temp 16-20°C		
Comet assay; % DNA tail	Mytilus edulis	10	
	Dab	5	
	Cod	5	
Stress on stress; days	Mytilus sp.	10	5
AChE activity; nmol min ⁻¹ mg ⁻¹ protein	Mytilus edulis	30 ¹ *	21 1 *
¹ Gills		26 ¹ **	19 ¹ **
² Muscle tissue	Mytilus galloprovincialis	29 ^{1 +}	20 1 +
³ Brain tissue	, , ,	15 ^{1 +}	10 1 +
*French Atlantic waters	Flounder	235 ² *	165 ² *
**Portuguese Atlantic waters	Dab	150 ² *	105 ² *
† French Mediterranean Waters	Red mullet	155 ^{2 +}	109 2 +
** Spanish Mediterranean Waters	Red mullet (M/F)-October	118 ^{3 ++}	83 ^{3 ++}
*** Baltic sea	` '	110	03
Dailic Sea	12-18 cm; GSI<1		
	Bottom temp 16-20°C		
	Eelpout (F)	124 2 +++	87 ^{2 +++}
Externally visible disease; Fish Disease Index (FDI)			
F: Females; M:Males; NA: Not applied;			
Ep,Ly,UI	Dab	F: 1.32,	F: NA, 54.0
		0.216	
		M: 0.96, 0.232	M: NA, 47.7
Ac,Ep,Fi,Hp,Le,Ly,St,Ul,Xc	Dab	F: 1.03, 0.349	F:50.6, 19.2
		M:1.17, 0.342	M: 38.8, 16.1
Ac,Ep,Hp,Le,Ly,St,Ul,Xc	Dab	F: 1.0, 0.414	F: 48.3; 21.9
		M:1.18, 0.398	M: 35.2; 16.5
Liver histopathology-non-specific	Dab	NA	Statistically significant increase in mean FDI level in the assessment period compared with a prior observation period or statistically significant upward trend in mean FDI level in the assessment period

Biological effect	Applicable to:	BAC	EAC
Liver histopathology- contaminant-specific	Dab	Mean FDI <2	Mean FDI ≥2 A value of FDI = 2 is, e.g. reached if the prevalence of liver tumours is 2% (e.g. one specimen out of a sample of 50 specimens is affected by a liver tumour). Levels of FDI ≥2 can be reached if more fish are affected or if combinations of other toxicopathic lesions occur
Macroscopic liver neoplasms	Dab	Mean FDI <2	Mean FDI ≥2 A value of FDI = 2 is reached, e.g., reached if the prevalence of liver tumours (benign or malignant) is 2% (e.g. one specimen out of a sample of 50 specimens is affected by a liver tumour). If more fish are affected, the FDI value is >2
Intersex in fish; % prevalence	Dab Flounder Cod Red mullet Eelpout	5	
Scope for growth Joules/h g ⁻¹ dry wt.	Mussel (<i>Mytilus</i> sp.; provisional, further validation required)	25	15
Hepatic metallothionein µg g ⁻¹ (ww) ¹ Whole animal ² Digestive gland	Mussel edulis Mytilus galloprovincialis	0.6 ^{1*} 2.0 ^{2*} 0.6 ^{3*} 2.0 ^{1*}	
³ Gills Differential pulse polarography	, , , , , , , , , , , , , , , , , , ,	3.9 ^{2*} 0.6 ^{3*}	
Histopathology in mussels	VVbas: Cell type composition of digestive gland epithelium; µm³ µm⁻³ (quantitative)	0.12	0.18
	MLR/MET: Digestive tubule epithelial atrophy and thinning; µm µm ⁻¹ (quantitative)	0.7	1.6
	VVLYS and lysosomal enlargement; µm³ µm⁻³ (quantitative)	VvLYS 0.0002	V>0.0004
	S/VLYS: µm² µm⁻³ Digestive tubule epithelial atrophy and thinning (semiquantitative)	4 Stage ≤1	Stage 4
	Inflammation (semiquantitative)	Stage ≤1	Stage 3
Imposex/intersex in snails	Nucella lapillus	<0.3	<2

[&]quot;Assessment criteria for the assessment of the fish disease index (FDI) for externally visible diseases in common dab (*Limanda limanda*).Ac, *Acanthochondria cornuta*; Ep, epidermal hyperplasia/papilloma; Fi, acute/healing fin rot/erosion; Hp, hyperpigmentation; Le, *Lepeophtheirus* sp.; Ly, lymphocystis; St, *Stephanostomum baccatum*; UI, acute/healing skin ulcerations; Xc, X-cell gill disease.

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