Current status of the development of marine monitoring indicators for microlitter and threshold levels with EU, OSPAR and HELCOM

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Front page photo:	Microplastic particles, examples found in a fulmar stomach, Photo: Jakob Strand
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1 Preface

This DCE note on the current status of the development of marine monitoring indicators for microlitter and threshold levels within EU, OSPAR and HELCOM has been prepared on request of the Danish Ministry for the Environment. The note provides a brief status on some of the relevant ongoing activities in different international frameworks involved in monitoring and assessment of microlitter in the marine environment.

2 Introduction

In recent years, increasing attention has been paid to marine litter, including microplastic in the marine environment. Microlitter (particles < 5mm) is classified in the categories 'artificial polymer materials' and 'other'. Microlitter comprises a variety of materials (e.g. plastics, metals, rubbers and glass), though many of them are not considered in the current definitions of microplastic. Although other properties may be considered in the definition (e.g. chemical properties, degradability and physical state), the MSFD TG ML agreed to define microlitter in terms of particle size in order to facilitate the classification, harmonisation and comparability of data. Therefore, the recommended definition of marine microlitter is derived from the general definition of marine litter as follows:

"Marine microlitter is marine litter with a length of its maximum dimension below 5 mm".

Most monitoring studies have mainly reported microlitter data for microplastics and sometimes also microrubbers for sediments, water and biota globally and also in European waters, incl. Denmark. In Europe, the development of marine indicators for amounts and composition of microlitter, mainly as microplastic, is of high priority due to the implementation of EUs Marine Framework Strategy Directive (MSFD). Similarly, the regional sea conventions for the Baltic Sea (HELCOM) and for the Northeast Atlantic, including the North Sea (OSPAR), have also acknowledged marine litter and microplastic as part of their monitoring and assessment programs.

With regard to MSFD indicators, microlitter is addressed as part of Descriptor 10 for marine litter. Microlitter is specifically mentioned under the criterion D10C2: "The composition, amount, and spatial distribution of microlitter on the coastline, in the surface layer of the water column, and in seabed sediment, are at levels that do not cause harm to the coastal and marine environment" according to the Commission Decision 2017/848/EU (EU, 2017). In addition, it is noted that "microlitter shall be monitored in a manner that can be related to point-sources for inputs (such as harbours, marinas, waste-water treatment plants, storm-water effluents), where feasible". In the latest Article 8 MSFD CIS Guidance Document (EU, 2022), the text has been slightly modified to "microlitter is monitored in the surface layer of the water column and in the seabed sediment and may be additionally monitored on the coastline". The GES Decision for D10C2 sets out 'artificial polymer materials' and 'other' as assessment elements for microlitter (particles <5 mm). For reporting, the EU enumeration list includes the option to use all types of microlitter as an element. This allows for assessing the status and the extent to which Good Environmental Status (GES) is achieved. A starting point should be the development of standardised monitoring methods, e.g. if monitoring of microlitter (and mesolitter) on coastlines is implemented as well.
In addition, D10C2 on microlitter is also mentioned under the secondary criterion D10C3: "The amount of litter and microlitter ingested by marine animals is at a level that does not adversely affect the health of the species concerned". However, D10C3 elements on microlitter will not be discussed more thoroughly in this note.

Microlitter and microplastics are also considered within the monitoring and assessment frameworks of the regional sea conventions for the Baltic Sea (HELCOM) and the Northeast Atlantic, incl. the North Sea (OSPAR). Currently, the OSPAR ICG-ML expert group has proposed a candidate indicator for microlitter in sediments. The reason is that the abundance of microplastics in biota and surface waters only represents snapshots of the occurrence of microplastics in the environment, while sediments are more stable matrices for both short and long-term monitoring of microplastics in the marine environment. With relevance for HELCOM monitoring, candidate indicators for microlitter are also under development for both the surface layer of the water column and sediment. Furthermore, microplastic in marine water, as well as in sediment, are also identified as primary indicators for monitoring and assessment of plastic pollution in the Arctic (AMAP, 2021).

However, further elements need to be considered before microlitter indicators can be implemented into the different international marine monitoring frameworks described above. The following checks must be fulfilled before internationally coordinated monitoring and assessments can be mandatory for example within OSPAR CEMP monitoring, e.g. marine monitoring of hazardous substances (OSPAR, 2019).

- Established monitoring protocols for sampling and analyses of microlitter.
- Quality assurance and quality control (QA/QC), both internal and external procedures need to be in place.
- Coordinated arrangements for data submission and management, preferably in international databases that can host and secure the monitoring data on applied methods and measured variables.
- Assessment tools such as assessment criteria/threshold values for assessing the environmental conditions and, where relevant, procedures for aggregation or integration of data prior to assessment.

Several initiatives have in recent years made significant progress on these important elements and with links to the international monitoring and assessment frameworks. The next chapters summarise some of the relevant progress made.

3 Status on monitoring protocols for sampling and analyses

Three important aspects need to be addressed in the development of monitoring guidelines for monitoring microplastics in the marine environment (Setäla et al., 2019):

- How to carry out the field sampling.
- How to eliminate other particulate matter in the sample without harming the plastic.
- How to accurately identify and quantify the particles.

In addition, guidelines should also describe relevant precautionary requirements to be considered throughout the entire monitoring procedure. It is important to prevent and assess the contamination of the samples at each step of the process, from sampling to analyses, as described in Chapter 3.

These aspects are also covered in the different monitoring guidelines that are currently under development for MSFD monitoring, and the regional sea conventions OSPAR and HELCOM, as listed in Table 1. Many of the important elements of these draft guidelines are harmonised according to methodological requirements and the list of variables to be reported. In addition, an ISO guideline is under development (currently only as a draft) for analysing plastic in water samples. The ISO guideline addresses water samples in general (e.g. drinking water, ground water and wet deposition) and it may therefore also become relevant for analysing marine water samples and other types of surface waters.

It should be noted that the target size fractions for microlitter vary among the guidelines and matrices. Consequently, the recommended methods for collection of samples, sample handling and analytical tools applied for identification and quantification of microlitter in environmental samples vary as well. These elements are also relevant to consider with regard to important elements as for instance complexity of different matrices and required sample amounts during analyses that are linked to analytical limits of detections and therefore also important when assessing indicator robustness needed for trend assessments.

Therefore, harmonisation of the different guidelines should be considered. For instance, better data harmonisation between environmental data and input/leakage data may become relevant for point sources such as effluents and riverine inputs.

The status of existing international guidelines relevant to microlitter/microplastic monitoring in marine environments in Europe, including Denmark, is listed in Table 1.

Table 1. Overview of existing international guidelines relevant to microlitter/microplastic monitoring in marine environments

International frameworks	Monitoring indicators on microlitter/micro-plastic	Current status on protocols
EU MSFD	D10C2 indicators for microlitter in water, sediment and potentially at beaches. D10C3 indicator microlitter ingested by biota.	Draft protocols on guidance for MSFD monitoring microlitter in marine environments. Latest available versions from December 2022 from JRC/TGML.
HELCOM	Candidate indicator for MP in water column and sediment.	HELCOM guidelines on monitoring of microlitter in the water column in the Baltic Sea. HELCOM (2022a). HELCOM Guidelines on monitoring of microlitter in seabed sediments in the Baltic Sea. HELCOM (2022b). These guidelines have been developed within the frame of the HELCOM BLUES project.
OSPAR	Candidate indicator for MP in sediment.	Draft guidelines for the monitoring of microlitter (including microplastics) in seafloor sediments for the OSPAR Maritime Area. Latest available version from December 2022 from OSPAR ICG-ML MPEG.
ISO	Plastics (including microplastics) in waters and related matrices.	Draft guideline ISO/NP 16094-1 for analysis of plastics in water. Latest available version from July 2022.

The upper size limit of <5 mm is equally important as the lower size limit. According to the current drafts for monitoring protocols from EU MSFD, HELCOM and OSPAR, the prioritised lower size limits for the microlitter particles as mandatory elements are 300 µm and 100 µm for microlitter in surface waters and sediment, respectively. According to these guidelines, additional data for microlitter particles in smaller classes with lower size limits of e.g. 50 µm or 20 µm can also be reported optionally. Even though the amounts of detected particles generally increase significantly when analysing lower size fractions, larger size classes of microlitter have been prioritised. With regard to the ISO guideline, the two size classes, 1 – 5 mm and 1 µm – 1000 µm, are currently identified for microplastic.

It is not fully clear in the guidelines whether the size limits for certain size classes are based on mesh size cut-off values determined by sampling gear (as in manta trawl) and by sieves applied during sample preparation or based on actual measurement of length (e.g. Feret maximum) of individual microlitter particles, e.g. by using imaging software or microscopy. The scientific community has not fully agreed on this yet, and both ways are currently applied even though it can add an additional element of uncertainty when comparing and aggregating monitoring data. Further decisions on this may become relevant before the different monitoring guidelines are finalised.

4 Development of QA/QC procedures

An important element when setting up an international framework for monitoring is having adequate procedures for quality assurance and quality control (QA/QC) as there are for other environmental monitoring variables. With regard to microlitter monitoring, several measures to reduce air contamination, cross-contamination and contamination control must be taken during both field sampling and laboratory analysis. In addition to precautionary actions taken during the handling and analyses of the samples, additional procedures for QA/QC are also needed for documenting the laboratory performances, including verifying the liability and reproducibility of the generated data. The latest recommendations include both internal and external QA/QC procedures. Reporting of relevant QA/QC data for blank samples and reference samples together with the actual microlitter data for the analysed environmental samples are also important for assessing the validity of the generated monitoring data.

Blank samples include both field blanks and laboratory blank samples for assessing the potential contamination of the samples, either during sampling or handling of the sample in the laboratory. In addition, such blank samples can be used for determining the limits of detection (LOD) of microlitter. For instance, in the draft guidance from TGML, it is recommended to use the formular LOD - mean + 3 x standard deviation of the particle concentration in the field and laboratory blank samples. Currently, similar methods are often applied as the ones used for LODs for contaminants. However, the blank data for microlitter are usually not normally distributed around a mean value, as they are counts for numbers of particles, often 0 or only a few in numbers.

Reference samples can be used for determining recoveries for the applied extraction and identification methods, as they reflect the efficiency of the respective laboratory protocols and are treated in the same manner as the samples throughout all steps. Depending on the scope of the analyses, QA/QC data can be reported for both size categories, morphologies and polymer composition of the particles to be detected. Both internal and external reference samples can be analysed in parallel with each sample series. An important prerequisite for this type of quality assurance method is commercially available certified reference materials (CRMs), which are currently lacking for microlitter analysis. Internationally, there are ongoing efforts for developing different approaches for producing adequate CRMs. One of the main challenges is to produce reference samples, where microplastic particles are distributed almost completely equally in a homogenised sample that is needed for producing reproducible results. For instance, as part of the EU-funded project EUROqCHARM (www.euroqcharm.eu), multiple tests have been performed on ways to produce and use "soda" tablets containing several polymers in specific size fractions. Such "soda" tablets can potentially be used as reference materials after being dissolved in water or other types of environmental matrices. To my knowledge, other institutions have also been working on other methods for preparing adequate reference samples for microplastic, e.g. JRC (Seghers et al., 2021).

International intercalibration studies are another important frameworks as external QA/QC procedures, where laboratory performance is assessed by comparing results between different participating analytical laboratories.

Below are some examples of such types of intercalibration exercises. The general outcome has been that a high variation occurs between the participants' results. An important element is that different methods are applied for both sample handling, identification and quantification of microplastic between the participants. Consequently, more efforts are required on harmonisation or even standardisation of the applied methods, which still need further development to be fully established.

Examples of some international intercalibration exercises from recent years:

- QUASIMEME/NORMAN Interlaboratory Studies on the Analysis of Microplastics in Environmental Matrices with three separate rounds organised (Round 1 – 3 , 2019-2022), covering water, sediment and/or biota samples, Organised by: WEPAL-QUASIMEME: https://participants.wepal.nl/participation/index.php

- JRC/BAM Proficiency test. Microplastics in drinking water and sediments, Round 1, 2021. Organised by: JRC (IT) and BAM (DE): https://joint-research-centre.ec.europa.eu/jrc-news/towards-reliable-measurement-microplastics-water-2021-11-16_en

- MEDCIS project, https://www.sciencedirect.com/science/article/pii/S0025326X20302150?via%3Dihub

- SCCWRP intercalibration 2019. https://globe.setac.org/towards-harmonized-methods-to-measure-microplastics/

5 Status on international monitoring databases

Internationally coordinated arrangements for data submission and management are important elements for comparing and aggregating data for wider regional assessments at the EU level. Therefore, there is a need for international databases that can host and secure the data on applied methods and measured variables. There are currently two main international databases with the capacity to host Danish and other European monitoring data for microlitter/microplastics in the marine environment:

- ICES DOME database: https://www.ices.dk/data/data-portals/Pages/DOME.aspx. Data are reported according to the Environmental Reporting Format version 3.2 in order to be quality controlled and entered into the database.

- EMODnet database: https://www.emodnet-chemistry.eu/marinelitter. From EMODnet, the data are fitted into the SeaDataNet for pan-European infrastructure for ocean & marine data management using following proposal for submission guidelines: https://nodc.ogs.it/catalogs/doidetails;jsessionid=3EEEC27DE015377DE7AA2613F3F96E10?0&doi=10.6092/d3e239ec-f790-4ee4-9bb4-c32ef39b426d

These databases are constructed to contain relevant metadata, such as station description, sampling methods, applied analytical methods and data originator as well as the actual data measured for microlitter/microplastics in the environment. The relevant information and measured data are reported into the databases using the vocabularies based on different code lists. The reporting formats have also established which information and parameters to be reported are regarded as mandatory or supplementary or optional. Parameters and related attributes are under continuous development. Therefore, it is recommended to consult the latest tables and vocabularies online at EMODnet or ICES.

Regarding the options for data flow of national monitoring data to the EMODnet database, data can either be submitted directly using the EMODnet ingestion portal or can initially be submitted to the ICES DOME database, wherefrom EMODNet has access to harvest the data. ICES DOME does not (yet) have the same possibility to gather data from EMODnet. ICES DOME is currently also used as the main international database storage for other marine monitoring data, e.g. from the Danish NOVANA monitoring program, where the regional sea conventions OSPAR and HELCOM have direct access to the data.

Currently, the EMODnet database contains the largest dataset for microlitter in surface waters, with currently 1476 data entries for the period 2015-2021 mainly submitted by EU member states after a data call for MSFD relevant assessments. Other datasets, e.g. for microplastics in sediment, have also been submitted by some member states to the EMODnet database after being uploaded using the EMODnet ingestion portal.

With regard to data on microlitter/microplastic ingested by biota, neither the ICES DOME database nor the EMODnet database have developed an adequate report format covering the relevant metadata. Currently, only OSPAR database ODIMS (https://odims.ospar.org) contain such types of monitoring data, more specifically for the seabird indicator: plastics ingested by the seabird fulmars. The ODMIS data format for plastic ingested in fulmars contains the basic information used for e.g. OSPAR QSR23 assessments. However, some important information is also missing, e.g. on sampling events and methods, analytical methods and data originator, and the raw data for the different types/morphologies of microplastic as only "user plastic" and "industrial plastic" are included. The ICES DOME format will usually require such types of additional information as mandatory.

Currently, both the EMODnet and ICES DOME databases are still in a process that can lead to modifications of the final data formats. Among others, it is considered to prioritise a report format for microlitter data for individual particles rather than aggregated data for certain size fractions, particle types/morphologies or polymers. However, this data strategy has not yet been fully decided within the international expert groups. Another factor is if more simplified report formats can be developed as a more user-friendly tool for submitting data into the databases. More simplified data formats (e.g. in excel-formats) can also be considered as an element that can facilitate the submission process for the data owners/originators and thereby increase the data amounts submitted into the databases.

An ICES Workshop on revising the DOME litter data format (WKLIDA) was organized in January 2023. The workshop focused on the monitoring community's needs for reporting litter data and quality assurance information compared to the present possibilities in the ICES Environmental Database (DOME) as well as harmonisation with the EMODnet Chemistry standards for litter reporting and looking into how this can be improved in ICES DOME. Several elements were identified at the workshop that can improve the ICES DOME database formats for reporting microplastic data for different compartments like water, sediment and biota. This included both basic information required for reporting data in individual particles as well as missing vocabularies on the code lists. It was therefore recommended that new data submissions should wait until these modifications of the current DOME format for reporting microplastic data have been revised. This will probably first happen during the next year, among others because such changes have to be approved by the relevant expert groups like ICES WGML and OSPAR ICG-ML.

6 Status on development of threshold levels for microplastic in the marine environment

Environmental status assessment can be performed using different approaches by analysing monitoring data against specific targets, or desired state can be assessed on a qualitative or quantitative basis. The main approaches for target setting are illustrated in Figure 1:

(1) threshold level,

(2) distance to target

(3) directional/trend-based

(4) a baseline value against which to measure change.

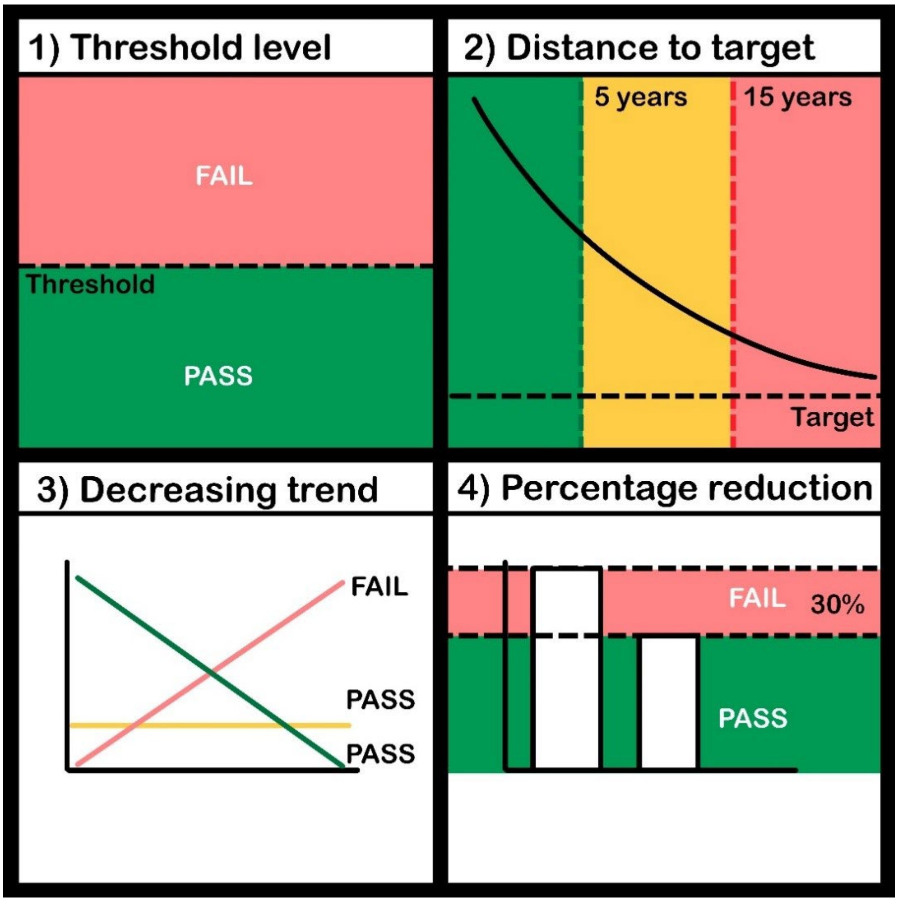

Figure 1. The four main principles identified for target settings for litter and microplastic (Copied from OSPAR MPEG draft guideline on candidate indicator for microlitter in sediments, version December 2022)

The setting of target values as `threshold values' (TVs) for monitoring indicators is, therefore, an important element in assessments of the environmental conditions based on monitoring data. As described in MSFD Commission De monitoring schemes, that TVs are derived for microlitter in the relevant compartments, i.e. for both D10C2 and D10C3. For D10C2, it is more specifically described as "Member States shall establish threshold values for these levels through cooperation at Union level, taking into account regional or subregional specificities" (EU, 2017).

Currently, threshold values remain to be developed at the EU level for all the relevant environmental compartments for microlitter under D10C2, i.e. for the surface layer of the water column, sediment and on shorelines (Table 2).

Table 2. Current status of the development of threshold values for microlitter, updated table from JRC TV report presented 13. December 2022 by JRC at TGML meeting.

D10 Criterion	Compartment	Agreed threshold methods	TVs available	Comments
D10C2 micro-litter	Surface layer of the water column	Baseline under develop-ment	No	Discussed in TGML
	Seabed sediment	No	No	Discussed in TGML, Collection of microlit-ter data in progress
	Coastline	No (Pellets under devel-opment)	No	Discussed in TGML

According to Article 8 MSFD CIS Guidance Document (EU, 2022), it is mentioned that in the interim, for the compartments which the Member States decide to assess, the assessment should be based on trend analysis to detect the evolution or tendency of amounts of microlitter (all). With regard to the guidance for D10C3, it is stated that the threshold values for D10C3 will be developed by the Member States through cooperation at the (sub)regional level. To date, only a threshold value is available for the ingested litter in fulmars in the North-East Atlantic: 'Over a period of at least five consecutive years, no more than 10 % of northern fulmar (*Fulmarus glacialis*) in samples of at least 100 birds may exceed the level of 0.1 g plastic particles in the stomach', whereas a similar type of threshold value is under development for plastic ingested by sea turtles.

The application of these TVs for MSFD assessments is more thoroughly described in the Article 8 MSFD CIS Guidance Document (EU, 2022) as; "For each parameter monitored (amount on the coastline, in the surface layer of the water column and on the seafloor) for D10C1 and D10C2, use survey data per litter categories concerned over time and space. Combine the litter categories data for macro-litter (all) (D10C1) and microlitter (all) (D10C2) per compartment used. The parameter outcomes are assessed against threshold values. In the absence of a threshold value, the achievement of the parameter is assessed by trend analysis. The combination of parameter outcomes for D10C1 and for D10C2 depends on the number of compartments used, i.e. on the extent to which compartments, in addition to those for which GES Decision requires monitoring, are assessed."

With regard to setting threshold levels for marine litter, including microlitter, different options and concepts can be considered. For instance, Werner et al. (2018) described a range of options as a non-exhaustive list to stimulate discussion (the order does not represent a prioritisation) as listed below:

1) The zero option: The setting of a TV of zero litter, even in the long-term, may not be a realistic/operational option because it would not be achievable. However, it can still be treated as a reference condition considered as the ultimate goal to achieve.

2) Points-of-no-return and tipping points: Both approaches, point-of-no-return and tipping points, are only applicable to population-level effects. Since they both refer to a status that is opposite to GES, being points of no return in terms of values, above which harm is already occurring, they are not options

for setting ML thresholds. In addition, there is currently not enough data available to support such an approach.

3),rRecautionary approach: There is vidence that increasing numbers of species are experiencing encounters with ML with multiple/numerous consequences. However, conclusive scientific knowledge on the quantitative relationship between the amount and the exact rate of harm caused by ML is currently only partially available, especially regarding sub-lethal effects. As research is currently unable to provide clear TVs, an initial approach to setting TVs should make use of the precautionary principle, thus providing maximum protection against adverse effects by introducing large safety margins. This could be based on an expert judgement approach (a subjective opinion based on scientific evidence) by eliciting the expertise of a wide range of qualified contributors to make sure that such judgement is demonstrably robust and explicitly stated. Currently, models for using expert judgement are under development, especially in quantitative risk assessment. This approach should be used especially for microplastics, tackling their input from various sources. This approach may, for instance, allow the setting of thresholds if the quantitative concentration/risk relation is not known, but there are indications of risk.

4) Pristine conditions: Another way is the concept of using the same litter pollution levels from pristine or near-pristine areas as TV, as was done for the OSPAR litter in fulmar stomachs indicator. This means accepting a slight deviation from the reference (pristine) condition by using the situation of least pollution found elsewhere in the environment of concern.

5) Cut-off values: Can be defined as a proportion or a percentile in relation to reference conditions, averages or maximum concentrations. The rationale for this approach is that the lower concentrations, which already exist in certain areas, should be the goal for other areas. This concept would be supported by the requirement of an equal level of protection in all areas and seas. For instance, 10^{th} percentile can be used for determining background concentration for metals according to the TGD for derivation for environmental quality standards (EU, 2019). An approach using percentile cut-off values was also applied for setting the beach litter threshold value in combination with an expert judgement approach for the risk of environmental impacts (van loon et al., 2021). In appendix 2, an example is shown of the descriptive statistics using percentiles on a global data set on concentrations of floating microplastic in the size class 300 µm – 5mm in the surface layer of the water column from a systematic review by Bohdan (2022). This study finds microplastic concentrations between 0 and 20 particles per m^3 with 10^{th} and 25^{th} percentiles of 0 particles and a median value of 0.031 particles per m^3. Other field studies targeting smaller size fractions for microlitter particles <100 µm have documented that number of particles can be 1-2 orders of magnitude higher compared to the size fraction >300µm (e.g., Montoto-Martinez et al., 2022; Kuddithamby et al., 2022).

6) Lowest endpoint: In this case, the threshold is set to the lowest concentration causing an adverse effect on one of the specific types of harm. For ML, this denotes that, while it might be difficult or impossible to decide on the relative importance of different adverse effects such as toxicological, entanglement or socio-economic, the lowest TV will be most relevant. Thus, if a TV based on a perceived disturbance by beach visitors is occurring at a lower litter abundance level than other impacts, it would determine the threshold

level. However, this option requires substantial knowledge of the different adverse effects before it can be applied in practice. An overview of the lowest effect levels extracted from the "Toxicity of Microplastics Explorer database" (Hampton et al., 2022) is highlighted in Appendix 1. The lowest observed effect concentrations for the size fraction 100µm - 1mm correspond to 20.300 particles per m^3 and 62.5 mg per m^3 of microplastics. These concentration levels are orders of magnitudes above what has been reported for this size category in the literature for the occurrence of microplastic in the marine environment, as mentioned under 5) for cut-off values.

7) Non-deterioration: The setting of absolute TVs at EU level should not allow increases in litter pollution in countries, where litter pollution levels are already below the TV. This is especially important as we struggle to determine thresholds for microplastics. There is no consensus yet on how risk assessment for microplastics should be conducted, and attempts are currently hampered by a lack of data. It is therefore important to stress that, especially for microplastics, there should be no allowances for increases. A combined approach of non-deterioration, based on existing data, alongside a precautionary approach with proposed TVs, which are reviewed when new scientific and monitoring data are available, is a possibility.

There is an increasing urgent need to have threshold levels ready for assessing microlitter in the marine environment, as the microplastic indicators are beginning to be implemented more widely into national and international monitoring frameworks, such as EU MSFD and the regional sea conventions. Subsequently, decisions on adequate options for deriving threshold values must be made. The next step for international expert groups will be to recommend an adequate approach for deriving relevant threshold levels for microplastic for assessing environmental conditions such as GES in the marine environment. However, before any developed assessment criteria can become fully operational, this will also require that harmonised monitoring schemes, including common protocols and appropriate QA/QC schemes, are widely adopted. In addition, accessibility of large monitoring datasets for microlitter from European waters or regional seas in the appropriate databases such as EMODnet or at ICES will also become an important prerequisite for evaluating such types of threshold values. Currently, there are still outstanding issues before the challenges with covering all relevant microlitter size classes and environmental compartments are resolved.

7 Conclusions

In recent years, progress has been made on several important elements required before internationally coordinated monitoring and assessments of environmental indicators for microlitter/microplastics can become mandatory within the international monitoring frameworks, such as EU MSFD and the regional sea conventions OSPAR and HELCOM. This includes progress on establishment of common monitoring protocols, quality assurance and quality control (QA/QC), coordinated arrangements with international databases and development of assessment tools, e.g. assessment criteria/threshold values for assessing the environmental conditions.

Different monitoring guidelines are under development for MSFD monitoring as well as for monitoring within the regional sea conventions OSPAR and HELCOM. Many of the important elements in these draft guidelines are harmonised according to methodological requirements and the list of variables to be reported. In addition, an ISO guideline is also under development for analysing plastic in water samples, including surface waters.

An important element when setting up an international framework for monitoring is having adequate procedures for QA/QC. Both internal and external QA/QC procedures are needed. Currently, most analytical laboratories rely on internal procedures. There are also initiatives to develop adequate frameworks for international intercalibration exercises that can be used as external procedures, but certified reference materials are still not commercially available.

There are two main international databases with the capacity to host Danish and other European monitoring data for microlitter/microplastics in the marine environment, EMODnet and ICES DOME. Currently, EMODnet contains most data, but not all relevant microlitter size fractions and environmental compartments are adequately covered. Both the EMODnet and ICES DOME databases are still in a process that can lead to modifications of the final data formats.

With regard to setting threshold levels for marine litter, including microlitter, different potential options and concepts have been described for setting thresholds for assessing the environmental status. For microlitter, there are still unresolved issues to handle before the analytical challenges for covering all relevant microlitter size classes and environmental compartments are resolved.

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Appendix 1. Summary of some available microplastic toxicity data

The tables below summarise information on toxicity data for different size fractions of nano- and microplastics selected from "Toxicity of Microplastics Explorer database" using the data file "43591_2022_40_MOESM2_ESM" (Hampton et al., 2022) and using the following search criteria: 1) Size category, 2) Effects detected: Yes, 3) Adverse effects (selecting the specific endpoints for growth, development, reproduction, behavior and mortality); 4) Excluding non-adverse endpoints for all biomarkers based on molecular and RNA expression responses.

In the tables below, the lowest concentration of nano-/microplastics based on particles per volume and mass per volume of each taxonomic groups are listed divided into the size categories 1nm - <100nm, 100nm – <1 µm, 1µm – <100µm, 100µm – <1mm and 1mm - <5mm

Table A1.1. Toxicity data with lowest effect concentrations for size category 1nm – <100nm for different taxonomic groups in the "Toxicity of Microplastics Explorer database" (extracted from Hampton et al., 2022).

1nm - <100nm	Number of entries for selected endpoints	Lowest Concentration (Particles/mL)	Lowest Concentration (µg/mL)	Effect Detected ?	References
Plants	1	5.72E+12	1100	Yes	doi.org/10.1021/es503001d
Rotifera	11	1.43E+06	1.00E-04	Yes	doi.org/10.1021/acs.est.6b01441
Crustacea	21	4.95E+06	0.001	Yes	doi.org/10.1016/j.envpol.2019.113506 0269-7491
Mollusca	28	1.43E+09	0.1	Yes	doi.org/10.1016/j.envpol.2018.08.020
Fish	1	1.72E+10	1	Yes	doi.org/10.1016/j.scitotenv.2017.01.156
All	62	1.43E+06	1.00E-04	Yes	

Table A1.2. Toxicity data with lowest effect concentrations for size category 100nm – <1µm for different taxonomic groups in the "Toxicity of Microplastics Explorer database" (extracted from Hampton et al., 2022).

100nm - <1µm	Number of entries for selected endpoints	Lowest Concentration (Particles/mL)	Lowest Concentration (µg/mL)	Effect Detected ?	References
Plants	4	1.78E+10	10	Yes	10.1016/j.chemosphere.2018.05.170
Bacteria	4	1.79E+08	20	Yes	10.1016/j.scitotenv.2018.06.141
Rotifera	6	1.43E+03	1.00E-04	Yes	10.1021/acs.est.6b01441
Crustacea	37	1.78E+06	0.125	Yes	10.1021/es401932b
Echinodermata	3	7.14E+07	5	Yes	10.1021/es502569w
Fish	1	1.00E+03	0.19	Yes	10.1016/j.jhazmat.2019.120861
All	55	1.00E+03	1.00E-04	Yes	

Table A1.3. Toxicity data with lowest effect concentrations for size category 1µm - <100µm for different taxonomic groups in the "Toxicity of Microplastics Explorer database" (extracted from Hampton et al., 2022).

1µm – < 100µm	Number of entries for selected end-points	Lowest Concentration (Particles/mL)	Lowest Concentration (µg/mL)	Effect Detected ?	References
Plants	25	9.91E+03	1	Yes	10.1016/j.scitotenv.2018.03.176; 10.1016/j.envpol.2016.11.005
Bacteria	7	2.43E+07	20	Yes	10.1016/j.scitotenv.2018.06.141
Rotifera	2	7.96E+05	1	Yes	10.1016/j.jhazmat.2018.07.101
Cnidaria	1	9.00E+10	50	Yes	10.1016/j.envpol.2018.08.045
Crustacea	121	7.00E+00	0.0013	Yes	10.1016/j.envpol.2017.11.014; 10.1016/j.envpol.2019.03.085
Annelida	6	1.00E+02	0.056	Yes	10.1016/j.marpolbul.2017.10.072
Echinoder-mata	3	3.00E+02	3.65	Yes	0.1021/es404295e
Mollusca	8	6.41E+02	0.023	Yes	10.1073/pnas.1519019113

100µm – < 1mm	Number of en-tries for se-lected end-points	Lowest Concentration (Particles/mL)	Lowest Concentration (µg/mL)	Effect Detected ?	References
Plants	12	30	100	Yes	10.1016/j.aquatox.2019.105296; doi.org/10.1016/j.envpol.2019.112980
Cnidaria	7	0.020	2.5	Yes	10.1016/j.envpol.2019.113074
Crustacea	15	0.54	0.063	Yes	10.1021/acs.est.7b03574
Fish	1	136	10	Yes	10.1016/j.scitotenv.2017.11.103
All	35	2.03E-02	6.25E-02	Yes	

Table A1.5. Toxicity data with lowest effect concentrations for size category 1mm - <5mm for different taxonomic groups in the "Toxicity of Microplastics Explorer database" (extracted from Hampton et al., 2022)

1mm – < 5mm	Number of en-tries for se-lected end-points	Lowest Concentration (Particles/mL)	Lowest Concentration (µg/mL)	Effect Detected ?	References
Mollusca	24	0.0065	100	Yes	doi.org/10.1016/j.watres.2020.115890
All	24	0.0065	100	Yes	

Appendix 2. An example of available data on microplastic levels in the environment

Table A2.1. Descriptive statistics of microplastic abundance in a global dataset reported in volumetric units, reviewed by Bohdan (2022).

Statistics	**Microplastics m^{-3}**
Median	0.031
1st quartile	0
3rd quartile	0.15
Maximum-value	20
Number of datapoints	701