Report of the Marine Chemistry Working Group (MCWG)

1–5 March 2010

Ostende/Ghent, Belgium

International Council for the Exploration of the Sea
Conseil International pour l'Exploration de la Mer

Recommended format for purposes of citation:

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Executive summary

The Marine Chemistry Working Group [MCWG] (Co-Chairs: Evin McGovern, Ireland and Katrin Vorkamp, Denmark) met at Ghent University, Belgium, 1–5 March 2010. The meeting was attended by 25 participants representing 10 different countries. MCWG worked in a combination of plenary, subgroups and specific task groups.

Following OSPAR work requests to ICES, MCWG continued to provide advice to support coordinated monitoring activities of both **organic contaminants** and **ocean acidification (OA)**. The latter was addressed by a **chemical oceanography subgroup (COSG)** consisting of seven MCWG members and four chair-invited external experts to assist with this particular task. COSG produced a comprehensive document including recommendations on parameters, protocols and quality assurance, initial advice on spatial and temporal coverage of OA monitoring, a review of the present state of knowledge with regard to methods, instrumentation and logistics and identification of key issues that need to be addressed.

Other CO-related agenda points included a plenary presentation by David Hydes of NOC, University of Southampton, UK on nutrient and oxygen dynamics and a presentation and discussion of the **international nutrient scale system**, large-scale production of **reference materials for nutrients in seawater** and measurements of **absolute salinity**.

Regarding organic contaminants, a draft technical annex was prepared for the monitoring of **dioxins/furans and dioxin-like PCBs** in marine sediment and the corresponding technical annex for biota monitoring was revised. Two sets of monitoring guidelines were now completed for publication in the **ICES TIMES series** (Silicone rubber passive samplers and Determination of perfluorinated compounds (PFCs) in marine sediment, water and biota). In response to a further OPSAR request, MCWG also concluded that more scientific information would be needed to understand the atmospheric-marine link before including PFCs in **atmospheric monitoring**. MCWG will follow up on developments in PFC research and monitoring at MCWG 2011.

New information on PFCs in the atmospheric and marine environment was also presented under the agenda point **"emerging contaminants"**. Further presentations on emerging compounds of environmental interest included pharmaceuticals, benzotriazoles, phosphorous ester flame retardants and the brominated flame retardant hexabromocyclododecane. An overview of recent and current research on anthropogenic and naturally produced brominated compounds was given in a plenary presentation by Adrian Covaci, University of Antwerp, Belgium.

Other **projects of general interest** to the group included updates on analytical methods for the determination of methylated PAHs and the mapping of contaminants in sediments around Norway. Both contaminants and nutrients were addressed in task group reports on **Good Environmental Status (GES)** under the **Marine Framework Directive Strategy (MSFD)** which were discussed by MCWG. MCWG also provided comments on a draft version of a **guideline for the monitoring of contaminants** in sediment and biota under the **Water Framework Directive (WFD).** Furthermore, a UK suggestion of **compliance checking** of **Environmental Quality Standards (EQS)** under WFD was discussed and alternative statistical approaches were suggested.

A plenary presentation was given by Colin Janssen, University of Ghent, Belgium on **links between chemistry and biology** in marine monitoring. MCWG's interest in this

field was also mentioned during the discussion of the **cooperation with other expert groups**, as well as links to the working group on marine sediments, the eel working group and the study group on nutrient standards.

Quality assurance (QA) was addressed in a presentation by the **QUASIMEME** representative Wim Cofino. MCWG continues to provide feedback and information exchange with QUASIMEME, a key service underpinning marine chemistry monitoring and research activities. The reporting of **QA information** to the **ICES Data Centre** was discussed following a proposal of alterations presented by the German monitoring laboratories and resulted in several recommendations for updates. Following an ICES Data Centre request, MCWG also provided advice on the reporting of **metal concentrations in seawater**.

1   Opening of the meeting

The Marine Chemistry Working Group (MCWG), chaired by Evin McGovern, Ireland and Katrin Vorkamp, Denmark, met at the University of Ghent, Belgium, from 1–5 March 2010. The meeting was jointly organized by Patrick Roose (MUMM) and Koen Parmentier (ILVO). Patrick Roose welcomed the participants to Ghent and opened the meeting on behalf of his organization.

The participants introduced themselves and their affiliations and described their specific interests within the field of marine chemistry. A chemical oceanography subgroup (COSG) was formed, consisting of Naomi Greenwood, Caroline Kivimae, Klaus Nagel, Solveig Olafsdottir, Carmen Rodriguez and Elisabeth Sahlsten to deal with CO-specific agenda points. They were joined by David Hydes, David Pearce and Alberto Borges on the Tuesday and Wednesday to focus primarily on agenda point 5.h (Section 5.8 of this report).

2   Adoption of the agenda

The agenda was adopted as presented in Annex 2. There were considerations of removing point 5g from the agenda, but it was decided that COSG would discuss whether or not this point should be kept on the agenda and if so, how it should be addressed at the next meeting.

3   Report of the 97th ICES Statutory Meeting

Evin McGovern highlighted the ICES JAMP Review Group's comments on advice provided by MCWG 2009 and other working groups report in response to OSPAR work requests to ICES (Annex 17 of the MCWG 2009 report).

Evin also highlighted that the MCWG 2009 report was presented to ICES SCICOM Steering Group on Human Interactions on Ecosystems SGHIE at the Annual Science Conference (ASC) in Berlin in September 2009. The heavy work load of MCWG and the efficient response to work requests were at SGHIE and the appreciation expressed by the ICES secretariat and the Chair of SGHIE was conveyed to MCWG 2010.

It was highlighted that there is one theme session of direct relevance to MCWG members at ASC 2010 (20–24 Sept, Nantes). Session F "Monitoring biological effects and contaminants in the marine environment: where do we go from here", convened by John Thain (UK), Catherine Couillard (Canada) and Dick Vethaak (The Netherlands), has particular relevance in the context of emerging MSFD developments. MCWG members are encouraged to consider submitting an abstract for this theme session by 15 April 2010.

4   Plenary presentations

Three speakers had been invited by the Chair to present their work at the meeting.

4.1   Colin Janssen: Quo vadis? Linking biology and chemistry in marine monitoring

Professor Dr Colin Janssen from the University of Ghent had been invited by the Chair to give a presentation at MCWG 2010. The suggested topic on linkages between

biology and chemistry was very welcomed by MCWG. As an agenda point it has been addressed repeatedly and MCWG has followed developments in this field with great interest.

The presentation consisted of i) a sneak preview of a position paper prepared by the Marine Board of the European Science Foundation (MB-ESF) and dealing with emerging substances and ii) results of a Belgian study (INRAM, http://www.vliz.be/projects/inram) dealing with integrated chemical, biological effects and ecological monitoring.

The MB-ESF position paper evaluates the present state of marine environmental monitoring in relation to hazardous substances and the various selection mechanisms that are or have been used to identify them. The paper also discusses emerging substances and gives two case studies, one on the evaluation of the offshore industries and one in relation to MARPOL. The main conclusions are the discrepancy in the number of chemicals considered by the various monitoring programmes, a lack of harmonization in selection procedures for hazardous substances, the fact that these mechanisms are hazard-based rather than risk-based and a lag between the scientific consensus about the risk of a substance and its introduction in routine monitoring. Furthermore, the processes should be science-based, as much as possible, and dynamic allowing both the selection of new substances as well as the elimination of existing ones. Also, data gathering and data accessibility is seen as an area for improvement. The paper will be published later in 2010.

With INRAM, Belgian scientists attempt the integration of the major fields of expertise (chemistry, biological effects and ecology) in order to assess the state of their coastal environment. The project also assesses the role of harbours as a source of contaminants to the coastal waters. Novel is the extensive use of passive samplers (PS) both to determine dissolved concentrations of chemicals and as a source of contaminants for in-vitro tests. PS, which were deployed for about 6 weeks, are used to expose test organisms to relevant concentrations of the mixtures found in the environment. The initial results show a clear toxicity of the samplers and a link with the contaminants. Both need to be further investigated and the results of this are expected in 2011. These observations are, to an extent, backed by the results of biological effects techniques. There are also indications of a relation between the condition of test organisms exposed during cage experiments, the species composition of the benthic community and the pollution levels. Professor Janssen sees great value in the use of PS for environmental monitoring and believes that they are ready for a more general application.

4.2 David Hydes: Portsmouth-Bilbao Ferry-Box: Inter-annual and seasonal dynamics in nutrients and oxygen

Dr David Hydes from the National Oceanography Centre (NOC), Southampton had been invited by the Chair to give a presentation at MCWG 2010. With several agenda points on chemical oceanography, it was of particular interest to the group to be informed about state-of-the-art data collection.

David Hydes first presented the general idea of ships of opportunities, i.e. automated instrument packages installed on cargo ships and ferries. Compared with monitoring of surface waters using buoys, piles and platforms, ships of opportunities have several advantages, e.g. reduced risk of biofouling, power supply, sheltered placement within the ship, easy maintenance in harbours and data collected along-transects instead of single locations. Especially ferry-boxes, i.e. analytical instruments on ferries

operating regular routes, have generated a multitude of oceanographic data, among these data on eutrophication, temperature and sediment transport etc. Details on European ferry-box routes can be found on the website www.ferrybox.com. An analogues project in North Carolina (USA) is www.ferrymon.org.

Since 2002, NOC has run the ferry-box data collection on board the Pride of Bilbao, a P&O ferry operating between Portsmouth (UK) and Bilbao (Spain). Data is transmitted to NOC using the Orbcomm satellite system and displayed on the NOC webpage in real time (www.noc.soton.ac.uk/ops/ferrybox_index). Originally, water temperature, salinity and fluorescence were measured. In 2005, extra sensors measuring turbidity, the concentration of dissolved oxygen and the partial pressure of carbon dioxide were fitted along with sensors monitoring the movement of the ship.

4.3 Adrian Covaci: Anthropogenic and naturally produced brominated compounds in the marine environment

Dr Adrian Covaci of the University of Antwerp had been invited by the Chair to give a presentation at MCWG 2010. His presentation on brominated compounds in the marine environment was of great interest for many MCWG members.

Polybrominated diphenyl ethers (PBDEs) are one group of important anthropogenic compounds among the brominated flame retardants (BFRs). In the past, they have been used extensively in electronic equipment, textiles, household products and transportation. Although the use and production of commercial mixtures have been banned in the EU since 2004 (Penta-BDE and Octa-BDE) and 2008 (Deca-BDE), PBDEs are still widespread and ubiquitous contaminants in humans, aquatic and terrestrial wildlife (Law *et al.* 2006). Furthermore, PBDEs have been shown to biomagnify in marine food chains, with high levels measured in apex predators, such as marine mammals.

Recently, the increased attention has been given to the presence of naturally produced brominated compounds, including structural analogues of PBDEs (hydroxylated and methoxylated PBDEs, HO-PBDEs and MeO-PBDEs, respectively) or polybrominated hexahydroxanthenes (PBHDs) in the marine environment.

The bioaccumulation and biomagnification potential to anthropogenic and naturally produced BFRs will be discussed in greater detail in harbour seals and harbour porpoises, as top predators of the North Sea foodweb. For comparison reasons and to underline species-specific accumulation of contaminants, PCB data will also be presented. An interesting comparison has been made between blubber and blood concentrations, the latter allowing the measurement of major metabolites, such as HO-PCBs and HO-PBDEs, as well.

Differences in the bioaccumulation between anthropogenic and naturally produced brominated compounds have not only been investigated in the North Sea, but also in various other foodwebs, such as foodwebs from Sydney Harbour (Australia), the Mediterranean Sea and the Brazilian coasts. High levels of naturally produced brominated compounds have been found in some of these food chains and these compounds show similar biomagnification potential compared to anthropogenic brominated compounds.

Although the literature is scarce about the toxicity of the naturally produced brominated compounds, their presence in the marine environment and possible synergistic effects, which can lead to an increase in the toxic potential to anthropogenic com-

pounds, are of concern for marine mammals, which are already considered to be particularly vulnerable and sensitive to pollution.

References

Law, R., Allchin, C. R., de Boer, J., Covaci, A., Herzke, D., Lepom, P., Morris, S., Tronczynski, J., and de Wit, C. 2006. Levels and trends of brominated flame retardants in the European environment. Chemosphere, 64: 187–208.

5   Main agenda

5.1   Report on developments with regard to quality assurance of marine chemistry, in particular with respect to QUASIMEME.

Wim Cofino visited MCWG on Wednesday to present information about QUA-SIMEME and to discuss with MCWG.

In his presentation he explained ongoing changes at QUASIMEME and plans for the future. QUASIMEME has joined forces with WEPAL, an accredited proficiency testing organization (Wageningen, The Netherlands). This gives QUASIMEME the benefit of a larger staff (more flexible, more robust) and allows QUASIMEME to become accredited as an extension of the existing WEPAL accreditation. QUASIMEME's plan is to become fully accredited in 2010. As one step in this process, the QUASIMEME method for statistical evaluation of the data has been approved by the Dutch accreditation board.

Actions have also been taken to change the submission of data by Internet, towards a more user-friendly, interactive system. QUASIMEME would like it to become operative this year, but this depends on IT progress. Wim Cofino concluded his presentation with three questions which had been posed to the QUASIMEME office, and asked for MCWG's view.

- There had been comments on the types of filters used for chlorophyll. There was no preference of a specific filter among MCWG members, but the group replied that the filter brand should not be mentioned, instead the characteristics of the filter used, e.g. material and pore size.

- Is there still an interest in passive sampling exercises? The problem is that not many labs use passive samplers on a routine basis and that there are many types of samplers in use. MCWG considered it as unlikely that there would be enough participants for regular passive sampling exercises.

- Should Methyl-Hg be added to the BT-1 exercise (trace metals in biota)? There are some customers, but not many. MCWG replied that for many species near 100% of total Hg can be assumed to be methyl-Hg and this might be sufficient.

The following items were addressed by members of MCWG:

PBDE in sediment and biota: At the moment, these two matrices are included in the same exercise. It was suggested that two exercises should be offered by QUA-SIMEME with biota and sediment, respectively, as some laboratories only need either biota or sediment. However, this was not supported by all members, as those laboratories that analyse both biota and sediment might have to double their resources. It was generally discussed whether this could be solved by providing separate types of materials and letting the participants choose, e.g. two biota, two sediment samples or

one of each. It was not clear how that would affect costs (organization) and performance and Wim Cofino pointed out that the number of results per sample would decrease, which is not desirable. Steven Crum informed later by e-mail that the BS-1 exercise would be divided into separate biota and sediment tests by 2011.

It was commented that deadlines for submission were sometimes changed at a very short notice. This was done by QUASIMEME to receive more data, leading to a better output (assigned values). But it was acknowledged that this could be frustrating for those laboratories that had worked hard to submit the results in time. MCWG appreciated that the communication had improved and that QUASIMEME was aware of its importance.

The suggestion to use freeze dried samples possibly providing high stability, lower storage and shipment cost, was rejected. There is no real gain in efficiency and most laboratories use fresh samples.

Wim Cofino informed that discontinued samples, used in previous rounds, can be bought from QUASIMEME. MCWG members were encouraged to contact the QUASIMEME office if they required specific samples or analysed the determinants in a different context. It has been arranged with some participants, for example, to supply DOC samples without the nutrients, as part of AQ-1 and AQ-2.

MCWG members enquired about international links and plans of extension. There are links with Japan (nutrients in seawater) and with the NORMAN network, but this has not directly lead to an increase in activities. QUASIMEME is currently producing new flyers for promotion purposes.

The discussion showed that participants use QUASIMEME for different purposes and thus have different expectations. Some participants ask for a wide variety of samples of different contaminant level, whereas others prefer the same type of samples every round. QUASIMEME tries to accommodate both preferences.

Wim Cofino addressed further issues that he wanted MCWG to discuss. The background was that it had become difficult to obtain naturally contaminated samples with a large variety of contaminants in adequately high concentrations.

QUASIMEME welcomes suitable biota samples, preferably mussels that could be provided by MCWG members. The costs of collection, transportation will be covered by QUASIMEME. This partner will also receive additional samples, prepared from his material. Approximately 100–120 kg of fresh weight is required as for one batch approximately 22–25 kg of mussel flesh is needed.

QUASIMEME considers the use of freshwater fish which often has higher contaminant levels than marine fish. There was no objection against such an approach to obtain good sample material.

Spiking of samples has generally been discussed, but QUASIMEME hesitates to introduce spiked samples. MCWG also had reservations for this method. A compromise could be an approach where living mussels are contaminated indirectly, by spiking of the water phase. This would result in more natural sample, but it would also be rather expensive. From a scientific point of view, MCWG would prefer this option to spiking of homogenized material.

Wim Cofino proceeded with a presentation about statistical methods; datasets were tested with old and new statistical methods, including the QUASIMEME method. It was demonstrated that different statistical methods could result in very different outcomes, including the mean value of the analyte and thus the Z-score. This was

partly due to the way these methods deal with outliers. The QUASIMEME method was shown to be very applicable and reliable. However, it was emphasized that the preferred number of data entries is 20 or higher.

MCWG thanked Wim Cofino for his presentations and time for discussion and expressed the wish to maintain the close dialogue with QUASIMEME.

5.2 Report on the developments in Water Framework Directive monitoring programmes for physico–chemical parameters (priority substances, other pollutants, nutrient status) in transitional and coastal waters; and developments in defining MSFD GES descriptors (Task groups 5 – eutrophication, TG8 – contaminants and their effects in the marine environment, TG 9 – contaminants in seafood)

Water Framework Directive (WFD)

a) Compliance checking

Peter Lepom was unable to attend this meeting, but had been contacted by the Chair prior to the meeting with regard to recent developments under WFD and reported by e-mail that suitable methods of compliance checking were a topical issue under WFD. As a working document for discussion of suitable methods, he provided a note by the UK, prepared for the WG E meeting on priority substances ("Statistical methods for compliance checking of environmental quality standards").

Katrin Vorkamp presented the document which suggests expressing the Maximum Allowable Concentration (MAC) as a percentile instead of an absolute value. The background was, according to the note, that the expression MAC implied that this concentration must never be exceeded. This does not agree with the discrete sampling as there is always some time that is not accounted for and during which MAC might be exceeded. Furthermore, the more samples are taken, the higher the probability of failure. For this reason, the note suggests using 95% percentile of the MAC and setting limits of confidence of failure.

The group discussed this suggestion in plenary, as well as the calculation of the annual average concentration under the WFD. There was general agreement that the arithmetic mean is a critical parameter for the determination of environmental levels as it assumes a normal distribution of the data, which is rarely given for environmental concentrations. Outliers can occur easily and would have a large effect on the arithmetic mean. The median would be a more robust parameter. Patrick Roose explained that a minimum of monthly samples is required under the WFD. Additional sampling might produce data which are not representative of the whole year, e.g. because of seasonal variations, which also would affect the arithmetic mean.

Regarding MAC, the group remarked that absolute values do not exist, but always include some uncertainty, e.g. natural variation and measurement uncertainty. Situations can occur where values would exceed MAC if their uncertainty was taken into account. There was general agreement that independent of the approach chosen for compliance checking, the uncertainty should be addressed. Regarding the UK note, MCWG was of the opinion that a percentile approach had merits, but there would be other suitable approaches as well. MCWG would be interested in following the development in this area and discussing other forthcoming suggestions.

MCWG will follow up on this item at their next meeting and discuss statistical approaches on the basis of actual environmental monitoring data.

Recommendation

A suitable statistical approach should be chosen for compliance checking, taking into account i) that most environmental monitoring data do not follow a normal distribution and ii) that measurement data include an uncertainty.

b) Guidance on monitoring of sediment and biota under WFD (draft)

A previous version of this draft document was reviewed by MCWG at last year's meeting. As remarked in the MCWG 2009 report, the group welcomes this initiative as sediment and biota offer a practical alternative to water. The group appreciates that the previous comments were taken on board and that MCWG was given the opportunity for further review.

A subgroup, consisting of Koen Parmentier, Michiel Kotterman, Michael Haarich, Victoria Besada and Els Monteyne, worked on this document and inserted their comments. It was agreed with Patrick Roose, a member of the drafting group for this document, that he would bring the MCWG comments to the attention of the drafting group. They are not listed specifically in this report.

The subgroup commented that the draft guideline included the relevant aspects to be considered for biota and sediment analysis and that it had moved closer towards a final document. The following general remarks were made:

- Because the information has to be collected from different domains, there is need for harmonization in terms and expressions and outline of the topics to be addressed. In the current draft, some points are dealt with in various sections (sometimes partly). The draft document would improve if the relevant information was given only once and referred in other sections of the guideline. Repetitions should be avoided.

- The subgroup thinks that in some cases, too many details are given on relevant species and contaminants, thus impeding the reading flow.

- In very few cases, the subgroup has suggested to remove parts of the text (indicated in the annotated document forwarded to Patrick Roose) which appeared questionable or irrelevant.

- Some definitions and verifications are required, as specified in the annotated document.

Recommendation

CMA to consider the comments of MCWG as forwarded to Patrick Roose in the final revision of the biota and sediment monitoring guideline

Marine Strategy Framework Directive (MSFD; Directive 2008/56/EC)

Final reports of three task groups were available for the information of MCWG. These reports addressed three of eleven quality descriptors (QD) to evaluate Good Environmental Status (GES), as stated in Annex 1 of the directive, i.e.

- Descriptor 5: "Human-induced eutrophication is minimized, especially adverse effects thereof, such as losses in biodiversity, ecosystem degradation, harmful algae blooms and oxygen deficiency in bottom waters"

- Descriptor 8: "Concentrations of contaminants are at levels not giving rise to pollution effects"

- Descriptor 9: "Contaminants in fish and other seafood for human consumption do not exceed levels established by Community legislation or other relevant standards"

Katrin Vorkamp presented the report of TG5 to MCWG. The task group provided the following definition of eutrophication "Eutrophication is a process driven by enrichment of water by nutrients, especially compounds of nitrogen and/or phosphorous, leading to: increased growth, primary production and biomass of algae; changes in the balance of organisms; and water quality degradation. The consequences of eutrophication are undesirable if they appreciably degrade ecosystem health and/or the sustainable provision of goods and services." MCWG noted, as also emphasized in the report, that the consequences themselves, i.e. changes in organism balance etc., were only undesirable if they lead to the degradation of ecosystem health and/or provision of goods and services. The report states that harmful algae blooms (HAB) should only be treated as part of the undesirable consequences if they increase in correspondence with increased nutrient input.

Several actions have been taken to address and combat eutrophication in coastal waters, under OSPAR, HELCOM, the Urban Wastewater Treatment Directive and the Water Framework Directive (WFD). Whereas WFD covers coastal waters, MSFD manages open marine waters so there is some overlap. The report notes that most of the offshore areas show limited eutrophication symptoms, however, it also mentions that turbidity near the coast is often too high to allow strong primary production. The group discussed differences in the conceptual approaches of WFD and MSFD, e.g. the "one out, all out" approach under WFD, in contrast to the integration of all (or most) descriptors under MSFD.

The report also emphasizes that this descriptor should not be considered in isolation, but with links to QD1 (Biological diversity), QD4 (Marine foodwebs) and QD9 (see above) with regard to HAB/biotoxins. However, marine biotoxins were excluded from QD9 as well as they are not necessarily linked to human activity.

The report provides a review of existing methods and recommends including other characteristics in addition to chlorophyll a, e.g. changes in community composition, occurrence of toxic species etc. Satellite observations, remote sensing, automatic recordings etc. should be used for open sea monitoring. Models may provide insight into long-range-transport effects. Regarding spatial and temporal scales, the report suggests that annual cycling may be more adequate for marine areas than seasonal datasets and initial screenings may be useful to identify impaired water bodies.

Patrick Roose presented the report of TG8, having participated in the work as an OSPAR observer. "Contaminants" are defined as "substances or groups of substances that are toxic, persistent and liable to bioaccumulate and other substances or groups of substances which give rise to an equivalent level of concern". However, this report also excludes HAB and marine toxins as they are natural products rather than contaminants. This means that HAB are not covered by any of these QDs.

The report also gives a definition of "pollution effects" and recommends the integration of contaminant monitoring and quantitative biological effect monitoring. These data should be interpreted against suitable assessment thresholds, e.g. environmental quality standards (EQS) under WFD or environmental assessment criteria (EAC) as defined within OSPAR. MCWG discussed how this integrated monitoring could be achieved. Examples were to be presented in Colin Janssen's plenary presentation the following day, see above.

TG8 identified a close link with QD9, and additional connections with QD1, QD4, QD6 (Sea-floor ecosystems), QD9 and QD10 (Marine litter). The report presents an extensive literature review of contaminants and biological effects, as well as a detailed description of relevant policies and conventions related to the descriptor. In the following discussion, it was clarified that WFD chemical status applied to the territorial waters where member states have jurisdictional rights, i.e. 12 nautical miles from baseline. MSFD deals only with marine matters that are not already covered by WFD.

Being a member of TG9, Victoria Besada presented the report of this task group to MCWG. In view of QD9, "contaminants" are defined as "hazardous substances present in fish as a result of environmental contamination for which regulatory levels have been set for human consumption or for which the presence in fish is relevant". Contaminants in fish and seafood are monitored against these regulatory levels, if available, whereas other relevant contaminants should be monitored over time.

The report emphasizes a strong link to QD8 and recommends integrating results from QD8 and QD9. It also states that food safety regulatory levels and GES are not linked. Regulatory levels for human consumption are generally too high to be used as indicators of marine pollution. Furthermore, they often lack the necessary data to link the samples to specific regions and might be contaminated during transport, storage or treatment. It can be noted that concentrations above regulatory levels indicate bad environmental status, but the opposite is not necessarily valid.

Following this line of argumentation, MCWG discussed what TG9 added to the evaluation of GES, beyond the other QDs. The group expressed the view that if GES was achieved for QD8, QD9 would be met as well. In order to obtain as much information as possible regarding GES, the report recommends that levels should be expressed as absolute values and not relative to regulatory levels. It also suggests an approach for monitoring of contaminants in fish and seafood for determining GES. MCWG generally welcomed the contact to public health specialists, but also noted that the food and environmental monitoring programmes are designed differently and that important information might get lost if compromises are attempted (ref. MCWG 2006 report Section 8.4).

The elements and compounds currently regulated in fish and seafood are Pb, Cd, Hg, PAHs, dioxins (including dioxin-like PCBs) and radionuclides, on the list of compounds relevant for time-trend monitoring. As is mentioned, which MCWG wondered about as can occur in high concentrations naturally in a non-toxic organic form.

5.3 MCWG members to report information on projects of relevance to MCWG activities

Two presentations were given under this agenda point.

Stepan Boitsov: Hydrocarbons in marine sediments of the Barents Sea and the North-Eastern part of the Norwegian Sea

The Norwegian Institute of Marine Research and the Norwegian Geological Survey have collected 65 sediment cores from the South-West Barents Sea and the areas off the Lofoten and Vesterålen islands in the North-Eastern Norwegian Sea, in the period 2006–2009. The sampling was part of the MAREANO program which includes detailed geochemical, geological and biological mapping of the seabed on the Norwegian shelf. Sediment samples were analysed for a number of geological and geochemical parameters, including grain size, total organic carbon contents, and lev-

els of organic (PAHs, TBT) and inorganic (heavy metals, arsenic) contaminants. A selection of sediment cores was dated by 210Pb-radiodating, and the historical development of contamination trends has been studied. Possible sources include long-range transport of man-made contaminants as well as local natural sources (in case of PAHs). The levels of PAHs in the MAREANO area are relatively low, under 250 µg/kg dry weight for the sum of 49 compounds in surface sediments in South-Western Barents Sea and under 500 µg/kg dry weight in the shelf areas of the North-Eastern Norwegian Sea, whereas deeper areas of the North-Eastern Norwegian Sea (down to 2000 m deep) exhibit somewhat higher levels, up to 2500 µg/kg dry weight for the sum of 49 compounds in surface sediments, due to higher total contents of organic material. These levels are considerably lower than what had previously been found in other parts of the Barents Sea, in particular south of Svalbard (Spitsbergen), where the natural contribution from eroded coal-bearing bedrock leads to PAH levels of up to 6000 µg/kg dry weight in surface sediments. The results obtained under the MAREANO programme are reported every year and are presented as maps on the website of the programme, www.mareano.no, whereas the detailed scientific results are published in international journals.

Michiel Kotterman: Identification and quantification of methylated PAHs by GCxGC–MS

Alkylated PAHs are ubiquitously present in the environment but the analysis of these compounds is not fully established yet, although their contribution to the toxic activity of environmental PAH mixtures is already acknowledged. In this project the identification and quantification of methylated PAHs was studied. The resulting method was used to analyse contaminated Elbe sediment as well as two QUASIMEME samples; shrimp (QPH052BT) and mussels (QPH056BT), in order to compare methylated PAH levels with PAH levels.

A GCxGC-FID method was designed to improve separation of 23 congeners of methylated chrysenes, benz[a]anthracenes and benzo[c]phenanthrene congeners from each other and from other possible interferences. This method was applied and further developed on GCxGC-qMS. Of the 23 standards available for the methylated PAH studies, 14 were fully separated with a combination of 60 m DB5 x 1.5 m LC50.

This study shows that there can be large differences of up to a factor of 10 in chromatographic responses for each congener. This response factor must be taken into account for proper quantification, as it is shown in the literature that different alkylated congeners give different toxic responses.

Using this method we were able to identify different classes of methylated PAHs in Elbe sediment. A total of 12 congeners were identified and quantified. The peaks of the unresolved compounds were also identified, but quantification was not possible. The levels of methyl-PAHs in the biota samples were low, only 3-methylcrysene could be quantified in the shrimp sample. In the mussel sample, higher concentrations were observed and 8 methylated PAHs of the available 23 congeners could be quantified. Although the levels of methyl-PAHs were lower than the PAH levels, the reported high toxicity of methylated PAHs suggests that the presence of methylated PAHs in these biological samples may contribute to the toxicological importance of PAHs.

5.4 Provide expert knowledge and guidance to the ICES Data Centre

Reporting of QA/QC data

The following working document was presented to the group: "Proposal by Germany how to improve the reporting of data on quality assurance and control within the ICES Integrated Environmental Reporting Format" (Annex 5. N.B.: The recommendations in Annex 5 are part of the German proposal and not MCWG's recommendations). The aim was to refresh the discussion about what quality assurance information is necessary and useful to be reported to the ICES database in the light of recent developments.

The proposal suggests that the obligation status of selected information should be amended to reduce the requirements for inputs on internal QA/QC data because this detailed information is specific for the measuring laboratory and could overload the national and international databases unnecessarily. The suggestions are based on the quality assurance and control requirements of EN ISO/IEC 17025 and the Commission Directive 2009/90/EC which include rules on the uncertainty of measurements and on analytical method limits of quantification for WFD monitoring.

In general the group supported the approach, in particular that the reporting of the uncertainty of an analytical measurement is important. Therefore, the provision of an uncertainty value should be mandatory, as well as information on the method for calculation of uncertainty. Information on the laboratory's Quality Management (QM) system and accreditation status, regarding the analytical method in question should also be mandatory.

The QA/QC data in the ICES database are used by OSPAR MON to give analytical weights to monitoring data. One of the QA/QC parameters used for the evaluation of data quality is the participation in (and results of) a proficiency testing scheme. No change of obligation status had been suggested for this parameter, and the group confirmed that this information was necessary ("mandatory") to be reported in the database. It was also discussed that the concentration ranges and matrix of the particular proficiency test were not always representative of the samples monitored.

Another QA/QC parameter used for data evaluation by OSPAR MON is the deviation from concentrations in certified reference materials (CRM). MCWG also discussed this parameter broadly and pointed out some limitations as the available CRMs might not be entirely suitable for the monitoring task, e.g. the samples matrices and concentrations might differ from those of the monitoring programme. Therefore, deviations from the certified values do not necessarily reflect problems with the analytical procedures used for monitoring samples.

MCWG concluded that the "Reference material mean value found" could be reported optionally, as suggested by the German proposal, if this is acceptable for the OSPAR MON work. However, the group did not entirely agree with the line of argument in the German proposal according to which reference values do not need to be reported as the analysis of reference materials is part of the internal QA under a QM system. Whereas it is correct that only validated data are reported as part of the QM system, the assessment work might need a quantitative parameter to evaluate the data quality. It was generally agreed, however, that the measurement uncertainty (with additional information on the method) and results from proficiency testing would provide equally valid or even better information.

The information on the use of an internal reference material where no appropriate CRM is available will need no additional changes to the database if LRM is used for an internal reference material (IRM) from the existing possibilities of type (CRM, LRM, and SRM).

Limits of quantification (LOQ) were discussed as well, but no suggestions were made to change their current reporting status. It was recommended, however, to report the method of determination of LOQ as different methods are currently used, e.g. based on the presence or absence of matrix.

Recommendations

MCWG supports the German proposal, provided that the changes do not conflict with OSPAR assessment approaches. As a minor detail, MCWG suggests changing the reporting requirements to "mandatory" regarding the information of the laboratory being accredited for the particular method or parameter. Other recommended changes are:

- Information on QM system: Mandatory

- Reporting of "Uncertainty value" and "Method of calculating uncertainty": Mandatory.

- Reporting of "Reference material mean value found": Optional, subject to OSPAR acceptance.

Further changes could be considered, e.g. information on method for LOQ determination.

Storage of data on trace metal concentrations in seawater

The ICES Data Centre has accepted data on concentrations of trace metals and organic contaminants in water for many years based on the specifications of Working Groups and Commission clients from the 1980s. The participation of the data centre in the development of a European Marine Observation and Data Network (EMOD-NET) required a mapping to data from other organizations and triggered a review of the database and the current reporting format ERF3.2. ICES asked MCWG to advice on this matter; first for trace metals, later for other parameters (Annex 6).

Comment and recommendations given by Kees Booij and Gert Asmund prior to the meeting were discussed at MCWG 2010. The general issue of metal speciation in seawater was described in Kees Booij's paper and will not be repeated here. MCWG discussed that two different approaches were possible for distinguishing between the so-called dissolved and particulate metals in a general sense: (i) operationally based, as used in the ICES database and (ii) matrix based. The following terms have been used in the database and in Kees Booij's document:

OPERATION–BASED		METALS ARE	MATRIX–BASED	
before filtration (BF)	Unfiltered water	Dissolved, particulate, colloidal, complexed	Total water	
after filtration (AF)	filtered water	dissolved	water	Filtrate
		particulate	suspended particulate matter (SPM)	Residue

MCWG discussed that the matrix based terminology was not really an improvement, because the real condition of the matrix is also determined by the operations performed to separate both phases. So matrix–based terms

- do not necessarily reflect reality
- are a more "academic" approach, but not practical
- need also an operational definition, for comparable data.

Even for those filtration procedures defined by the filtration method, filter type and pore size, the mass spectrum passing the filter (and becoming part of the so-called dissolved fraction) is not known, as a cut-off provides information on a certain percentage of retention for one single particle diameter. The filtration process also depends on the amount filtered, the particle concentration, composition and size distribution, which all can have an effect on the retained material on the filter, e.g. in terms of absorption, chromatographic properties, porosity and flow. As this is a dynamic process the filtration conditions vary and thus the composition of the filtrate and the dissolved phase. Therefore, an exact description of the matrices will not be possible. This applies also to the distribution of colloids between the filtrate and the residue, which will be influenced by any separation procedure.

It was also noted in the discussion that the direct analysis of unfiltered water compared to the addition of the results from filtered water and suspended matter would lead to different results, in most cases. Therefore the metal concentration in 'total water' is not synonymous with the total metal concentration.

MCWG concluded that an operation-based approach was the preferred one. In case of matrix-based terminology, the matrices should be limited to water and particulate matter. Colloidal adsorbed metals cannot be determined with the available analytical techniques of most monitoring laboratories, has no real meaning for monitoring and, therefore, should not be taken into account. Ultrafiltration (also called cross-flow or tangential flow) can separate according to molecular weight cut-offs (Annex 7), but with restrictions comparable to normal filtration. In any case, no matter if operation- or matrix-based terminology is used, method information should be given, in particular on filtration method, filter material and porosity (see questions 1 - 3 in Annex 6).

Regarding question 4 in Annex 6, suspended particulate matter (SPM) should be reported in units of mass/volume, e.g. µg/l or ng/l. Consequently, it is correct to expect the basis "dry weight" for SPM data. The concentration of SPM itself must not be confused with the concentration of e.g. trace metals on SPM. For this parameter, both mass/volume and mass/mass units are meaningful, normalizing the metal concentration to the water volume or the SPM mass of the sample. If the SPM concentration of the sample is known (in mass/volume) the metal concentration can be transferred between the volume and mass basis.

MCWG was requested to review the list of pretreatment methods and make additions as needed (Annex 6). As several pretreatment methods did not apply to seawater, MCWG sorted the methods according to their application in water, sediment and biota analysis. The result is shown in Annex 7. Furthermore, the group recommends removing certain codes of pretreatment, in particular for different filter manufacturers, and structuring information on filtration methods differently, with focus on the type of filtration, the filtration material and the porosity of the filter. Details are given in Annex 7.

The final question in Annex 6 is related to the reporting of metal concentrations in total water. MCWG confirms that the Water Framework Directive (WFD) does not require trace metal analysis in total water as the Directive 2008/105 sets environmental quality standards (EQS) for the dissolved phase (filtrate passing 0.45 μm) for trace metals. As metal concentrations will be determined for filtered water, there is no real issue to address here.

Recommendations

The reporting of trace metal concentrations in seawater should include information on the filtration method, i.e. the type of filtration; the filter material and the porosity (see Annex 7). The list of pretreatment methods in the database can be structured slightly differently, see suggestions in Annex 7.

5.5 Report to SSGHIE on your plans to promote cooperation between EGs covering similar scientific issues (Contaminants – WGMS, WGBEC, SGIMC; Chemical oceanography – SGONS)

The IOC-ICES Study Group on Nutrients Standards (SGONS) is currently developing reference materials for nutrients in seawater (RMNS – see Section 5.6). In addition, new sampling and measurement protocols using the RMNS will be developed by SGONS and a nutrients analysis manual will be produced. MCWG may be required to provide input and advise on the analytical methods and should keep up to date with the work of SGONS. The reference materials produced could be of use to Group members for the routine analysis of nutrients. The next meeting of SGONS is to be held in March 2010 and the outputs of this meeting will be of interest for MCWG and can be followed up at the MCWG meeting in 2011.

Collaboration between MCWG and The Working Group on Eels may also be required. High contaminant concentrations have been found in eels and within the Eel Group it has been suggested that pollution may be a possible cause of the decline of eel populations in some areas. However, there could be other reasons for this decline as well. MCWG may be able to provide advice to the Eel Group on analytical techniques for contaminants, and on quality assurance. Furthermore, there is expertise within MCWG on the analysis of contaminants in eel. MCWG can collaborate with the Eel Group and provide feedback on their studies.

There are strong links between MCWG and the Working Group on Marine Sediments (WGMS). The two groups have collaborated on the production of a number of technical annexes and ICES TIMES papers for the analysis of organic contaminants, and on the guidelines for passive sampling.

There has also been collaboration between MCWG and WGBEC in relation to dioxin analysis and bioassays for analysis. WGBEC provided draft documents on bioassay analysis as background documents prior to the 2010 MCWG meeting.

5.6 Report on developments in relation to ongoing chemical oceanography issues, including the International Nutrient Scale System and SCOR/IAPSO progress on developing algorithms for reporting absolute salinity of seawater

In plenary David Hydes presented information on an initiative to prepare and make available reference materials (solutions) for nutrient analysis in seawater to the worldwide community of analysts. This initiative has been lead by Michio Aoyama of

the Meteorological Research Institute and Hidekazu Ota of the General Environmental Technos Company in Japan. The Technos Company have sponsored the construction of a "clean" building for the preparations of these materials. The results of internal tests of these materials and of two international comparison exercises to which more than 50 laboratories contributed were reported on at the 2009 INSS International Workshop in Paris in February 2009. The development of the processes used to produce these materials owes much to the experiences of ICES-MCWG who initiated a series of intercomparisons led by Alain Aminot. Details were presented by David Hydes that demonstrate that the Technos facility in Japan is able to produce a high quality reference material for nutrient analysis on an industrial scale – batch sizes of up to 2000 bottles. This is something that previous attempts to produce nutrient standards have not been able to achieve.

This initiative was also discussed by the COSG. The COSG strongly supports the production of these reference materials for nutrient analysis and such materials should be used in routine analysis in all laboratories involved in monitoring concentrations of nutrients. COSG is sure that this would improve the comparability and reliability of nutrient data within the OSPAR and HELCOM community, when used in conjunction with participation in QUASIMEME. It fully supports the activities of the ICES-IOC Study Group on Nutrient Standards (SGONS - http://www.ices.dk/workinggroups/ViewWorkingGroup.aspx?ID=417).

An important feature of the materials is that they are produced by the sterilisation of a large volume of seawater collected from pertinent water masses in the ocean. The materials that are available at the moment have been collected in the Pacific and therefore have higher concentrations and nutrient ratios that are not appropriate to work in the North East Atlantic and on the North West European shelf. Also they have salinities which are too high to be appropriate to work with samples collected in the Baltic. Consequently members of COSG expressed their support for the idea working with Aoyama and Ota to supply them with seawaters that could be processed to provide materials appropriate to work in the OSPAR and HELCOM regions.

The IOC-ICES SGONS is holding an open meeting in Paris 23/24 March 2010 and it should be discussed there how materials appropriate to use in the OSPASR-HELCOM areas can best be produced. The COSG considered that the development of these processes should be presented and discussed during the next MCWG meeting.

Klaus Nagel informed MCWG about developments concerning the measurement and reporting of absolute salinity.

Salinity is one of the basic parameters in marine sciences. However, during the last 200 years there have been several changes in the definition of what is called 'Salinity'. The main reason for these different definitions is that a precise direct experimental determination of Salinity is practically impossible. Measurement of salinity was therefore related to other parameters which can be determined with high precision, e.g. chlorinity or conductivity. To achieve this relation between salinity and chlorinity or conductivity, different standards have been used over recent decades, resulting in slightly different definitions and 'salinity scales' and in confusing dimensions of salinity values. The currently most widely accepted measure of salinity is the Practical Salinity S (without dimension).

The major disadvantages of these different definitions of salinity are some inconsistencies in the thermodynamic description of seawater properties. Furthermore variations in composition of local seawater samples contribute to errors, for example if

density is calculated using salinity values. To overcome these disadvantages SCOR/IAPSO Working Group 127 on Thermodynamics and Equation of State of Seawater defined Reference Salinity S_R, which accurately represents the Absolute Salinity S_A of artificial seawater that has stoichiometry in molar fractions of the IAPSO Standard Seawater constituents. S_R is intended to be used as the new independent salinity variable for future thermodynamic functions of seawater, as an SI-based extension of Practical Salinity, as a reference for natural seawater composition anomalies and as the currently best estimate for Absolute Salinity of IAPSO Standard Seawater.

An approximate relation, which is suitable for most practical applications, is established that connects Absolute Salinity to currently used Practical Salinity. Details of the definitions of Reference Salinity and information for relating it to Practical Salinity are compiled in http://www.teos-10.org/ .

The dimension of salinity according to the new definition of salinity is $g*kg^{-1}$ and it is expected that it will be introduced in the near future as the official measure of salinity.

Recommendations

ICES to note this new definition of salinity and possible consequences for the database.

5.7 Review available information regarding the role and the elemental composition of organically bound nutrient species in relation to recycling of inorganic nutrients

COSG discussed whether or not this point should remain on the agenda as no new information was available. The conclusion was to take it off the agenda for the next meeting.

5.8 **Monitoring methodologies for ocean acidification (OSPAR request 2010/2) To provide, on the basis of a review of existing methodologies and experience, recommendations for cost efficient methods for monitoring ocean acidification (OA) and its impacts, including possibilities for integrated chemical and biological monitoring. Specifically this should provide:**

I. **advice on appropriate parameters, protocols and quality assurance for monitoring changes in pH and inorganic carbon chemistry in the OSPAR maritime area and other ancillary parameters that should be included in monitoring programmes**

II. **advice on the status of current knowledge of spatial and temporal variability of pH and inorganic carbon chemistry in the OSPAR maritime area**

III. **advice on appropriate spatial and temporal coverage for monitoring, considering different oceanographic features and conditions and key habitats/ecosystems at risk from OA in the OSPAR maritime area**

The COSG addressed the OSPAR request and the following text outlines the key conclusions and recommendations of MCWG. More detailed supporting information is presented in Annex 8. This includes information on the status of current knowledge of spatial and temporal trends as required under 5.h.ii of the agenda. The Chair addressed limited specialist expertise in MCWG by inviting additional experts in this area to the meeting and circulating the draft text to further experts subsequent to the meeting to invite their comments.

General Comments

Increasing concentrations of CO_2 in the atmosphere will have long-term impact on the carbonate system in marine environments. It is estimated that oceans absorb approximately a quarter [1,2] of the total anthropogenic releases of carbon dioxide each year. Ocean acidification is a persistent process. It is a direct consequence of this uptake and has already been observed through direct measurements [3]. These changes in the carbonate system are a cause for concern for the future health of marine ecosystems.

Painstaking and sensitive methods are necessary to measure changes against a background of high natural variability. It is a long-term process requiring long-term observations. A monitoring programme needs to be designed to distinguish long-term trends from shorter term natural variability. Monitoring needs to start with a research phase which assesses the scale of short term variability of different regions. The Arctic Ocean needs to be monitored because its waters are potentially most sensitive to change. The Atlantic Ocean needs to be monitored because it provides the source waters to the shelf seas and is already known to show more variability than is predicted by numerical models [3]. In near shore environments increased production resulting from eutrophication has probably driven larger changes in acidity than CO_2 uptake [5]. Although the cause is different, data are equally required from these regions to assess potential ecosystem impacts.

MCWG considered the scientific background to the request and the overall objectives of OSPAR monitoring. It was considered that the key broad objective for OSPAR was to measure long-term changes in pH, carbonate parameters and saturation states ($\Omega_{ARAGONITE}$ and $\Omega_{CALCITE}$) in support of assessment of risks to and impacts on marine ecosystems in the OSPAR regions. Monitoring in the OSPAR region should also be strongly linked with proposed global observational networks and international carbon research projects and initiatives [6,7]. One example of such an initiative is the IMBER/SOLAS Ocean Acidification Working Group (http://www.imber.info/C_WG_SubGroup3.html) which has the following tasks:

1) Coordinate international research efforts in ocean acidification;

2) Undertake synthesis activities in ocean acidification at the international level.

MCWG reviewed the present state of knowledge with respect to the points raised in the OSPAR request. MCWG compiled information regarding the measurements already performed or planned in the OSPAR area and the methods and instrumentation available to perform these measurements (covering laboratory instruments as well as those that can work autonomously in ships of opportunity or on moorings). The work covered is presented in the Annex 8 of this report. <u>It is highlighted that sustained funding will be required to mount the programme of sustained observations that is necessary.</u> However much of the sample collections and measurements required can be carried in conjunction with existing work. The overlap of interest with monitoring of the CO_2 exchange process itself and initiatives such as ICOS - Integrated Carbon Observation System (http://www.icos-infrastructure.eu) will reduce the likely cost burden of specifically acidification related monitoring. MCWG made the following recommendations:

Advice on measurements parameters, protocols and quality assurance

A review of methods and instrumentation available to undertake these measurements, covering laboratory instruments as well as those that can work autonomously in ships of opportunity or moorings are presented in Annex 8.

1) Coordinated observation of the carbonate system in the OSPAR areas should be started as soon as possible to establish long-term datasets. Measurements of carbonate system parameters should therefore be included in monitoring programmes, taking into account the requirements for these parameters in the Marine Strategy Framework Directive.

2) There are four parameters which describe the carbonate system (total hydrogen ion concentration, total alkalinity (TA), total dissolved inorganic carbon (DIC) and partial pressure of carbon dioxide pCO_2). A minimum of two must be determined to derive the other two. If three of the variables are determined this allows verification of the computed value. DIC and TA are the most widely measured for discrete samples while pCO_2 is the most common for underway measurement. At present there are practical and technological limitations to pH measurements but because pH is of primary concern this parameter should be measured when possible in future.

3) Both discussion in the meeting and a recent papers [8, 9,10] have raised concerns about reliability of these widely used calculations in different types of natural waters and artificially modified waters used in experimental systems. There is urgent need to resolve these questions (See MCWG 2010 Annex 8 Section 9.2).

4) Necessary parameters to be measured alongside the carbonate parameters are temperature, salinity, dissolved phosphate and silicate as these are required for carbonate system calculations. It is strongly recommended that dissolved oxygen and nitrate also be measured to provide information on the timing of the data relative to the seasonal production/respiration cycle. OSPAR has existing guidelines for the determination of these ancillary parameters.

5) To achieve consistency, the widely accepted procedures outlined in the handbook by Dickson *et al.*, (2007) [11] must be followed, although limitations have to be taken into account in estuarine systems. Monitoring requires a long-term commitment to both observation and methods (technological) development. Present methods are slow and require high skill levels. Improvements in the methods should be sought to improve their ease and speed of use. Monitoring will need to adapt to these changes.

6) To obtain reliable and consistent datasets it is essential that a high level of quality assurance is established from the beginning of the programme. This can be based on the certified reference materials (CRMs) for DIC and TA which are available from Professor Andrew Dickson (Scripps Institution of Oceanography, USA), and reference gases for pCO_2 measurement are available from NOAA. (Certified buffer solutions for use in the measurement of pH are currently underdevelopment). Use of CRMs should be coupled with inclusion of the appropriate determinands in a laboratory proficiency testing schemes (QUASIMEME).

7) Data should be reported to both the Carbon Dioxide Information Analysis Center (CDIAC - http://cdiac.ornl.gov/oceans/home.html) and to the ICES data repository (http://www.ices.dk/datacentre/Submissions/index.aspx). Reporting data to the ICES data repository enables it to be linked to many related OSPAR datasets e.g. nutrients and integrated ecosystem data. Globally most research groups measuring carbonate parameters submit data to CDIAC. CDIAC has established reporting formats for these data and related metadata, and also has worked with the community to develop systems for effective data access and review, such as the SOCAT (Surface Ocean CO_2 Atlas - http://www.ioccp.org/SOCAT.html). ICES should consider how data reporting would evolve so that relevant data are available and accessible to both databases without replicating reporting requirements.

Spatial and temporal variability of pH and inorganic carbon chemistry in the OSPAR maritime area

Information on spatial and temporal variability of pH and inorganic carbon chemistry in the OSPAR maritime area is summarized in the detailed technical document at MCWG 2010 Annex 8 Section 8.

Advice on appropriate spatial and temporal coverage for monitoring

MCWG cannot give precise guidelines on spatial and temporal coverage for monitoring. The attached detailed report at Annex 8 describes the currently available sources of data and suggests known locations where continued observations and new observations should be made (see Sections 3 and 7). The design of a monitoring programme should take the following into consideration.

1) A lack of data on seasonal and interannual variability means that an initial research phase of more widely dispersed and more frequent measurements is necessary to provide the information required to design a statistically robust long-term monitoring programme.

2) Measurements need to cover the range of waters from estuaries, shelf seas and ocean mode waters and abyssal waters where sensitive ecosystems may be present. That is the spectrum of waters in all OSPAR areas. Particular emphasis should be placed on key areas at risk within the OSPAR area, for example high latitudes where ocean acidification will be most rapid, and areas identified as containing ecosystems and habitats which may be particularly vulnerable, e.g. cold water corals.

3) There are several national and international projects that have been/are currently active in the OSPAR area making sustained measurements using a variety of platforms. These can be used as a basis for the development of the future monitoring programme, details are provided in Annex 8 Section 7. Building relevant measurements into other related programmes may also support cost-effective monitoring. For example, in some locations, incorporation of carbonate parameters into OSPAR eutrophication monitoring may provide a cost-effective approach to coastal and inshore monitoring.

4) Monitoring is foreseen as a combination of low-frequency repeat ship-based surveys enabling collection of extended high quality datasets on horizontal and vertical scales, and high-frequency autonomous measurements for more limited parameters using instrumentation deployed on ships of opportunity and moorings. MCWG noted the standard World Ocean Circulation Experiment (WOCE) horizontal sampling strategy for oceanic studies of 30 nautical mile spacing for physical measurements, with higher resolutions in regions of steep topography and boundary currents, and 60 nautical miles or better for carbon and tracer measurements [12].

Advice on future planning and coordination

Development of coordinated and harmonized monitoring of ocean acidification involving OSPAR contracting parties would be best developed through a (finite life) OSPAR/ICES study group to bring together the required expertise. The study group would need to include national representative from the observing teams, experts on (1) the development of appropriate analytical methods (2) carbonate system chemistry (3) ecosystems function potentially affected by acidity changes (4) ecosystem modellers (5) statisticians involved with the efficiency of monitoring programmes. Terms of Reference for the study group would include:

1) Identify available resources (including appropriate national and international projects and monitoring).

2) Identify vulnerable areas requiring particular emphasis for monitoring (e.g. through modelling output and consultation with biologists).

3) Plan coordinated monitoring activities of contracting parties based on existing and any necessary new initiatives.

4) Agree on harmonization of approaches for sample collection, analytical methods, QC, QA and data reporting.

5) Design a statistically robust monitoring programme on the basis of the baseline data and considering related output from numerical models,

6) Address the current state of play with regard to the direct measurement of pH in marine and estuarine systems and promote the development and use of direct methods for measuring pH.

7) Develop suitable "indicators" for the reporting of ocean acidification.

Additional points

MCWG identified several points that need further investigation or research in order to achieve measurements of carbonate system parameters that fulfil the requirements of the OSPAR monitoring system.

1) Direct measurements of pH are the preferred option but there are practical and technical challenges. Additionally, calculation of pH from other parameters has to be done taking into account the limitation of the carbonic acid dissociation constants being used and using an appropriate pH scale (See MCWG 2010 Annex 8 Sections 2.1 and 9.1).

2) Measurements of carbonate system parameters are specialised methods. It should be considered if specialist laboratories with a track record in making these measurements should be contracted to do the analyses (at least in the first stages of this work). Workshops within the monitoring community will probably be necessary for the exchange of expertise and harmonizing of methods applied in regions other than the open ocean.

3) Substantial research is necessary to evaluate variability of the system in time (daily, monthly, annual scale) as well as in space. Such information is necessary to define the appropriate frequency and distribution of future long-term monitoring.

4) A cause for concern is the future supply of CRMs. Presently these are produced on limited scale ("cottage industry") in Andrew Dickson's laboratory. Increased research into ocean acidification has the potential the increase the demand for CRMs beyond the capacity of this laboratory. Andrew Dickson is working with Akihiko Murata (JAMSTEC, Japan) and others to develop an alternative and larger source of supply.

Topics for further discussion at the next ICES MCWG meeting

MCWG 2010 suggests four items for the MCWG 2011 agenda (Annex 3):

1) Report on recent developments in methodology of pH measurements and required standardization procedures

2) Report on recent results of measurements of pH in marine systems (especially in the OSPAR area)

3) Report on recent developments in quality assurance issues in the measurement of carbonate system parameters.

4) Review new information for determination of pH and carbonate parameters in estuarine and brackish water and the associated calculations.

Furthermore, COSG felt that the information presented in Annex 8 would have a broad interest given the increasing interest in this field and could be published as an ICES report. The MCWG Chair undertook to explore this possibility with the ICES secretariat.

Recommendations

- OSPAR to note the advice of MCWG in support of coordinated monitoring of ocean acidification in the OSPAR maritime area.

- A limited life ICES/OSPAR study group should be established to further develop coordinated monitoring of ocean acidification in the Northeast Atlantic.

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5.9   Guidelines for Monitoring PCDD/Fs and "dioxin like" PCBs

I.   Finalize technical annex for monitoring PCDD/Fs and "dioxin like" PCBs in sediments (with WGMS); (OSPAR 2008/4)

MCWG compiled, in collaboration with the Chair of WGMS, a new draft based on the 2009 draft technical annex of WGMS and the technical annex for monitoring of PCDD/Fs and "dioxin like" PCBs in biota. The new draft was presented at WGMS by the Chair for review and editing by the group. Having received comments from WGMS, MCWG finalized the technical annex shortly after the meeting (Annex 10).

MCWG suggests combining, in collaboration with WGMS, both technical annexes (i.e. biota and sediment) in the form of an ICES Times Series paper. The Chair of WGMS will take this proposal to his group for their approval.

II.   Respond to comments received by ICES on consideration of cell based assays in biota monitoring guidelines for dioxin

At MCWG2009, at technical annex was prepared for the monitoring of PCDD/Fs and dioxin-like PCBs in biota. ICES was contacted subsequently by Dr Peter Behnisch, Commerce and Marketing Director of BioDetection Systems bv who commented on the presentation of bioassays for the determination of these compound groups. It was agreed by ICES and the MCWG Chair that these comments should be addressed at MCWG2010, to be discussed by the experts who were involved in the preparation of this technical annex.

The technical annex cited COMMISSION DIRECTIVE 2002/69/EC describing a two-step approach for the determination of these compound groups in foodstuffs: An initial screening and a subsequent verification of positively tested samples. It then continues "This approach has not been applied in the monitoring of dioxins/furans under the OSPAR Coordinated Environmental Monitoring Programme (CEMP), but will be briefly described in this guideline (see "Screening methods based on bioassays"). Primarily, it is applied for the screening of a large number of samples, as the method, based on cell lines, is relatively costly to maintain or to use for lower sample numbers. In environmental monitoring, it might for instance be useful for the identification of suitable sampling locations."

Peter Behnisch's comment was related to the description of the bioassay as "costly to maintain" which he considered as incorrect. This was discussed by MCWG2010 with the conclusion that the maintenance of cell lines and the application of a bioassay to a small number of samples can be costly and that the sentence itself was correct. MCWG also recognized, however, that their recommendations of certain methods should primarily be based on their technical and scientific merit and that costs should be a minor criterion.

Focussing on the technical and scientific aspects, it was confirmed, as also concluded in the technical annex, that bioassays were not suitable for the main purpose of OSPAR CEMP, which is the quantitative determination of individual compounds and congeners over time, specifically in areas already identified as contaminated. For this purpose, the bioassay lacks specificity. As mentioned before, MCWG saw opportunities for bioassay applications as screening tools, for instance in the identification of problem areas, and in integrated monitoring, in terms of risk identification.

MCWG agreed that the technical annex should be revised by deleting the whole paragraph in question, to focus the guideline on technical and scientific matters. Some discussion on applicability of bioassays remains under "7. Screening methods based on bioassays" (Annex 9).

Recommendations

OSPAR to note that the guideline has been revised (Annex 9).

5.10 Report on new information regarding emerging contaminants in the marine environment

Four presentations were given under this agenda point, i.e.

- Lutz Ahrens: Partitioning behaviour of polyfluoroalkyl compounds (PFCs) in the marine environment;
- Ralf Ebinghaus: PFCs in the marine environment;
- Norbert Theobald: Emerging contaminants in the North and Baltic Seas;
- Katrin Vorkamp: Temporal trend of hexabromocyclododecane (HBCD) in ringed seal from East Greenland.

5.10.1 Lutz Ahrens: Partitioning Behaviour of Polyfluoroalkyl Compounds (PFCs) in the Marine Environment

New results were presented of a study examined the partitioning behaviour of polyfluoroalkyl compounds (PFCs) in water, suspended particulate matter (SPM) and sediment. Sediment cores and seawater samples from three water layers (surface, middle and bottom) were collected at two sampling stations in Tokyo Bay, Japan, in 2008. Furthermore, the influence of physico-chemical parameters, such as organic carbon, was examined on the sorption of PFCs onto the sediment and SPM. Finally, the partition coefficient (K_d) and organic carbon normalized partition coefficient (K_{OC}) was calculated for the sorption on sediment and SPM (Ahrens *et al.*, 2009; Ahrens *et al.*, in press).

In the sediment cores the short-chain perfluoroalkyl carboxylic acids (PFCAs; C ≤ 7) were found exclusively in pore water, while long-chain PFCAs (C≥ 11) were found only in sediment. The perfluoroalkyl sulfonates (PFSAs), n-ethylperfluoro-1-octanesulfonamidoacetic acid (N-EtFOSAA) and perfluorooctane sulfonamide (PFOSA) seemed to bind more strongly to sediment than PFCAs. The enrichment of PFCs on sediment increased with increasing organic matter and decreasing pH. The perfluorocarbon chain length and functional group were identified as the dominating parameters that had an influence on the partitioning behaviour of the PFCs in sediment. The maximum ΣPFC contamination in sediment was observed in 2001–2002 to be a flux of 197 pg/cm²/yr. Concentrations of PFOSA and N-EtFOSAA increased between 1985 and 2001, but after 2001, the concentration decreased significantly, which corresponded to the phase out of perfluorooctyl sulfonyl fluoride-based compounds by the 3M Company in 2000.

References

Ahrens, L., Yamashita, N., Yeung, L. W. Y., Taniyasu, S., Horii, Y., Lam, P. K. S., and Ebinghaus, R. 2009. Partitioning behaviour of per- and polyfluoroalkyl compounds between pore water and sediment in two sediment cores from Tokyo Bay, Japan. Environ. Sci. Technol., 43: 6969–6975.

Ahrens, L., Taniyasu, S., Yeung, L. W. Y., Yamashita, N., Lam, P. K. S., and Ebinghaus, R. 2010. Distribution of polyfluoroalkyl compounds in water, suspended particulate matter and sediment from Tokyo Bay, Japan. Chemosphere, in press.

5.10.2 Ralf Ebinghaus: PFCs in the marine atmosphere.

Ralf Ebinghaus presented PFC concentration data from ship-borne sampling campaigns in the marine atmosphere. These measurements mainly focused on volatile precursor compounds; however, ionic PFCs in the particulate phase were also included in these studies. Target analytes were Fluortelomeralcoholes (FTOH), Fluortelomeracrylates (FTA), Perfluorsulfonamides (FASA), Perfluorsulfonamidoethanoles (FASE), Perfluorcarboxylic acids (PFCA) and Polyfluorsulfonic acids (PFSA).

Ship cruises can be summarized as follows:

North – South – Transects

Longyearbyen (Norway) – Kiel (Germany)	Maria S. Merian 2007
Bremerhaven (Germany) – Cape Town (South Africa)	Polarstern 2007, 2008
Cape Town (South Africa) – Neumayer (Antarctica)	Polarstern 2008

East – West – Transects

Las Palmas (Spain) – St John's (Canada)	Maria S. Merian 2007
Recife (Brazil) – Dakar (Senegal)	L'Atalante 2008

Regional

German Bight (North Sea)	Atair 2007
Baltic Sea	Maria S. Merian 2008

The results for PFCs detected in the **gas-phase** can be summarized as follows:

Only neutral volatile PFC (precursors) were determined in gas-phase samples and the total gas-phase PFC concentrations ranged from 4.5 pg m^{-3} (Southern Ocean) to 335 pg m^{-3} (source regions, here Gulf of Bothnia)

The detected concentrations were always FTOH > FASA > FASE > FTA with a relative contribution to the total concentration of 76%; 11%; 8%; and 5% respectively.

During all measurement campaigns 8:2 FTOH was always observed in the highest concentrations.

The results for PFCs detected in the **particulate phase** can be summarized as follows:

Both neutral volatile precursors and persistent ionic PFC were determined in particle-phase samples. Volatile precursors such as FASA and FASE were detected at low concentrations (< 1 pg m^{-3}) in the particulate phase, but FTOH and FTA were not detected above the blank

Persistent ionic PFC such as C4-C13 PFCA and PFSA were detected at low concentrations, namely 1 pg m^{-3} and 0.3 pg m^{-3} respectively.

Maximum concentrations in the particulate phase in potential source regions were reached for PFOA with 6 pg m^{-3} and for PFOS with 2 pg m^{-3} and the overall contribution of these substance groups in the particle phase was higher for PFCA (90%) than for PFSA (10%).

In general, the particle-phase contribution to the total atmospheric PFC concentration was consistently < 20%.

More details results of these studies can be found in the following references:

Dreyer, A., Ebinghaus, R. 2009. Polyfluorinated Compounds in Ambient Air from ship- and land-based measurements in northern Germany, Atmospheric Environment, 43 (8), 1527–1535.

Dreyer, A., Weinberg, I., Temme, C., Ebinghaus, R. 2009a. Polyfluorinated compounds in the atmosphereof the Atlantic and Southern Oceans: Evidence for a global distribution, Environmental Science & Technology, 43 (17): 6507–651.

Dreyer A., Matthias V., Temme C., Ebinghaus, R. 2009b. Annual time-series of air concentrations of polyfluorinated compounds (PFC). Environmental Science & Technology. 43 (11): 4029–4036.

5.10.3 Norbert Theobald: Emerging pollutants in the North and Baltic Seas.

In the presentation some new results on the investigation of mid-polar emerging pollutants were shown. Sea water samples of the North and Baltic Seas had been extracted by solid phase extraction and analysed with HPLC-MS/MS for selected pharmaceuticals, phosphorous ester flame retardants and the high volume chemical benzotriazole which is widely used as a corrosion inhibitor. The results show that many new compounds exhibit significantly higher concentrations in the water phase than "classical" pollutants (PAHs, chlorinated hydrocarbons). In particular, highest concentrations were found in coastal areas of the southern North Sea. By correlating the concentrations to salinity it can be shown that the rivers Rhine and Elbe are the main sources for the pollutants. In addition, the linearity of the correlation shows that these emerging pollutants behave conservatively and are stable in the marine environment. Highest concentrations were observed for Carbamazepine, Tris(1,3-dichlorisopropyl)phosphate and benzotriazole. For these compounds, concentrations of up to 20 ng/L were observed in coastal waters and 100 to 300 ng/L were found in the river Elbe. Clear gradients with decreasing concentrations were observed towards the North Sea. In the Baltic Sea concentrations were lower (1 to 5 ng/L) and the distribution was more homogeneous with only low gradients. A similar pattern of occurrence was shown for PFCs.

5.10.4 Katrin Vorkamp: Temporal trend of hexabromocyclododecane (HBCD) in ringed seal from East Greenland.

As part of the Arctic Monitoring and Assessment Programme (AMAP), the Danish National Environmental Research Institute (NERI) has monitored the pollution of the Greenland environment for many years. In the AMAP Core Project, key species have been identified which contain contaminants in high concentrations, e.g. as a result of bioaccumulation and biomagnification, and/or are important food items in the traditional Arctic diet. Samples of these species are collected in collaboration with local hunters with a two-year-interval and analysed for persistent organic pollutants (POPs) and heavy metals. In addition, the AMAP Core Project has performed retrospective time-trend studies on archived samples, including polybrominated diphenyl ethers (PBDEs) in ringed seals from East and West Greenland (Riget *et al.*, 2006; Vorkamp *et al.*, 2008) and polyflourinated compounds (PFCs) in ringed seals and polar bears (Bossi *et al.*, 2005; Dietz *et al.*, 2008). As a consequence of increasing trends established in the retrospective studies, these compounds have subsequently been transferred to the regular POP-monitoring.

Recently, a retrospective study has been conducted on the high volume brominated flame retardant hexabromocyclododecane (HBCD), analysing 50 samples of ringed seals from East Greenland collected in ten years between 1986 and 2008. The use of HBCD increased by 40% between 1999 and 2003 and, in contrast to the PBDE application pattern, almost equal amounts has been used in Europe and North America. Technical HBCD consists of approximately 80% γ-HBCD, while α- and β-HBCD account for approximately 12% and 6%, respectively. In biota samples, this profile has been found to be reversed, with α-HBCD being the dominating congeners. This was also the case in the ringed seal samples which did not contain any detectable concentrations of β- and γ-HBCD.

A statistically significant exponential time-trend was found with an annual increase of +6.1%. The median concentration of α-HBCD was 8.7 ng/g lw in 2008. Interestingly, no significant time-trend had previously been found for PBDEs in the same animals and most recent data indicate a downward trend for PBDEs in East Greenland. For PBDEs in ringed seals from West Greenland, however, a statistically significant linear increase of 5.3% had been found (Vorkamp *et al.*, 2008). Time trend studies on HBCD in the Arctic have recently been summarized, but included increases, decreases and inconclusive developments (de Wit *et al.*, in press).

The HBCD-concentrations of this study were comparable with previous results for ringed seals from East Greenland, although determined differently (i.e. GC-MS in the former study and LC-MS-MS in this study; Frederiksen *et al.*, 2007). The former GC-MS results showed lower concentration in West Greenland than in East Greenland, by about a factor of 4. At the same time, HBCD concentrations in ringed seals from Svalbard exceeded the East Greenland results by about a factor of 3, while concentrations were below detection limits in ringed seals from the Canadian Arctic (de Wit *et al.*, in press). These geographical differences could indicate the same spatial pattern that has been established for PBDEs and other POPs, however, the HBCD data are very limited.

References

Bossi, R., Rigét, F.F., Dietz, R. 2005. Temporal and spatial trends of perfluorinated compounds in ringed seal (*Phoca hispida*) from Greenland. Environ. Sci. Technol., 39 (19), 7416–7422.

de Wit, C., Herzke, D., Vorkamp, K. (in press). Brominated flame retardants in the Arctic – trends and new candidates. Sci. Total Environ.

Dietz, R., Bossi, R.; Rigét, F.F., Sonne, C., Born, E.W. 2008. Increasing perfluoroalkyl contaminants in East Greenland polar bears *(Ursus maritimus)*: a new toxic threat to the Arctic bears. Environ. Sci. Technol. 42 (7), 2701–2707.

Frederiksen, M., Vorkamp, K., Bossi, R., Riget, F., Dam, M., Svensmark, B. 2007. Method development and preliminary screening of HBCD, TBBPA and dimethyl-TBBPA in marine biota from Greenland and the Faroe Islands. Int. J. Environ. Anal. Chem. 87 (15), 1095–1109.

Riget, F., Vorkamp, K., Dietz, R., Rastogi, S.C. 2006. Temporal trend studies on polybrominated diphenyl ethers (PBDEs) and polychlorinated biphenyls (PCBs) in ringed seals from East Greenland. J. Environ. Monitor, 8: 1000–1005.

Vorkamp, K., Rigét, F.F., Glasius, M., Muir, D.C.G., Dietz, R. 2008. Levels and trends of persistent organic pollutants in ringed seals *(Phoca hispida)* from Central West Greenland, with particular focus on polybrominated diphenyl ethers (PBDEs). Environ. Int., 34(4): 499–508.

5.11 MCWG to contribute, as may be required, to ICES activities on integrated chemical and biological effects monitoring and review new information on effect directed chemical analysis

No new information was presented under this agenda point. It was noted that SGIMC had met in January 2010 but the report was not as yet available. There was no specific request from ICES to MCWG 2010 in relation to this agenda point. It was agreed that this topic should remain on the 2011 agenda given MCWG's interest in the topic. A number of presentations were suggested for MCWG 2011 (see 8. Action list).

5.12 Complete guidelines for publication in ICES TIMES series: Determination of PFCs in marine sediment, biota and seawater; Guidelines on use of silicone rubber passive samplers

Determination of PFCs in marine sediment, biota and seawater

A draft paper was prepared by Lutz Ahrens prior to the working group meeting. The draft document was based on the technical annexes for monitoring PFOS in sediments and water produced by MCWG and WGMS in 2009, with additional information on biota. A small subgroup consisting of Lutz Ahrens, Philippe Bersuder, Stepan Boitsov, Ralf Ebinghaus and Norbert Theobald reviewed the draft document. The reviewed paper is to be sent to two external experts for a final review, with the aim of a submission to ICES for publication before the end of March 2010.

Silicone rubber passive samplers

A small subgroup consisting of Lynda Webster, Katrin Vorkamp and Stepan Boitsov reviewed the guidelines for passive sampling of hydrophobic contaminants in water using silicone strip samplers prepared at MCWG2009. A videoconference with Foppe Smedes was organized during the meeting to discuss editorial changes and possible new developments to include into the guidelines. Comments made by the subgroup will be sent to Foppe Smedes who will include new information and transfer the document into the ICES TIMES series format so as to submit for publication before 15 April 2010.

5.13 Provide advice on whether it is appropriate to include PFOS in atmospheric monitoring programmes and if other perfluorinated compounds should be included in such monitoring to support assessments of inputs of PFOS to the marine environment (OSPAR request 2010/6)

Perfluorooctane sulfonate (PFOS) can be found ubiquitously in the remote marine environment and even in polar ecosystems (Giesy and Kannan, 2001). However, it is still under debate how PFOS reaches those regions. At present, no scientific consensus exists whether the atmospheric transport by volatile precursor compounds (Ellis *et al.*, 2004; Martin *et al.*, 2006; Schenker *et al.*, 2008) or the transport by oceanic currents or by means of sea-spray is the dominant transport pathway (Armitage *et al.*, 2006; McMurdo *et al.*, 2008).

It is generally accepted that 8:2 fluorotelomer alcohol (FTOH), a potential perfluorooctanate (PFOA) precursor, is by far the dominating compound in the remote atmosphere. 8:2 FTOH is considered to occur exclusively in the gaseous phase (Dreyer *et al.*, 2009a; Dreyer and Ebinghaus, 2009).

Potential PFOS precursors, such as perfluorooctanesulfonamides (FASA) and perfluorooctanesulfonamidoethanols (FOSE) have been detected both in the gaseous and in the particulate phase at very low concentrations (Dreyer *et al.*, 2009a; Dreyer *et al.*, 2009b).

To our knowledge, a link between atmospheric transport of precursor compounds and occurrence and accumulation of PFCs (i.e. mainly PFOS) in biota has not been scientifically established yet.

Direct atmospheric measurements of PFOS are very limited. Only a few studies could detect PFOS and other ionic perfluorinated compounds (PFCs) in the particulate phase (Barber *et al.*, 2007; Jahnke *et al.*, 2007; Dreyer *et al.*, 2009a,b; Dreyer and Ebinghaus, 2009). However, the concentration levels were close to the method detection limits and the analytical capabilities to analyse PFOS in air samples were insufficient.

Recommendations

Further method development (including sampling and sample preparation) is necessary before MCWG is in the position to provide advice to include PFOS or other ionic PFCs, including PFOS precursors, in atmospheric monitoring programmes.

To support assessments of inputs of PFOS to the marine environment, further research on the wet deposition of PFCs and the partitioning of ionic PFCs between the particulate and gaseous phase would be needed for an improved understanding of the importance of atmospheric transport and deposition.

Current research programs should be used to fill the gaps explained above, e.g. in terms of existing infrastructure from monitoring programmes.

MCWG will continue to review the scientific literature on the role of atmospheric transport and deposition for the assessment of inputs of PFOS to the marine environment (see 8. Action list).

References

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McMurdo, C. J., Ellis, D. A., Webster, E., Butler, J., Christensen, R. D., and Reid, L. K. 2008. Aerosol enrichment of the surfactant PFO and mediation of the water-air transport of gaseous PFOA. Environ. Sci. Technol., 42: 3969–3974.

Schenker, U., Scheringer, M., MacLeod, M., Martin, J. W., Cousins, I. T., and Hungerbühler, K. 2008. Contribution of volatile precursor substances to the flux of perfluorooctanoate to the Arctic. Environ. Sci. Technol., 42: 3710–3716.

6   Plenary discussion of draft report

The plenary discussion of the draft report took place on Friday 5 March 2010. The final draft version of the report was circulated by e-mail after the meeting, for approval by MCWG.

7   Any other business

Evin McGovern announced that he would step down as Chair after MCWG 2010. The group thanked him for his excellent work in these last four years.

Katrin Vorkamp is going to continue as MCWG Chair, but asked for assistance with chemical oceanography matters. Elisabeth Sahlsten was appointed as informal sub-group chair.

8   Recommendations and action list

Recommendations are listed in Annex 4.

The following actions have arisen from MCWG 2010:

ACTION	WHO
Report on new results on PFCs in the atmosphere and discuss implications for the marine environment.	Lutz Ahrens
Present new information on organophosphorous and new brominated flame retardants in the marine environment	Philippe Bersuder
Update MCWG on new results on PFCs, including relevant information for marine and/or atmospheric monitoring	Ralf Ebinghaus
Report on contaminant concentrations and biological effects in eel	Michiel Kotterman
eport on the effect of multicontaminant exposure on early life stages of sole	Michiel Kotterman
Present draft technical annex for dioxins/furans and dioxin-like PCBs in sediment to WGMS for finalization and approval. Report back to the Chair of MCWG during WGMS meeting.	Patrick Roose

Action	Who
Bring relevant environmental monitoring data for tests of statistical approaches of compliance checking under WFD	Patrick Roose
Bring MCWG comments on draft CMA document to the attention of the CMA group	Patrick Roose
Follow up on the INRAM project as presented by Colin Janssen at MCWG 2010 (see 4.1)	Patrick Roose
Prepare a list of certified reference materials available for nutrient analyses in seawater	Elisabeth Sahlsten
Report to MCWG 2011 on pH monitoring activities in the Baltic sea	Elisabeth Sahlsten, Klaus Nagel
Present new information on emerging compounds, including time-trend data on PFCs.	Norbert Theobald
Prepare draft TIMES series paper on the analysis of dioxins/furans and dioxin-like PCBs in biota and sediment	Katrin Vorkamp, Patrick Roose
Prepare draft TIMES series paper on the analysis of PCBs in biota and sediment. First step is to get in touch with original authors and discuss this plan with them.	Lynda Webster

9 Date and venue of the next meeting

MCWG received and welcomed an invitation from Elisabeth Sahlsten to host MCWG 2011 in Gothenburg, Sweden. Dates to be confirmed.

10 Closure of the meeting

The meeting was closed at 1 p.m. on Friday, 5 March 2010.

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Annex 2: Agenda

ICES Marine Chemistry Working Group: 32nd meeting

University of Ghent, Belgium 1–5 March, 2010

DRAFT AGENDA

1   OPENING OF THE MEETING

The meeting will begin at 10.00 am on the first day, and 09.00 am thereafter.
ADOPTION OF THE AGENDA

2   REPORT OF THE 97th ICES STATUTORY MEETING

3   PLENARY PRESENTATIONS

3.a Colin Janssen (University of Ghent): *Qua vadis? Linking biology and chemistry in marine monitoring*

3.b David Hydes (NOC Southampton): *Portsmouth-Bilbao FerryBox - Inter-annual and seasonal dynamics in nutrients and oxygen*

3.c Adrian Covaci (University of Antwerp): *Anthropogenic and naturally produced brominated compounds in the marine environment*

4   MAIN AGENDA

4.a Report on developments with regard to quality assurance of marine chemistry, in particular with respect to QUASIMEME.

4.b Report on the developments in Water Framework Directive monitoring programmes for physico-chemical parameters (priority substances, other pollutants, nutrient status) in transitional and coastal waters; and developments in defining MSFD GES descriptors (Task groups 5 - eutrophication, TG8 - contaminants and their effects in the marine environment, TG 9 - contaminants in seafood).

4.c MCWG members to report information on projects of relevance to MCWG activities.

Stepan Boitsov: *Hydrocarbons in marine sediments of the Barents Sea and the North-Eastern part of the Norwegian Sea.*

Michiel Kotterman: *Identification and quantification of methylated PAHs by GCxGC-MS.*

4.d Provide expert knowledge and guidance to the ICES Data Centre (via sub-group) on a continuous basis.

4.e Report to SSGHIE on your plans to promote cooperation between EGs covering similar scientific issues. (contaminants - WGMS, WGBEC, SGIMC; Chemical oceanography - SGONS)

Chemical Oceanography

4.f Report on developments in relation to ongoing chemical oceanography issues, including the International Nutrient Scale System and SCOR/IAPSO progress on developing algorithms for reporting absolute salinity of seawater;

4.g Review available information regarding the role and the elemental composition of organically bound nutrient species in relation to recycling of inorganic nutrients;

4.h Monitoring methodologies for ocean acidification (OSPAR request 2010/2) To provide, on the basis of a review of existing methodologies and experience, recommendations for cost efficient methods for monitoring ocean acidification (OA) and its impacts, including possibilities for integrated chemical and biological monitoring. Specifically this should provide:

   i) advice on appropriate parameters, protocols and quality assurance for monitoring changes in pH and inorganic carbon chemistry in the OSPAR maritime area and other ancillary parameters that should be included in monitoring programmes

   ii) advice on the status of current knowledge of spatial and temporal variability of pH and inorganic carbon chemistry in the OSPAR maritime area

   iii) advice on appropriate spatial and temporal coverage for monitoring, considering different oceanographic features and conditions and key habitats/ecosystems at risk from OA in the OSPAR maritime area,

   iv) Contaminants

4.i Guidelines for Monitoring PCDD/Fs and "dioxin like" PCBs

   i) Finalize technical annex for monitoring PCDD/Fs and "dioxin like" PCBs in sediments (with WGMS); (OSPAR 2008/4)

   ii) Respond to comments received by ICES on consideration of cell based assays in biota monitoring guidelines for dioxins

4.j Report on new information regarding emerging contaminants in the marine environment.

Lutz Ahrens: *Partitioning Behaviour of Polyfluoroalkyl Compounds (PFCs) in the Marine Environment.*

Norbert Theobald: *Emerging pollutants in the North and Baltic Seas.*

Ralf Ebinghaus: *PFCs in the marine atmosphere.*

Katrin Vorkamp: *Temporal trend of hexabromocyclododecane (HBCD) in ringed seal from East Greenland.*

4.k MCWG to contribute, as may be required, to ICES activities on integrated chemical and biological effects monitoring and review new information on effect directed chemical analysis.

4.l Complete guidelines for publication in ICES TIMES series: Determination of PFCs in marine sediment, biota and seawater; Guidelines on use of silicone rubber passive samplers.

4.m Provide advice on whether it is appropriate to include PFOS in atmospheric monitoring programmes and if other perfluorinated compounds should be in-

cluded in such monitoring to support assessments of inputs of PFOS to the marine environment (OSPAR request 2010/6).

5   PLENARY DISCUSSION OF DRAFT REPORT

6   ANY OTHER BUSINESS

7   RECOMMENDATIONS AND ACTION LIST

8   DATE AND VENUE OF THE NEXT MEETING

9   CLOSURE OF THE MEETING

Annex 3: MCWG terms of reference for the next meeting

The **Marine Chemistry Working Group** [MCWG], chaired by Katrin Vorkamp, Denmark, will meet in Gothenburg, Sweden from [Dates to be confirmed] 2011 to:

a) Report on developments with regard to quality assurance of marine chemistry, in particular with respect to QUASIMEME,

b) Water Framework Directive and Marine Strategy Framework Directive:

1. Report on the developments in Water Framework Directive monitoring programmes and developments under the Marine Strategy Framework Directive,

2. Discuss suitable statistical methods of compliance checking of Environmental Quality Standards.

c) Provide expert knowledge and guidance to the ICES Data Centre, as may be required,

d) Describe MCWG interests and activities on the interface to other expert groups (e.g. WGMS, WGEel, WGBEC, SGIMC, SGONS),

e) Discuss the need and availability of certified reference materials for the analysis of nutrients in seawater,

f) Ocean acidification:

1. Report on recent developments in methodology of pH measurements and required standardization procedures,

2. Report on recent results of measurements of pH in marine systems (especially in the OSPAR area),

3. Report on recent developments in quality assurance issues in the measurement of carbonate system parameters,

4. Review new information for determination of pH and carbonate parameters in estuarine and brackish water and the associated calculations,

g) Contribute, as may be required, to ICES activities on integrated chemical and biological effects monitoring and review new information on effect directed chemical analysis,

h) Report on new information regarding emerging contaminants in the marine environment,

i) Discuss the role of atmospheric transport and deposition for the assessment of inputs of PFOS and other PFCs to the marine environment,

j) MCWG members to report information on projects of relevance to MCWG activities,

k) Complete guidelines for publication in TIMES series: Determination of dioxins/furans and dioxin-like PCBs in biota and sediment.

MCWG will report by 30 April 2011 to the attention of the SCICOM and ACOM.

Supporting Information

Priority	This group maintains an overview of key issues in relation to marine chemistry, both with regard to chemical oceanography and contaminants. The activities are considered to have a high priority. MCWG provides input across the field of marine chemistry, which underpins the advice given by ICES, and also supports the work of national and international collaborative monitoring programmes, e.g. within OSPAR.
Scientific justification and relation to action plan	a) MCWG has a particular interest in quality assurance and maintains strong links with QUASIMEME with a view to supporting quality assurance activities in this field. b) This work was initiated by MCWG and will be of interest to EU/OSPAR/HELCOM. c) This is in direct response to possible requests by the ICES Data Centre. d) This was initiated by SSGHIE. MCWG will follow up on this point to identify areas of common interests and complementary expertise. e) This activity has arisen from a plenary presentation at MCWG 2010 and cooperation with SCONS. f) This follows up on an OSPAR request at MCWG 2010. g) This item was initiated by MCWG members and will be of interest to OSPAR in relation to OSPAR request no. 8, 2008. h) This was initiated by MCWG members on the basis of concerns regarding emerging contaminants in the marine environment and is an ongoing area of interest to the group. i) This follows up on an OSPAR request at MCWG 2010. j) MCWG members are interested in receiving reports on relevant projects and activities from other members. k) This work was initiated by MCWG with a view to achieving wider dissemination of guidelines initially prepared in response to an OSPAR request for technical annexes.
Resource requirements	The resource required to undertake activities within the framework of this group is negligible.
Participants	The Group is normally attended by some 20–30 members.
Secretariat facilities	None.
Financial	No financial implications.
Linkages to advisory committees	ACOM
Linkages to other committees or groups	WGMS, WGBEC, SCICOM, SGONS
Linkages to other organizations	The work of this group is closely aligned with work being undertaken within the EU Chemical Monitoring Group on the requirements and implementation of the Water Framework Directive. The group provides the basis for some advice to OSPAR.

Annex 4: Recommendations

RECOMMENDATION	FOR FOLLOW UP BY:
1. Compliance checking of Environmental Quality Standards A suitable statistical approach should be chosen for compliance checking, taking into account i) that most environmental monitoring data do not follow a normal distribution and ii) that measurement data include an uncertainty (MCWG 2010 report, Section 5.2).	WG-E (Priority substances)
2. Draft guidance on monitoring of sediment and biota under the Water Framework Directive CMA to consider the comments of MCWG as forwarded to Patrick Roose in the final revision of the biota and sediment monitoring guideline (MCWG 2010 report, Section 5.2)	CMA
3. German proposal on reporting of QA/QC data MCWG supports the German proposal, provided that the changes do not conflict with OSPAR calculations. As a minor detail, MCWG suggests changing the reporting requirements to "mandatory" regarding the information of the laboratory being accredited for the particular method or parameter. Other recommended changes are: Information on QM system: Mandatory Reporting of "Uncertainty value" and "Method of calculating uncertainty": Mandatory. Reporting of "Reference material mean value found": Optional, subject to OSPAR acceptance. Further changes could be considered, e.g. information on method for LOQ determination (MCWG 2010 report, Section 5.4).	ICES OSPAR
4. ICES Data Centre request The reporting of trace metal concentrations in seawater should include information on the filtration method, i.e. the type of filtration; the filter material and the porosity (see Annex 7). The list of pretreatment methods in the database can be structured slightly differently (MCWG 2010 report, Section 5.4 and Annex 7)	ICES
5. Absolute salinity ICES to note this new definition of salinity and possible consequences for the database (MCWG 2010 report, Section 5.6).	ICES
6. Ocean acidification: OSPAR to note the advice of MCWG in support of coordinated monitoring of ocean acidification in the OSPAR maritime area (MCWG 2010 report, Section 5.8).	OSPAR
7. Ocean acidification: A limited life ICES/OSPAR study group should be established to further develop coordinated monitoring of ocean acidification in the Northeast Atlantic (MCWG 2010 report, Section 5.8).	ICES OSPAR
8. Technical annex on PCDD/Fs and dioxin-like PCBs in biota. OSPAR to note that the guideline has been revised (MCWG 2010 report, Section 5.9 and Annex 9).	OSPAR

9. Advice on atmospheric monitoring of PFOS	OSPAR
Further method development (including sampling and sample preparation) is necessary before MCWG is in the position to provide advice to include PFOS or other ionic PFCs, including PFOS precursors, in atmospheric monitoring programmes.	
To support assessments of inputs of PFOS to the marine environment, further research on the wet deposition of PFCs and the partitioning of ionic PFCs between the particulate and gaseous phase would be needed for an improved understanding of the importance of atmospheric transport and deposition.	
Current research programs should be used to fill the gaps explained above, e.g. in terms of existing infrastructure from monitoring programmes (MCWG 2010 report, Section 5.13).	

Annex 5: Proposal by Germany on reporting of QA/QC data to the ICES Data Centre (to be addressed under 5.4)

Proposal by Germany how to improve the reporting of data on quality assurance and control within the ICES Integrated Environmental Reporting Format

The ICES Integrated Environmental Reporting Format version 3.2.3 (ERF3.2.3) is an important tool in order to provide access to marine data of a standard format and known quality. In order to ensure the comparability and quality of data information of quality assurance and control are requested. Supporting analytical quality control information includes e.g. data on analyses of certified reference materials (CRMs) and results from interlaboratory comparisons and laboratory performance studies.

At present expert task groups develop a concept for the status assessment under the Marine Strategy. The assessment of European Seas regarding contaminants and pollution effects under the Marine Strategy Framework Directive is recommended to be based upon monitoring programmes covering the concentrations of chemical contaminants in appropriate matrices (water, sediment, biota) and biological measurements relating to the effects of pollutants on marine organisms, populations, communities and ecosystems. These data should be interpreted against assessment thresholds designed to protect against of pollution effects, taking into account the provisions provided under the Water Framework Directive and the approaches developed under the Regional Seas Conventions.

To ensure the reliability and comparability of analytical results generated by laboratories performing the Water Framework Directive monitoring the COMMISSION DIRECTIVE 2009/90/EC laid down technical specifications for chemical analysis and monitoring of water status establishing minimum performance criteria, including rules on the uncertainty of measurements and on the limit of quantification of the methods. The basic principles of quality assurance and control are provided by the EN ISO/IEC 17025 which comprise general requirements for the competence of analytical laboratories and give advice in applying quality management systems.

In order to fulfil the requirements of the COMMISSION DIRECTIVE 2009/90/EC (see Article 6) the German marine monitoring laboratories agreed to establish quality management systems according to EN ISO/IEC 17025 in all laboratories of the German marine Monitoring Programme (GMMP). This includes a variety of internal and external quality assurance tools and will ensure that only validated data will be reported to national and international databases. Further they agreed to meet and report the minimum performance criteria for the uncertainty of measurements and the limit of quantification of the methods as set out in Article 4 of the COMMISSION DIRECTIVE 2009/90/EC.

According to the ICES Reporting Format following information on internal data quality assurance and control (QA/QC) are collected and have been reported by the German laboratories until now:

Parameter measurement record – RECID 10

mandatory: Limit of quantification

recommended: Uncertainty value, Method of calculating uncertainty

Reference material record - RECID 93

mandatory: Type (CRM; LRM, SRM) and Code of the reference material

recommended OSPAR: Reference material - basis of determination used in control chart analysis and Reference material mean value found

optional: Reference material′s standard deviation, Control chart - number of measurements, Control chart- period

In order to improve availability of high quality data, to reduce the effort of data input and to harmonize the data evaluation and assessment the German marine monitoring laboratories would like to suggest to reduce the amount and/or change the obligation status of selected information on <u>internal</u> quality assurance and control because these detailed information are specific for the measuring laboratory and will overload the national and international databases unnecessarily. This applies especially to the "Reference material mean value found". Instead of that it should be reported if a laboratory has established a quality management system according to EN ISO/IEC 17025 or not.

Further the reporting status of "Uncertainty value" and "Method of calculating uncertainty" should be changed from "recommended" to "mandatory" because we consider uncertainty of measurement to be a key indicator of the reliability of analytical results, and it should in any case be taken into account when evaluating compliance with assessment thresholds. The COMMISSION DIRECTIVE 2009/90/EC therefore demand (see Article 4): "Member States shall ensure that the minimum performance criteria for all methods of analysis applied are based on an uncertainty of measurement of 50% or below (k = 2) estimated at the level of relevant environmental quality standards and a limit of quantification equal or below a value of 30% of the relevant environmental quality standards."

Since the 2005 CEMP data assessment (OSPAR), data are given analytical weights according to the available information on QA/QC. Amongst others the CRM mean values are taken to be acceptable if the CRM lab concentration is within 25% of the CRM true concentration. Given the fact that not for all chemical parameters in relevant matrices CRM′s are available we would like to state further that also the use of internal reference materials should be documented within the ICES database.

Recommendation

The German marine monitoring laboratories recommend the following changes of the ICES Integrated Environmental Reporting Format, version 3.2.3:

Generally it should be reported "mandatory" if a laboratory has established quality management systems according to EN ISO/IEC 17025 or other equivalent standards accepted at international level and further it should be reported "optional" if it is accredited by an authorized organization

The status of reporting of "Uncertainty value" and "Method of calculating uncertainty" should be changed from "recommended" to "mandatory"

The status of reporting of "Reference material mean value found" should be changed to "optional" also within OSPAR reporting

For all chemical parameters for which no CRM′s are available the use of internal reference materials should be documented within the ICES database also

Literature

CEMP Assessment Manual "Co-ordinated Environmental Monitoring Programme Assessment Manual for contaminants in sediment and biota", OSPAR Commission 2008

COMMISSION DIRECTIVE 2009/90/EC of 31 July 2009 laying down, pursuant to Directive 2000/60/EC of the European Parliament and of the Council, technical specifications for chemical analysis and monitoring of water status

ICES Integrated Environmental Reporting Format, Version 3.2.3, 27 July 2009

<u>For further discussion please contact:</u>

Federal Environment Agency
Section II 2.5, Quality Assurance Panel (GMMP)

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Annex 6: Data Centre questions for MCWG (to be addressed under 5.4)

Data Centre questions for MCWG

Seawater matrix review

As described in Kees Booij's document, the participation of the ICES data centre in EMODNET triggered a review of the DOME database for handling dissolved, colloidal and total particulate trace metals and organics in seawater.

There are currently 3 matrixes for seawater data:

BF, "before filtration" (i.e. before separation of suspended solids by filtration or centrifugation) which was intended for reporting 'total' levels of contaminants in water samples, either by methods which involve analysis of the 'unfiltered' seawater, or by summing contaminant levels determined in the 'filtered' water with the appropriate values in the separated solids;

AF, "after filtration" which leaves the dissolved fraction after separation of solids by filtration or centrifugation; and

SPM "suspended particulate matter" where contaminants are reported in units of mass/volume.

A number of recommendations have been made which MCWG should consider in the broader context for all relevant parameter groups: i.e. metals, organics, nutrients and chlorophyll. We agree that the terminology is outdated and misleading and agree that MATRX terms "before filtration" (BF) and "after filtration" (AF) be deprecated. We prefer, however, to keep the matrixes because combining them would result in the loss of information from the export from DOME to EcoSystemData (which does not contain method information) and will affect other parameters.

Clearer definitions are needed and the addition of a colloid matrix will clarify how to report. Please comment on the following matrix suggestions considering metals, organics and other parameter reporting such as organic carbon (DOC, POC, CORG), nutrients (NTOT, NTRA,NTRZ) and chlorophyll etc.:

"total water – general oceanographic parameters and the sum of particulate and dissolved concentrations in the water column" (WATT)

"water filtrate – dissolved concentrations in the water column" (WATF)

"water colloid – colloidal concentrations in the water column" (WATC)

"water particulate matter" – particulate concentrations in the water column" (WATP).

Very few checks have been added in DATSU and, based on the Booijs document; MCWG should suggest other checks for DATSU and approve the following:

1) Data type "CW" (seawater), reported in ERF3.2 format, will require an analytical method record (type 21) to be associated with all parameter records (type 10). Data files that fail to comply will not enter DOME.

2) The field "Method of Pre-treatment" (RECID 21: METPT) will be mandatory for seawater data. Data files that fail to comply will not enter DOME.

3) All parameters reported in the filtrate and colloid matrix will require a filter to be reported with a size indication. Filters "MF", "PCF", and "SAR" without size should give an error.

4) The matrix "SPM" should report units of mass/volume such as "ug/l", "ng/l" etc. Particulates quantified on an SPM mass basis (ug/g SPM) will receive an error and be prevented entry from the database. Question: will this affect the current generic check "if MATRIX is "SPM" then basis should be "D" (dry weight)"?

MCWG is requested to check the options of METPT and make additions as needed to limit the amount of inconsistent codes and definition requests from data submitters

EMODNET

ICES is involved in the chemical lot of EMODNET and will need ad hoc support from MCWG.

Question: Concern has been expressed that EU directives force monitoring agencies to generate "total water" data which, for metals, is "useless" data. In order for the data centre to take this concern to EMODNET and other organizations, MCWG should elaborate on this concern in their report.

Annex 7: Methods of pretreatment (agenda item 5.4)

The following table includes the codes and descriptions of pretreatment methods, which have been sorted according to their application for water, sediment and biota analysis. All filtration methods are marked with grey and addressed separately (see below).

CODE	DESCRIPTION	WATER	SEDIMENT	BIOTA
ASH	Ashing the sample		X	X
CGT	preconcentration in gold trap	X	X	X
CP	Centrifuged/precipitated	X		
D	Drying1		X	X
DAIR	Drying - by air [2] (suggest ambient air)		X	X
DCHEM	Drying - chemically (with Na2SO4)		X	X
DEPUR	Depurated			X
DFRZ	Drying - freeze dried		X	X
DNO	Drying - none (fresh material)		X	X
DOVN	Drying by oven - temperature unknown [2]		X	X
DRY100	Drying >100 degrees Celsius [2]		X	X
DRY99	Drying <100 degrees Celsius [2]		X	X
F	Filtration/fractioning			
FCN	Filtered on cellulose nitrate filter			
FRZND	Frozen directly without pretreatment	X	X	X
GF/F	glass fibre syringe filter			
GFC	glass fibre cartridge			
GFC1.2	filtration, glass fibre, Whatman GF/C - particle retention of 1.2 μm and a weight of 53 g/m2			
GFF	glass fibre filter			
GFF0.7	filtration, glass fibre, Whatman GF/F - particle retention of 0.7 μm and a weight of 70 g/m2			
GFF100	glass fibre filter 100			
GFF200	glass fibre filter 200			
HMG	Homogenized	X	X	X
HYDL	Hydrolysis		X	X

[1] Should become drying - other; [2] temperature should be mentioned e.g. in a different subset of data or new field.

CODE	DESCRIPTION	WATER	SEDIMENT	BIOTA
HYDM	Hydrometer	X	X	
IGN	Ignition		X	X
MAN	Manual milling (mortar and pestle)		X	X
MF	Filtered on membrane filter			
MF120	Filtered on membrane filter 1.20 µm			
MF20	Filtered on membrane filter 0.20 µm			
MF45	Filtered on membrane filter 0.45 µm			
MF80	Filtered on membrane filter 0.80 µm			
MMG	Mechanical milling/grinding		X	X
N20	Filtered on Nuclepore filter 0.20 µm			
N40	Filtered on Nuclepore filter 0.40 µm			
NA	Not applicable	X	X	X
NDEPUR	Not-depurated			X
NF	Not filtered	X		
NONE	No pretreatment	X	X	X
PCF	Filtered with polycarbonate filter			
PCF20	Filtered with polycarbonate 0.2 µm			
PCF45	Filtered with polycarbonate 0.45 µm			
PIP	Pipette analysis		X	
S	Sedimentation	X		
SAR	Sartorius filter			
SON	Sonicate	X	X	X
SVD	Sieving - Dry sieving		X	
SVW	Sieving - Wet sieving		X	
UF1	Filtered with 1 kDa ultrafilter			
UV	Ultraviolet radiation	X		
WGFC	Whatman glassfibre filter concentrate			

MCWG suggest structuring the filtration methods in a different way, which removes the information on specific suppliers (e.g. Sartorius filter, Nuclepore GF/F are Whatman trademarks, etc.) The important piece of information is the type of filtration (membrane filtration, fibre filtration or ultrafiltration), the filtration material and the porosity of the filter. We suggest dropdown menus for the filtration method, membrane material and porosity, e.g.:

Membrane filtration

Membrane material	Abbreviation	Pore size (μm)
Cellulose nitrate	CN	0.1
Cellulose acetate	CA	0.2
Regenerated cellulose	RC	0.22
Polyethersulfone	PES(U)	0.45
Polyvinylidene fluoride	PVdF	0.65
Polytetrafluoroethylene	PTFE	0.8
Polycarbonate	PC	1.2
Polyamide	PA	3.0
Other	…	5.0
		8.0
		10.0
		20.0

Fibre filtration

	Abbreviation	Nominal retention diameter (μm)
Glass fibre	GF	0.7
Quartzfiber	QF	1.0
Polypropylene	PP	1.2
Other	…	1.5
		1.6
		2.0
		2.7

Ultrafiltration

Membrane material	Abbreviation	MWCO (kD)
Regenerated cellulose	RC	1
Polyethersulfone	PES(U)	2
Polyvinylidene fluoride	PVdF	3
Polytetrafluoroethylene	PTFE	5
Polyamide	PA	10
Other	…	20
		30
		50
		80
		100
		300
		1000

Annex 8: Report in response to OSPAR request for information needed to support the development of monitoring of ocean acidification (agenda item 5.8)

1. Background information and monitoring objectives

1.1 Background

Recent reports have identified ocean acidification (OA) resulting from the absorption of anthropogenic CO_2 by the oceans as a major concern because of its potential effects on marine biogeochemistry and ecosystems and the lack of appropriate information for assessing the risks (e.g. Raven *et al.*, 2005). Acidification (measured as reduction in pH) is a certain consequence of the rise in atmospheric concentrations of CO_2 (carbon dioxide) and the resulting net oceanic CO_2 uptake.

There is only limited data available to assess the vulnerability of different areas to change and to understand the already identified spatial and interannual variability of oceanic CO_2 uptake (Schuster *et al.*, 2009). Atmospheric CO_2, has small spatial gradients, and is dispersed. However, local factors influence air-sea CO_2 exchange. For example, local variations in total alkalinity (TA) have an influence on CO_2 uptake.

Some marine regions will be more rapidly affected than others but ultimately all marine regions will be impacted. The susceptibility of a water to change depends on the chemical composition and temperature of the water. The rate of decrease in pH is small (*ca*. 0.001 pH units yr^{-1}) and the build up of anthropogenic CO_2 is likely to become biologically significant over decades. However local processes have already been seen to cause more intense than expected changes (Thomas *et al.* 2007; 2009; Feely *et al.*, 2008; Wooton *et al.*, 2008).

At temperate latitudes the natural annual cycles and interannual fluctuations of temperature and biological production result in a natural cycle and interannual fluctuations in pH that are large compared to the likely net annual rate of decrease, consequently a long-term monitoring programme must be designed to discern between the long-term trend and these short term fluctuations. Waters where there is enhanced production due to nutrient enrichment will have a larger cycle in biological production and respiration and consequently greater than natural range in acidity through a year.

The seas of NW European shelf area may be flushed by ocean water at such a rate that it is the change in pH in the ocean water that may be the primary determinant of the underlying long-term rate of change in pH of these shelf seas. In turn the rate of increase in acidity in ocean waters will vary from year to year in line with the changes in the amount of uptake of atmospheric CO_2. Variations in uptake are a result of variations in temperature, biological activity and mixing between surface and deeper waters.

Many potentially relevant processes in shelf seas are poorly described at present, such as inputs from rivers producing enhanced production and respiration, factors influencing TA and reactions with the benthos. Both monitoring and process studies in shelf seas are required so a distinction between ocean control and control by local processes can be made.

The international research community through the SOLAS/IMBER ocean acidification working group are also considering how a global observational network for ocean acidification can be established (Feely *et al.*, 2010).

1.2   Objectives of monitoring

As context for responding to the request it was considered necessary to elaborate on the rationale and primary objectives for an OSPAR ocean acidification monitoring programme as this relates to the physico-chemical aspects of such a programme.

Key objectives were considered to be:

- o   Assemble a baseline dataset against which long-term ocean acidification monitoring in key water bodies in OSPAR areas can be judged. The work required to measure the variability of the carbonate system on seasonal and interannual time-scales has already started (See Section 8). To ensure that results are not aliased by this variability, monitoring programmes to assess long-term trends will need to be scientifically and statistically robust (See Tables in Section 8).
- o   Determine medium to long-term temporal trends in pH and carbonate (calcite and aragonite) saturation state chemistry in surface, mode and deep water where appropriate in all OSPAR areas in order to:
    - •   Better understand the role of processes such as variation in the extent of deep winter mixing that control the properties of the carbonate system in the surface layer of the ocean.
    - •   Determine temporal changes to saturation status in deep waters (e.g. track Aragonite Saturation Horizon and Calcite Saturation Horizon) which will affect ecosystems such as cold water corals.
    - •   Support ecological monitoring and assessment of, for example, sensitive species and habitats especially those dependent on calcification, as part of an integrated chemical and biological monitoring programme.
    - •   Fulfil MSFD requirements for monitoring of the carbonate system as included in annex III of the Directive[1]).
- o   Carry out a programme of work that ties into and is consistent with other extant and planned global monitoring, modelling, assessment and research activities (See Section 10).
- o   Provide information to validate and improve models to better forecast environmental perturbations and ecological risks.
- o   Work in conjunction with groups gathering evidence of ecological status, in order to inform national and international policymakers of the impacts of increased global CO_2 concentrations and underpin the need for international agreements to reduce CO_2 emissions.

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[1] Table 1 of Annex III of Directive 56/2008/EC (MSFD) lists following physical and chemical features among the indicative list of characteristics: "pH, pCO_2 profiles or equivalent information used to measure marine acidification".

2   Required measurements and sampling

2.1   Required measurements

The four measurable parameters of the carbonate system are total alkalinity (TA), total dissolved inorganic carbon (DIC), carbon dioxide fugacity (fCO_2) and pH. The chemical equilibria connecting these species in solutions have been extensively quantified for seawater. Consequently measurements of any two components allow the concentration of the other two to be calculated. However the precision of this assessment varies with the pair chosen. There is no optimal choice of parameters and each has advantages and disadvantages (Dickson *et al.* 2010).

For these calculations, measurements of temperature and salinity are required with precisions of better than 0.05°C for temperature and 0.1 for salinity to achieve 0.001 precision in calculation of pH.

When calculating pH from measurements of DIC and TA concentrations of phosphate and silicate need to be known. Standard protocols for measuring these parameters in seawater are appropriate. (See MCWG 2010 Annex 13 for guidelines on nutrient monitoring). To achieve a pH precision of 0.001, precisions of 0.3 uM and 15 uM for phosphate and silicate are respectively required.

pH and fCO_2 can be measured with higher precision than they can be calculated, however the procedures for direct measurement of pH are not well established because universally accepted standardization procedures are only currently being developed.

TA and DIC is the preferred pair for calculation of pH and fCO_2 if these are not measured directly.

TA, DIC and pH together provide full coverage of the inorganic carbon system and also allow check of the internal consistency.

N.B. pH is an operationally defined concept and there are four different scales (US National Bureau of Standards (NBS), free scale, total hydrogen ion scale, seawater scale), which result in significantly different numerical values. The recommended scale for use in seawater related calculation is the total hydrogen ion scale. It is critical that the scale used is reported as part of the meta-data when data are included in a database. The temperature of the pH measurement must also be reported.

The assessment of change in the carbonate system is greatly assisted when ancillary data are available on hydrography, concentration of nutrients, dissolved oxygen and biomass.

Further details can be found in the EPOCA Project's handbook on acidification research (http://www.epoca-project.eu/index.php/Home/Guide-to-OA-Research/) and in Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) 2007. Guide to best practices for ocean CO_2 measurements. PICES Special Publication 3, 191 pp.(http://cdiac.ornl.gov/oceans/Handbook_2007.html)

2.2   Sampling and sampling platforms

Ocean acidification monitoring will require a combination of traditional hydrographic surveys and (semi-)continuous measurement of key parameters using autonomous instruments deployed on a variety of platforms. Building relevant measurements into other related programmes may also support cost-effective moni-

toring. For example, in some locations, incorporation of carbonate parameters into OSPAR eutrophication monitoring may provide a cost-effective approach to coastal and inshore monitoring.

Hydrographic cruises: Traditionally most marine research and monitoring surveys have been conducted using research vessels. These still provide the only mechanism by which subsurface samples can be collected over large areas and facilitate collection of high quality data for a much broader range of parameters than can be acquired using, for example, ships of opportunity. Monitoring of ocean acidification requires regular hydrographic cruises to measure the accumulation of anthropogenic DIC in mode waters and in deep waters in the region of sensitive eco-systems such as cold water corals. Monitoring of ocean acidification requires observations over large spatial scales because the input of atmospheric CO_2 is diffuse over the oceans and is not localized like many contaminants that, for example, come from estuaries. The standard World Ocean Circulation Experiment (WOCE) suggests a sampling strategy for oceanic studies of 30 nautical mile spacing for physical measurements, with higher resolutions in regions of steep topography and boundary currents, and 60 nautical miles or better for carbon and tracer measurements [Hood *et al*. 2010]. Additionally high frequency sampling is needed to quantify intra- and interannual variability. To achieve the range of sampling required other more widely available and cost-effective ways of sampling need to be developed and used. Much of the carbonate system data that has been collected in the North Atlantic Ocean on hydrographic cruises is now being systematically assessed in the CARINA project (see Section 10).

Ships of opportunity (SOO) and FerryBoxes: One mechanism for providing the needed spatial and temporal coverage is through the use of ships of opportunity carrying autonomous sensors. These are used widely with great success (Watson *et al*., 2009) for monitoring the surface water pCO_2 values. pCO_2 is closely related to the pH of the water. pH can be calculated successfully ($\pm$ 0.002) by estimating the TA of the water from the salinity in many areas. The regions in which this is possible is being extended by the addition of the routine collection of water samples on these ships by the ships crews on a number of underway lines. In areas where such estimates are not available or possible the measurement of a second parameter needs to be added for ocean acidification monitoring and research. The activities are coordinated globally by the IOC sponsored IOCCP (International Ocean Carbon Coordination Project - www.ioccp.org). The extent of global coverage can be seen via the CDIAC web page http://cdiac.ornl.gov/oceans/VOS_Program/VOS_home.html.

Much of the effort of the IOCCP project focuses on the oceans. Within NW European Shelf seas, additional systems fitted to FerryBoxes (www.FerryBox.org) could provide a very effective network for monitoring (see below).

Similarly pCO2 systems have and are being fitted to research vessels operating in shelf seas. This also provides another valuable source of data with the advantage that the measurements can be coordinated with process studies on the research vessel. (In the UK this has been coordinated in the Carbon-OPS project (www.bodc.ac.uk/carbon-ops, Hardman-Mountford *et al*., 2008).

Buoy/Moorings: Instrumented buoys and moorings also provide platforms for the collection of detailed time series data see http://cdiac.ornl.gov/oceans/Moorings/moorings.html & http://www.eurosites.info/. At present, technologies for autonomous moorings are not fully mature. Several different strategies are being employed in the development of reliable instrumentation and sensors. The Batelle pCO2 buoy systems, developed by NOAA and MBARI, are

currently used operationally in the US for open ocean and shelf-sea monitoring systems, with more than 20 systems deployed to date.

3   Suggested sampling

3.1   Rationale

At present we lack reliable knowledge of how ocean acidification is likely to progress in different areas. The information that is available suggests that the rate of change is variable with both time and location. Observations need to document current variability of the full spectrum of areas covered by OSPAR from the open Atlantic and Arctic Oceans to estuarine regions. It is essential that we have knowledge of the daily to seasonal to interannual variations in each area. This knowledge is required for the design of a long-term monitoring programme that will avoid aliasing assessments due to a poor knowledge of short-term variability. Relevant recent and planned activities in the OSPAR area are listed in Section 7.

Monitoring needs to cover the range of waters from estuaries, shelf seas, ocean mode waters and abyssal waters where sensitive ecosystems may be present. That is the spectrum of waters in all OSPAR areas. Particular emphasis should be placed on key areas at risk within the OSPAR area, for example high latitudes where ocean acidification will be most rapid, and areas identified as containing ecosystems and habitats which may be particularly vulnerable, e.g. cold water corals.

Much of the required monitoring can be done in conjunction with existing activities carried out by operational agencies and scientific groups making sustained observations. Work will need to be done using the range of platforms described in Section 2.2.

High repeat rate observations will be necessary in the first phase for example sampling of shelf seas should be carried out with an a minimum of a monthly repeat rate. For off shelf work coordination is required with regular hydrographic cruises which are being under taken at least once a year such as IEO's cruises in the Bay of Biscay and NERC "Ellett Line" cruise between Scotland and Iceland.

3.2   Suggested measurements scheme based on locations for repre-
sentative monitoring that exist or could be rapidly established
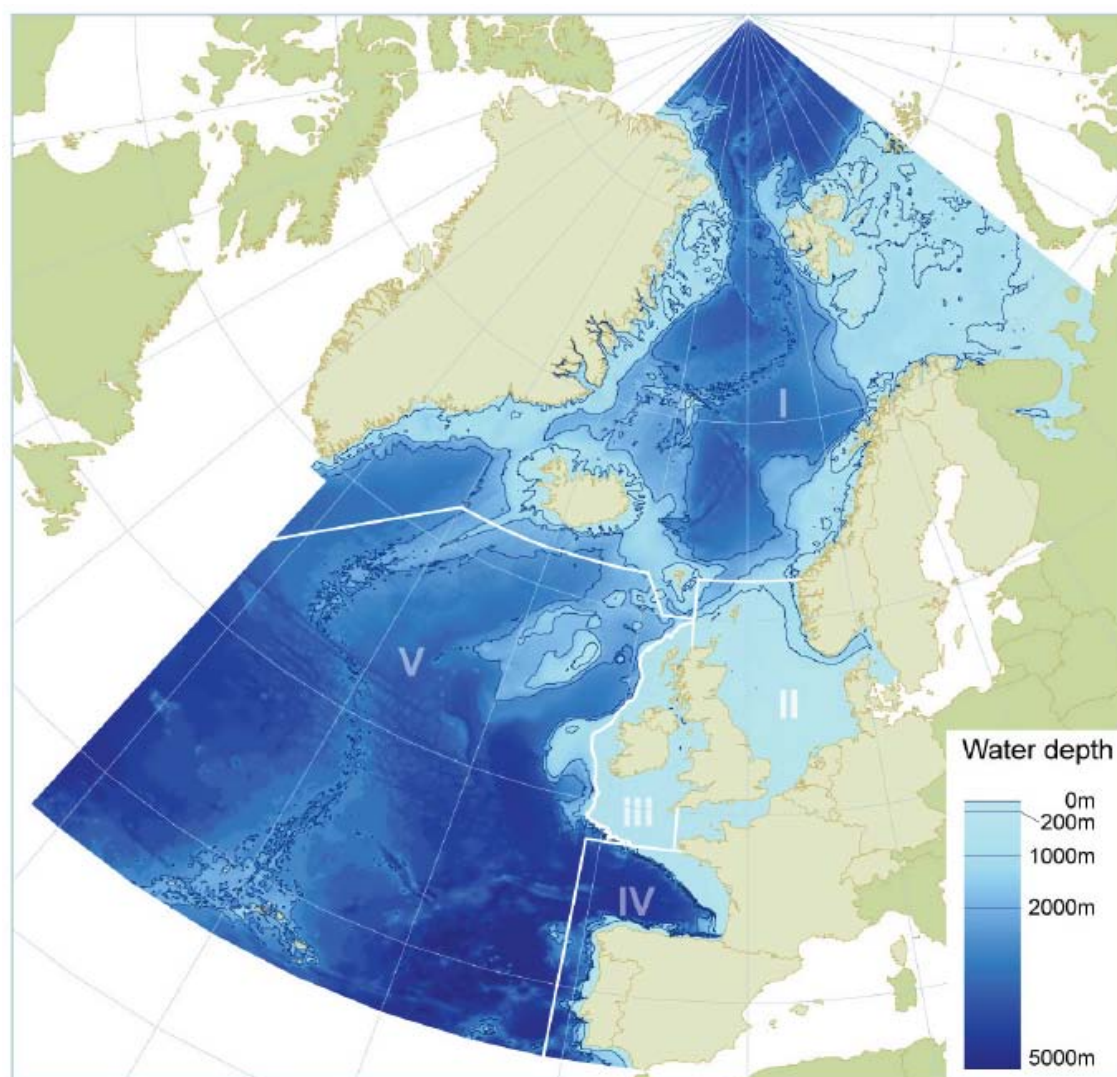
Figure 3.1. Map of the OSPAR maritime area.

3.2.1   Open Ocean – Arctic, Atlantic (areas 1 and 5)

In the long term, the accumulation of CO_2 in ocean waters will determine what happens in shelf seas as these are the main source waters for the shelf.

<u>SOO (Ships of Opportunity)</u> An effective basis for monitoring of surface waters exists in the SOO operations that are already being conducted for the study of air-sea CO_2 fluxes. These may in future be supported by ICOS. For the study of acidification, measurement of TA is planned to be added to the work on lines that are being included in national OA measurement programmes.

At present, measurements in the surface are under taken by:

1) UK (UEA) – Portsmouth- Caribbean – with a time-series extending back to the 1995/6 and continuous from 2002. This samples water adjacent to NW European shelf and the route crosses the PAP mooring site (which provides data on deep mixing). (pending CarboChange,)

2) No (UB) – Copenhagen- Greenland route sampling on this route began in 2002. Provides data from OSPAR areas 1, 2 and 5 (pending CarboChange)

3) No (NIVA) New SOO line Tromsø to Spitsbergen – existing SOO line to which CO_2/TA measurements could be added to collect data in the Arctic. Should be supported due to the potential sensitivity of Arctic waters to acidification.

Hydrography: Knowledge is needed of: (1) the variability of subsurface CO_2 accumulation and transport back to the surface as result of variation in the depth of deep winter mixing, (2) subsurface structures of water adjacent to the shelf that are source waters to the shelf. (3) the change in saturation state of aragonite and calcite. This requires cruises that are conducted on a regular basis reoccupying the same stations at least once per year. Consideration needs to be given to sampling in deep waters in the location of potentially sensitive ecosystems such as cold water corals.

This type of hydrographic measurements are undertaken by:

1) Es (IEO) winter and summer cruises in the Bay of Biscay. The Bay of Biscay is a system with limited amount of advection in the deeper waters making it a good location to study as changes tend to be due to local vertical processes rather than large-scale advection. (Spanish monitoring funded – CO_2 is not supported for regular work.)

2) UK (NERC) Ellett Line Scottish west coast to Iceland (samples in OSPAR areas 3 and 1/5) surveys – annual or more frequent surveys through the source waters for the North Sea. – (measurements of TA/DIC are not supported for regular work; surface pCO2 has been measured for 3 years through the CarbonOPs project)

3) IC (MRI) Time series measurements (quarterly) of carbon parameters in the Irminger and Iceland Seas as a part of repeat hydrography network (OSPAR area 1)

4) Other relevant hydrographic cruises on the northern edge of the North West European shelf are conducted by Norwegian MRI and UK Marine Scotland. The Marine Institute with the National University of Ireland Galway (IE) have conducted surveys extending across the Rockall Trough to the west of Ireland.

Buoy/Moorings: Buoys and moorings provide high resolution data on both the air sea transfer of CO_2 and deep mixing related transport.

At present mooring measurements are undertaken by:

1) UK (NOCS) Porcupine Abyssal Plain -PAP observatory mooring (OSPAR region 5) – pCO2 measurements were made successfully at this site in 2005 and a newly designed mooring with measuring pCO2 will be deployed in May 2010. This site is important as the observations complement those made by the UEA –SOO line (see above) and are in an area of the N Atlantic where we now know CO_2 uptake is variable but the cause of the variability has not been defined.

2) No UB Ocean Weather-ship Station Mike site (OSPAR region 1): Time series of information for CO_2 at this site is available up to 2009. A mooring replacing the Weather-ship is planned.

3.2.2 Open Seas North, Celtic and Iberian (areas 2,3, and 4)

A key to understanding the impact of OA in open shelf seas is the identification of the transition between waters that are influenced by changes in the ocean source waters and those waters where local shelf sea processes are important, such as river inputs or interaction with seabed processes such as denitrification.

SOO – SOO based observations provide the most cost-effective way of monitoring this transition. This can be done in the North Sea using one line that is already instrumented for pCO_2 observation which runs North/South. A second orthogonal SOO line is operated at the moment but needs to be upgraded for pCO_2 measurements.

1) No (UB) and N (NIOZ). SOO line Bergen – Amsterdam has been making pCO_2 measurements since 2006

2) No (NIVA) and UK (Marine Scotland). SOO line Bergen – Aberdeen recommended to add pCO_2 to look at the inflows and outflows to the North Sea

Hydrography – detailed surveys of the greater North Sea have been undertaken by NIOZ in 2001/2 (4 seasons), and the summers of 2005 and 2008, with a new survey planned for summer, 2011. These provide considerable background knowledge of regional difference and the ability to estimate fluxes to and from the North Sea. These are research exercises rather than the basis for a cost-effective monitoring programme. In recent years the Marine Institute/National University of Ireland Galway (IE) have undertaken surveys of shelf waters to the west of Ireland, including an annual winter transect on 53°N that further extends NW across the Rockall Trough (region 5).

3.2.3 Coastal and Estuarine

Regions of freshwater influence (ROFI) are already areas of concern for marine monitoring because of high inputs of nutrients and enhanced levels of production and respiration. These should also be monitored for pH because of the associated enhancement in the variation of the acidity (e.g. Rhine and Thames plumes in the North Sea). Coastal waters are complex environments in which pH changes are determined by processes that control both the DIC and TA content of the water. These waters need to be studied both from the point of view of their potential vulnerability to ocean acidification and because the pH is an important variable affecting the speciation of other components such as ammonia and trace metals making them more or less available to biota. In these areas studies of acidification need to be fully integrated with existing monitoring which provides information on the processes driving changes in the carbonate system.

OSPAR should look to promote and support work that covers the range of coastal and estuarine areas, from relatively pristine regions with freshwater inputs that are low in both nutrients and organic carbon such as those on the west coasts of Scotland, Norway and Sweden, to the estuary of the Scheldt which has high inputs of organic carbon and the Thames and Severn with high inputs of nutrients.

Collection and preservation of samples for the determination of carbonate species is a relatively simple task so that potentially many existing coastal and estuarine monitoring programmes could be extended to include the collection of the required samples. The degree to which this can be done will depend on the capability and capacity of individual national groups to measure the samples. Certainly, additional funding commitments will need to be obtained from the government departments responsible

for financing national monitoring programmes. Initially this might be done by sub-contracting the processing of samples to laboratories which already have a recognized capability in these measurements.

Existing work which provide example of the approach needed include:

- UK PML Stations E1, L4 Coastal sites salinity >34. Existing monitoring on a monthly and weekly basis respectively in surface and subsurface waters. These sites represent relatively pristine waters. These also provide a historical context to changes in hydrography that can in the case of the E1 site be traced back over 100 years.
- NL RIKZ/Deltares Dutch Coastal Grid; high flow.
- UK NERC POL Liverpool Bay; high load and production.
- BE Scheldt Estuary; high nutrients and high organic input, low pH, high CO2 water.
- Other key areas are the rivers Gironde and Loire which have high flows greater than that of the Rhine.

4   Methods, Quality Control and Quality Assurance

4.1   Methods

Work on ocean acidification will build on the research and development activities that have gone into the precise studies of carbonate chemistry and the work of the CO_2 gas transfer community. Current best practice for the analyses has been carefully described by Dickson *et al.* (2007) in a series of standard operating procedures that cover both the methods and basic quality control procedures. The Dickson manual is available on line at http://cdiac.ornl.gov/oceans/Handbook_2007.html. Further relevant information is also being compiled by the EPOCA project and is available at http://www.epoca-project.eu/index.php/Home/Guide-to-OA-Research/. (Dickson *et al.,* 2010).

The basic methods in common use are:

- TA: Potentiometric titration (open or closed cell)
- DIC: Acidification followed by infrared detection or coulometric titration
- pH: Spectrophotometric detection or glass electrode.
- fCO_2: Sea water in equilibration with air and infrared detection

Details of the equipment available to carry out the analyses and current suppliers of the equipment can be found on the IOCCP web pages at http://www.ioccp.org/Sensors.html. Following best practice it is considered that experienced laboratories should be able to attain the following precisions when making direct measurements of particular variable.

- TA: For closed cell ± 3 μmol kg^{-1}, for open cell ± 1.0 μmol kg^{-1}
- DIC: ± 1.5 μmol kg^{-1}
- pH:
    - Spectrophotometrically ± 0.001 pH units
    - Glass electrode 0.003 pH units
- fCO_2: ± 1–2 μatm

4.2   Calibration and quality control

4.2.1   DIC and TA

To assess measurement accuracy certified reference materials (CRM) are available to control measurements of DIC and TA. The carbonate analysis community has set up a reference material supply service provided by Andrew Dickson's laboratory at the Scripps Institute of Oceanography (University of California). This operation is part funded by NSF and runs on a non profit basis. These reference materials consist of natural seawaters sterilized by a combination of filtration, ultraviolet radiation and addition of mercuric chloride. They are bottled in 500 ml borosilicate glass bottles sealed with greased ground glass stoppers. Each batch prepared is then analysed for salinity, total dissolved inorganic carbon and total alkalinity, using the best available methodologies. (http://andrew.ucsd.edu/co2qc/index.html).

A cause for concern is the future supply of CRMs. Presently these are produced on limited scale ("cottage industry") in Andrew Dickson's laboratory. Increased research into ocean acidification has the potential to increase the demand for CRMs beyond the capacity of this laboratory. Dickson is working with Akihiko Murata (JAMSTEC, Japan) and others to develop an alternative and larger source of supply.

The Dickson CRMs are used widely to control the accuracy of DIC and TA measurements; however there is no feedback of the data obtained by individual laboratories. The ICES MCWG was one of the first groups to recognize the value of intercomparison exercises in improving the comparability of data from different laboratories and this lead to the establishment of QUASIMEME ("Quality Assurance of Information for Marine Environmental Monitoring in Europe" – www.QUASIMEME.org). MCWG 2010 discussed the possibility of using the Dickson CRMs as the basis of a laboratory performance study for determinations of DIC and TA with the QUASIMEME representative.

4.2.2   pCO$_2$

The NOAA Carbon Cycle Greenhouse Gases Group (CCGG http://www.esrl.noaa.gov/gmd/ccgg/refgases/stdgases.html) is currently responsible for maintaining the World Meteorological Organization mole fraction scales for CO_2, CH_4, and CO. With the mission of propagating this scale for data intercomparison, CCGG can fill and calibrate compressed gas cylinders for use as standard reference gases by other laboratories for measurements of CO_2, CH_4, CO, and the stable isotopes of CO_2 (^{13}C and ^{18}O). These gases form the basis of calibration of the non-dispersive infrared (NDIR) analysers that are used as the detector in most underway pCO$_2$ systems. Depending on the design of the system, two to four different concentrations of gas are used to provide regular calibrations. This enables accuracies of better than 1 μatm CO_2 to be achieved by some systems.

4.2.3   pH

At present no CRM is available for pH measurements. For work in seawater the measurement system needs to be calibrated with buffer solutions made up in water of similar ionic strength to the seawater being measured. The buffer compound used is "TRIS" (2-amino-2-hydroxymethyl-1,3-propanediol). Carefully prepared solutions have a high level of stability with a drift rate typically$\leq$ 0.0005 pH units per year (Nemzer and Dickson, 2005). The uncertainties arising from the preparation of such buffers is typically less than 0.002 in pH.

5 Novel methods and platforms

5.1 Methods

Methods for the determination of DIC and TA in discrete water samples and underway measurement of pCO_2 using equilibrator based systems are now well established. However measurements of DIC and TA are relatively time consuming with throughput of only 3 measurements per hour when using the VINDTA system. There are obvious advantages to speeding up processing which ideally should be developed to a state where it could operate as part of an autonomous system. Measurements of pH using glass electrodes have been a cause of concern in recent years because of problems with both the stability of electrodes and the production of suitable buffer solutions. Recent developments on both fronts have lead to reassessment of the approach and further work is continuing in this area. The electrode approach looks like it will be best suited to use in the laboratory. Colorimetric methods are working well in some laboratories (IMM Vigo for example). Colorimetry has been used in experimental underway system (Bellerby *et al.*, 1995; Friis *et al.*, 2004) and this has lead to a number of development projects to produce instruments that can be operated reliably as part of underway systems.

Developments in sensors and instruments were reviewed by Schuster *et al.* (2009b) and Byrne *et al.* (2010) as were potential applications in the coastal ocean by Borges *et al.* (2010).

5.2 Platforms

Moorings: moorings such as the Smart-Buoys operated by Cefas have considerable potential to provide a platform for carbonate system measurements in coastal seas. Presently problems with the reliability of pCO_2 and pH sensors and their potential to bio-fouling need to be addressed – evaluations of likely systems are being carried out by ACT (Alliance for Coastal Technologies - www.act-us.info). See Section 5.3.

Argo floats: Currently available sensors for carbonate variables have response times that are too slow and power consumptions which are too high for use either on Argo floats or gliders. However those working on the miniaturisation of systems are aware that the study of the carbonate system would be greatly enhanced if fitting suitable sensor to Argo Floats and Gliders were possible.

5.3 Ongoing developments

The meeting developed the following summary of ongoing developments.

5.3.1 pCO_2 on moorings

Measurements of pCO_2 on mooring have been made less widely than on SOOs and the technology is less mature than for underway systems. Particular issues to be considered for mooring applications are long-term stability of sensors, biofouling and power consumption. Measurements may be made with high temporal resolution from such platforms but have low spatial resolution.

a) Pro Oceanus (http://www.pro-oceanus.com/products_CO2.html), taking part in the Alliance for Coastal Technology evaluation of in situ pCO_2 analysers (http://www.act-us.info/evaluation/rft.php)

accuracy ± 1ppm CO_2, precision ±0.01 ppm CO_2

b) Battelle (NOAA) http://www.battelle.org/seaology/

accuracy 0.01 ppm CO_2, precision ~1ppm CO_2

c) Contros (www.contros.eu)

accuracy < 10ppm CO_2

d) SAMI [2]-CO_2 (http://www.sunburstsensors.com/), taking part in the Alliance for Coastal Technology evaluation of in situ pCO_2 analysers (http://www.act-us.info/evaluation/rft.php)

accuracy ± 3 ppm CO_2, precision <1ppm CO_2, long-term drift <1ppm CO_2 over 6 months

e) CARIOCA (http://www.dt.insu.cnrs.fr/carioca/carioca.php)

accuracy ± 3 ppm CO_2, precision ±1 ppm CO_2

5.3.2 pCO_2 on vessels

pCO_2 measurements have been made on vessels since the mid 1990s and the technology is more mature than that for *in situ* measurement systems. A mixture of commercial and custom made systems exist. Measurements made on regular transects, such as from ferries and shipping lines, allow a system to be characterized over time. Measurements made from research vessels may provide wider spatial resolution but lower temporal resolution.

a) Pro OCEANUS (http://www.pro-oceanus.com)

accuracy ± 1 ppm CO_2, precision ± 0.01 ppm CO_2

b) General Oceanics (http://www.generaloceanics.com/home.php?cat=69)

accuracy ± 1ppm CO_2, precision 0.01 ppm CO_2

c) Dartcom-PML   (http://www.bodc.ac.uk/carbon-ops/instrumentation/telemetry_and_pco2/)

accuracy ± 1 ppm CO_2, precision (LI-COR) 0.01 ppm, repeatability 0.2 ppm CO_2.

d) Kimoto air marine CO_2 system (Japan)

accuracy ?, precision ± 0.3 ppm CO_2, minimum detectable 0.1 ppm CO_2

e) Contros (www.contros.eu)

accuracy < 10 ppm CO_2

f) SAMI [2] – CO_2(http://www.sunburstsensors.com/)

accuracy ± 3 ppm CO_2, precision < 1 ±ppm CO_2, long-term drift < 1ppm CO_2 over 6 months

g) Apollo SciTech (http://www.apolloscitech.com/PCO2.htm)

Repeatability better than ±1 ppm CO_2

5.3.3   pH

SAMI[2] –pH (http://www.sunburstsensors.com/) designed for mooring and underway, measured on the total hydrogen ion scale precision ±0.001 pH, accuracy ±0.003 pH, long-term drift 0.001 pH over 6 months

6 Data management and assessment

Mechanisms to ensure timely data availability need to be considered and the barriers to this to achieving this addressed. As much relevant data are generated as part of research programmes, often by academic institutions, there may be a reluctance to report, especially unpublished, data. Nonetheless, the OA research community recognize the need for efficient data flows and products ultimately to inform stakeholders and policy development (Pesant *et al.*, 2010).

Data may be dispersed in a range of national and international projects (e.g. CAR-BOOCEAN, EPOCA) data centres. Data should be reported to both the Carbon Dioxide Information Analysis Center (CDIAC - http://cdiac.ornl.gov/oceans/home.html) and to the ICES data repository. Reporting data to the ICES data repository enables it to be linked to many related OSPAR datasets e.g. nutrients and integrated ecosystem data. Globally most research groups measuring carbonate parameters submit data to CDIAC. CDIAC has established reporting formats for these data and metadata, and has worked with the community to develop systems for effective data access and review such as the SOCAT (Surface Ocean CO_2 Atlas). ICES should consider how data reporting would evolve so that relevant data are available and accessible to both databases without replicating reporting requirements. Harmonised data vocabularies and meta-data reporting requirements need to be elaborated. Metadata should include details of methodologies and protocols including and protocols used for calculating parameters that are not directly measured.

ICES felt that the assessment process is one that needs further development. This needs to be done in coordinated way and be a process that includes chemists, ecologists and modellers to ensure that the data are used effectively. It was seen that numerical models could be used both to identify key areas at risk and guide the targeting of the monitoring programmes. The monitoring programme should also be designed to both support those modelling activities and respond to the findings of those activities.

Assessment should be based on:

- The collection of data in all OSPAR regions.
- An international and coordinated programme of long-term observations.
- Consistently measured data that are reported promptly with the appropriate meta-data.
- Assessment at an international level, in association with appropriate relevant global scientific activities (Currently IMBER for example).
- The key assessment will be the identification of temporal trends
- The development of appropriate indicators for summarizing findings for OSPAR, EEA and national policy related agencies.

7   Summary of past and current measurement activities in the OSPAR area and Baltic Sea

Listed are measurements on larger scientific cruises and/or repeated sections, time-series stations, ships of opportunity (SOO) and moorings that the group are currently aware of. The list should be considered to be incomplete.

Note: Only some of the listed activities have long-term funding commitments (greater than five years) as part of national monitoring programmes. If OSPAR/MSFD requires nations to monitor ocean acidification, long-term funding would need to be put in place.

LAND/INSTITUTE	AREA	PLATFORM/TYPE	PARAMETERS	TIME
Area I – Nordic Seas and Arctic Ocean				
Iceland / MRI	Iceland Sea and Irminger Sea	Single time-series stations	DIC, discreet pCO_2, pH	1983–2010
Norway / UiB and Bjerknes	75° N transect	Research cruises	DIC, TA	2003, 2006, 2008?
Norway / UiB and Bjerknes	OWS M	Monthly profiles	DIC, TA	?-2009
Norway / UiB and Bjerknes	OWS M	Continuous	pCO_2	2005–2009
Norway / UiB and Bjerknes	Nordic Seas	G. O. Sars (research vessel)	Underway pCO_2	?
Sweden / UG	Arctic Ocean	Research cruises	DIC, TA, pH	2005, ?
Germany / AWI?	Nordic Seas (Greenland Sea?)	Research cruises	?	?
Germany / ?	Irregular	Polarstern	Underway pCO_2	
Norway / UiB and Bjerknes	Aarhus – Nuuk	SOO (Nuka Arctica)	Underway pCO_2	?
Iceland / MRI	Icelandic waters?	Bjarni Saemundsson	Underway pCO_2	1995–2010 (irregular)
Iceland / MRI	Icelandic waters?	Bjarni Saemundsson	Research cruises 4 / year	
UK "Ellett Line"	Greenland – UK	Scientific cruise	Hydrography	2008??
Ferry-box line up to Svalbard				
Area I- North Sea				
Norway / UiB and Bjerknes	Bergen – Amsterdam	SOO / weekly	Underway pCO_2	2005–2009
Netherlands / ?				
Norway / UiB and Bjerknes	Aarhus – Nuuk	SOO (Nuca Arctica)	Underway pCO_2	?
Belgium / ULg	Southern Bight of North Sea	Weekly to monthly, Belgica	Underway pCO_2	2000-on going
a	(research vessel)			
UK / NOCS	Stonehaven Single time-series station	?	?	

LAND/INSTITUTE	AREA	PLATFORM/TYPE	PARAMETERS	TIME
Netherlands / NIOZ	Basinwide North Sea	Research cruises	?	2001, 2005, 2008?
UK / NOCS	English Channel	SOO (Pride of Bilbao)	DIC, TA	Since 2005
UK /PML	English Channel (E1, L4)	Weekly (L4) & monthly (E1)	TA/DIC	2008-
UK /PML	English Channel (E1, L4)	Weekly (L4) & monthly (E1)	Underway pCO_2Transects (Plymouth Quest)	
Netherlands / NIOZ	Southern Bight of the North Sea / German Bight	JetSet (53°N; 4° 46'E)Weekly time-series	Underway DIC, TA?	?
Belgium / ULg	Ste Anna (Scheldt estuary)	Fixed station, continuous	pCO_2	2002-on going
Belgium / ULg / NIOO	RV Luctor monitoring (Scheldt estuary)	monthly cruises	pCO_2, TA	2008- on going
Planned activities				
Cefas (UK): Install pCO_2 system on RV Endeavour				
Area III – North-western Atlantic shelf and Celtic Sea				
UK / Cefas	Liverpool Bay	Buoy, DEFRA tests	pCO_2	2010
UK / PML	Holyhead – Dublin,	Prince Madog (research	Underway pCO_2	2006–2009
UK / PML	Irish Sea Coastal Observatory	RV (quasi-monthly)	Underway pCO_2Transects (Prince Madog)	2007-
Ireland / MI	Mace Head Coastal Atmospheric research station	Buoy	pCO_2	2008-
Ireland / MI&NUIG	Irish Shelf (AreaIII) and off-shelf(area V)	RV Celtic Explorer	Underway pCO_2	2009-
Ireland / NUIG&MI	Irish Shelf (AreaIII) and off-shelf(area V)	Research Cruises	TA, DIC	2008 -
Belgium / ULg	Celtic Sea	Research cruises, OMEX-II, CCCC, PEACE	pCO_2, TA, pH	1997–1999, 2002, 2004, 2006–2009
Area IV –				
UK / NOCS?	Portsmouth - Spain	SOO (Pride of BIlbao), 2/week	Underway pCO_2	2005
IEO-Santander / IIM-VIGO	Cantabric Sea and west coast	VACLAN cruises	Underway pCO_2 , pH, TA	2005-
IEO-Gijon	Cantabric Sea	Radiales project	pH, TA	2010-
Belgium / ULg	Iberian upwelling system	Research cruises (OMEX-II)	pCO_2, TA, pH	1997–1999
Planned activities				
IEO-Santander	AGL Buoy(49°N; 16.5°W)	Mooring	pH	2011
IEO-Santander	Cantabric Sea	Radiales project	pH,	2011

Land/institute	Area	Platform/type	Parameters	Time
IEO-Vigo	Cantabric Sea	Radiales project	Underway pCO_2 pH	2010
Area V – Open Atlantic Ocean				
Germany / ?	Liverpool - Halifax	SOO (A. Companion)	2 per 5 weeks Underway pCO_2	2005
USA / France	Charleston – Reykjavik	SOO (Skogafoss) 12 / year	Underway pCO_2	2005
UK / UEA	Portsmouth (UK) Windward Islands -	SOO (Santa Lucia/Santa Maria)	Underway pCO_2	/monthly 2002-
France / Lefevre	France – French Guiana	SOO (MN Colibri) ~6/year	Underway pCO_2	2006-
France / Lefevre	France – Brazil	SOO (Monte Olivia) ~6/year	Underway pCO_2	2007-
Spain / ULPGC	Goteburg – Durban	SOO (Quima)	Underway pCO_2	2005-
UK / NOCS	Porcupine Abyssal Plain (49°N; 16.5°W)	Mooring	pCO_2	?-
Spain	OVIDE, Iberian Peninsula-	Research cruise	Underway pCO_2,pH,TA	2002–2010
Spain	Spain-Antartic	SOO	Underway pCO_2	2000–2009
Spain	GIFT (35.862°N, 5.974°W)	Time series station	water column pH, TA	2005-
France / ?	MAREL (48°22'N; 4°33'W)	Mooring	pCO_2 2003-	
UK / NOC/UEA	26° N line	?	?	?
Ireland / NUIG&MI	off-Shelf Rockall Trough	Research Cruises	TA, DIC	2008-
Planned activities				
Spain / ?	OVIDE, Iberian Peninsula/ Greenland -	Research cruise	?	2010
Baltic Sea				
Sweden / SMHI	Swedish waters	Monitoring cruises?	TA, pH	?
Germany / IOW/Schneider		Helsinki – Lübeck	SOO	Underway pCO_2
Germany / IFM-GEOMAR	Boknis Eck (54.52°.N 10.03° E)	Time-series station	?	?
Germany / ?	?	Research cruises	??	
Estonia/Lipps	Helsinki – Tallinn	SOO	Underway pCO_2	2010
Planned activities				
Sweden / SMHI	Kemi - Gothenburg	Baltoc SOO	Underway pCO_2	2010

8 Summary of spatial and temporal information for the OSPAR area

To avoid aliasing the interpretation of results from long-term monitoring, the programme has to be designed to take into account shorter term variability of the system. The current state of knowledge of the variability of the system is summarized here.

Figure 1 provides information on cross-system variability and the range of spatial and temporal variability of seawater carbonate chemistry variables. In general, the dynamic range of pH tracks that of pCO_2. The dynamic range of pCO_2 and of pH is more intense in estuarine environments and decreases towards marginal seas, showing intermediate dynamic range in nearshore and coastal upwelling systems. Estuaries show the largest dynamic range in TA, followed by nearshore ecosystems (due to the influence of run-off) and then marginal seas (related to strong gradients in the Arctic Ocean also due to the influence from run-off).

Table 1 summarizes available information on the temporal variability of seawater carbonate chemistry variables in the OSPAR regions from daily to interannual timescales. The daily variability due to the night-day cycle of biological activity (photosynthesis and respiration) is relatively uniform across the OSPAR regions, and 2 to 10 times lower than the seasonal amplitude. Note that these studies were carried out during the most productive periods of the year, typically in spring. During other less productive seasons (undocumented to our best knowledge) the daily variability is expected to be lower or even below detection levels. Pelagic calcification seems to be at cellular level coupled to photosynthesis, hence, it is also expected to follow a day-night cycle. Based on field studies (e.g. Robertson *et al.*, 1994; Harlay *et al.*, 2010; Suykens *et al.*, 2010), the maximal drawdown of TA during blooms of pelagic calcifiers is ~30 μmol kg^{-1}, for a characteristic time-scale typically of 15 days (roughly equating at a drawdown of TA of ~2 μmol kg^{-1} per day). Thus, the impact of pelagic calcification at daily scale on seawater carbonate chemistry is expected to be close to or below detection limits. In regions of strong horizontal salinity gradients (nearshore coastal environments such as Irish Sea, English Channel and Southern Bight of the North Sea), the tidal displacement of water masses leads to sub-daily variability of seawater chemistry that is equivalent or higher than the daily variability due to the day-night cycle of biological activity. For instance tidal variations of TA and pCO_2 of, respectively, 50 μmol kg^{-1} and 50 μatm have been reported in the Southern Bight of the North Sea (Borges and Frankignoulle, 1999).

Seasonal variations of seawater carbonate variables are mainly related to biological activity (organic carbon production and degradation, $CaCO_3$ production and dissolution) and to physical structure of the water column (mixing and stratification), and to the thermodynamic effect of seasonal temperature changes for pCO_2 and pH. The amplitude of the seasonal variations of seawater carbonate variables is strongest in OSPAR region II (North Sea), and more or less equivalent in the other four OSPAR regions (Table 1).

Inter-annual variability of seawater carbonate variables is strongest in OSPAR region II (North Sea), and roughly equivalent in OSPAR regions III, IV and V and lowest in OSPAR region I (Table 1). Except for OSPAR region I, interannual variations are equivalent to the amplitude of seasonal variations. Table 1 shows the maximum interannual variations that are typically observed during the most productive season (spring). Inter-annual variability of seawater carbonate variables is usually lower

during the other periods of the year (e.g. Schiettecatte *et al.*, 2007; Omar *et al.*, 2010 for the North Sea). Inter-annual variability of the seawater carbonate variables is related to variable river inputs in nearshore ecosystems (Borges *et al.*, 2008a), to biological activity in nearshore and offshore ecosystems (Borges *et al.*, 2008a; Omar *et al.*, 2010), to vertical mixing (Borges *et al.*, 2008a,b; Dumousseaud *et al.*, 2009), and to changes in temperature (Dumousseaud *et al.*, 2009; Omar *et al.*, 2010). These drivers of interannual variations interact; for instance, milder and warmer years will be characterized by lower winter-time mixing that will lead to a lower seasonal replenishment of nutrients and lower primary production, but also a lower vertical input of DIC (e.g. Borges *et al.*, 2008b).

Spatial gradients of seawater carbonate variables can be related to the heterogeneity of water masses and will to some extent track the spatial gradients of salinity or of temperature. Spatial gradients of seawater carbonate variables can also be related to the more or less marked patchiness of biological activity. The spatial gradients of seawater carbonate variables are strongest in the Iberian upwelling region of OSPAR region IV, followed by OSPAR region II (Table 2). Note that we reported in Table 2 the large-scale spatial gradients (at basin scale), but mesoscale spatial gradients can be much more intense such as across frontal structures (Borges and Frankignoulle, 2003) or across river plumes (Borges and Frankignoulle, 1999).

Long-term changes of pH are poorly documented and most available information on long-term changes of seawater carbonate variables is based on the analysis of seawater pCO_2 data. In all OSPAR regions, the reported rate of increase of pCO_2 in seawater is equivalent or higher than the increase of atmospheric CO_2 (Table 3). The fact that pCO_2 could be increasing faster in surface waters than in the atmosphere has been attributed to changes in circulation both in terms of vertical mixing (Corbière *et al.*, 2007) and in terms of horizontal distribution of water masses (Thomas *et al.*, 2008), or to the decrease of buffering capacity of seawater (Thomas *et al.*, 2007). In nearshore regions influenced by river inputs, such as the Southern Bight of the North Sea, the decadal changes of seawater carbonate variables due to changes in nutrient inputs have been evaluated by model simulations to be more intense than expected from the response to ocean acidification (Gypens *et al.*, 2009; Borges and Gypens, 2010). The effect of eutrophication on carbon cycling could counter the effect of ocean acidification on the carbonate chemistry of surface waters. But changes in river nutrient delivery due to watershed management could also lead to stronger changes in carbonate chemistry than ocean acidification. Whether antagonistic or synergistic, the response of carbonate chemistry to changes of nutrient delivery to the coastal zone (increase or decrease, respectively) could be stronger than ocean acidification (Borges and Gypens, 2010).

Note that the long-term yearly rates of change of pCO_2 and pH are close or below the analytical detection level. Also, the long-term yearly rates of change of pCO_2 and pH are between 3 and 10 times lower than the typical interannual variability of these quantities in the OSPAR regions (Table 1). This implies that to detect long-term changes of seawater carbonate variables, a sustained monitoring of more than 10 years is required to obtain a signal that is analytically significant and discern the long-term trend from natural interannual variability.
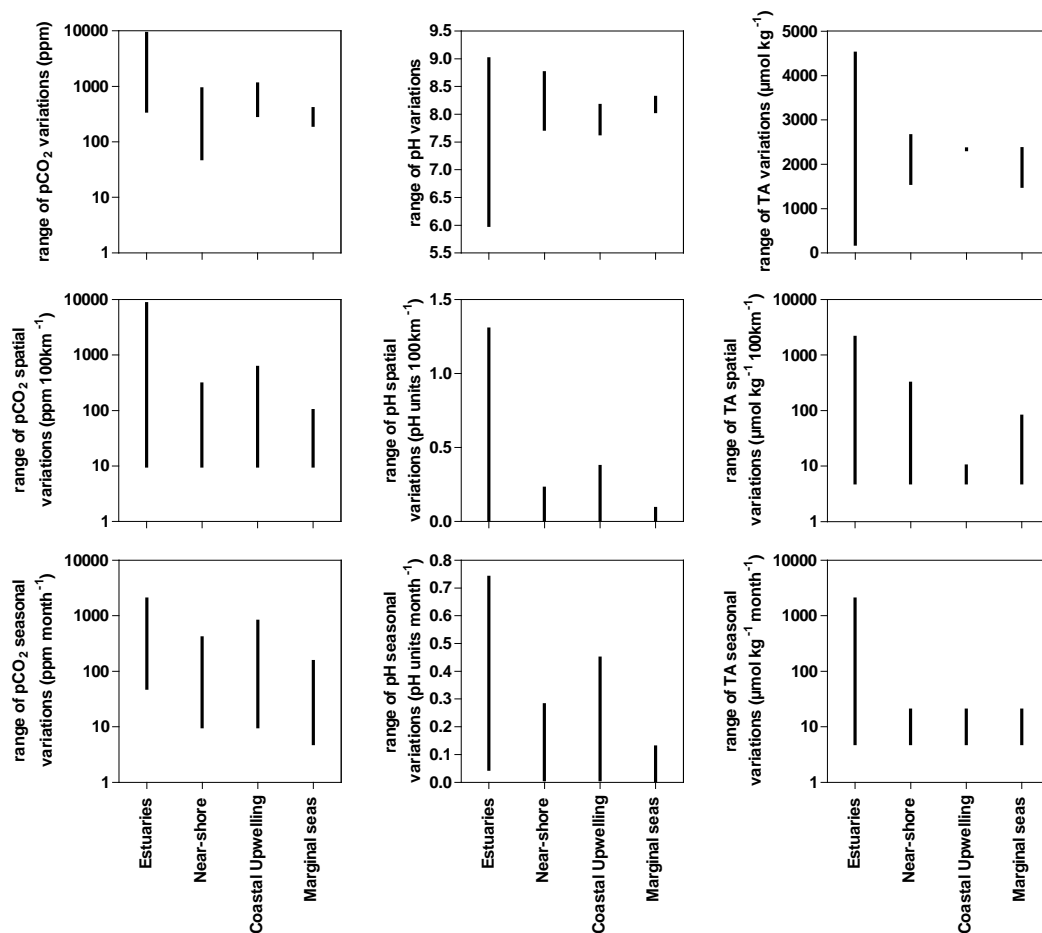
Figures and Tables

Figure 1. Range of spatio-temporal variability across different coastal environments of the partial pressure of CO₂ (pCO₂), pH and total alkalinity (TA). Adapted from Borges *et al*. (2009).

Table 1. Amplitude of daily and seasonal variations and interannual variability of the partial pressure of CO_2 (pCO_2), total alkalinity (TA), pH and dissolved inorganic carbon (DIC) in the OSPAR regions. The variations of pH and DIC were computed from pCO_2 and TA and were broken down into changes due to pCO_2 (ΔpCO_2) and due to TA (ΔTA).

OSPAR REGION	PCO2 (µATM)	TA (µMOL KG-1)	PH		DIC (µMOL KG-1)	
			ΔpCO2	ΔTA	ΔpCO2	ΔTA
Amplitude of daily variations (maximum, i.e. most productive period)						
I	20 [a]	~0	0.020	~0	10	~0
II	20 [b]	~0	0.020	~0	10	~0
III	15 [b]	~0	0.015	~0	7	~0
IV	15 [b]	~0	0.015	~0	7	~0
V	20 [a,c]	~0	0.020	~0	10	~0
Amplitude of seasonal variations						
I	45 [d]	20 [h]	0.047	0.003	23	17
II	220 [e]	60 [i]	0.309	0.010	175	52
III	70 [b]	50 [b,j]	0.075	0.008	37	43
IV	30 [f]	20 [k]	0.031	0.003	15	17
V	60 [g]	20 [h]	0.064	0.003	31	17
Inter-annual variability						
I	5 [l]	?	0.005	?	2	?
II	150 [e]	?	0.183	?	96	?
III	50 [k]	20 [b]	0.052	0.003	25	17
IV	50 [k]	10 [k]	0.052	0.002	25	9
V	20 [m]	?	0.020	?	10	?

[a] Robertson *et al.* (1993) ; [b] Frankignoulle and Borges (2001) ; [c] Borges and Frankignoulle (2001) ; [d] Olsen *et al.* (2008); [e] Omar *et al.* (2010) ; [f] Borges and Frankignoulle (2002); [g] Schuster and Watson (2007); [h] Robertson *et al.* (1994); [i] Thomas *et al.* (2009); [j] Harlay *et al.* (2010); [k] Dumousseaud *et al.* (2009); [l] Naka-oka *et al.* (2006); [m] Santana-Casiano *et al.* (2007).

Table 2. Typical spatial gradients at basin scale (per 100 km) of the partial pressure of CO_2 (pCO_2), total alkalinity (TA), pH and dissolved inorganic carbon (DIC) in the OSPAR regions. The variations of pH and DIC were computed from pCO_2 and TA and were broken down into changes due to pCO_2 (ΔpCO_2) and due to TA (ΔTA).

OSPAR REGION	PCO2 (µATM 100KM-1)	TA (µMOL KG-1 100KM-1)	PH (PH UNITS 100 KM-1)		DIC (µMOL KG-1 100KM-1)	
			ΔpCO2	ΔTA	ΔpCO2	ΔTA
I	2 [a]	8 [g]	0.002	0.001	1	7
II	20 [b,c,d]	20 [h]	0.020	0.003	10	17
III	10 [b]	5 [b,i]	0.010	0.001	5	4
IV	10 [b] to 50 [e]	5 [e,i]	0.010 to 0.052	0.001	5 to 26	4
V	2 [f]	5 [f]	0.002	0.001	1	4

[a] Olsen *et al.* (2008); [b] Frankignoulle and Borges (2001); [c] Thomas *et al.* (2004); [d] Omar *et al.* (2010); [e] Borges and Frankignoulle (2002); [f] Schuster and Watson (2007); [g] based on salinity gradients from Olsen *et al.* (2008) ; [h] Thomas *et al.* (2009) ; [i] Dumousseaud *et al.* (2009).

Table 3. Long-term changes in surface waters of the partial pressure of CO_2 (pCO_2) and pH in the OSPAR regions. The changes of pH were computed from those of pCO_2 assuming a constant total alkalinity.

OSPAR REGION	PCO2 (µATM YR−1)	PH (PH UNITS YR−1)
I	1.5 to 3.0 [a]	-0.0015 to -0.0030
I	2.1±0.2 [a]	-0.0024 ± 0.002
II	4.4 [a]	-0.0044
III	3.2 [a]	-0.0032
IV	3.2 [a]	-0.0032
V	1.9 [a] to 4.9 [a]	-0.0019 to -0.0049

[a] Omar and Olsen (2006); [b] Thomas *et al.* (2007); [c] Schuster *et al.* (2009), [d] Olafsson *et al.* (2009).

9   Inconsistencies in the calculation of carbonate system variables when the system is over determined and concerns over the applicability of calculation schemes in estuarine waters

9.1   Validity of calculations in estuarine waters

MCWG 2010 discussed concerns about how universal calculation procedures such as the commonly used CO2sys program (Lewis and Wallace, 1998; Pierrot *et al.*, 2006) could be used in all waters of interest to the group. The limitations arise due the degree to which the various dissociation constants for carbonic acid have been measured over the whole range of salinities of interest. The choice of dissociation constants of carbonic acid used in the calculations needs to be appropriate. The Mehrbach *et al.* (1973) constants as refitted by Dickson and Millero (1987) are assumed to be the better choice to use in calculations based on ocean waters (Wanninkhof *et al.*, 1999; Lee *et al.*, 2000). However they are only valid down to a salinity of 15, so cannot be used reliably in estuarine waters. Recently Millero *et al.* (2006) have published dissociation constants of carbonic acid in seawater as a function of salinity and temperature which extend over the whole salinity range. They have been included in version 14 of CO2sys excel version (http://www.ecy.wa.gov/programs/eap/models.html) Software to provide an alternative to the commonly used CO2sys software (Lewis and Wallace, 1998) has and is being developed. Seacarb (http://CRAN.R project.org/package=seacarb) may be considered as an alternative. Among its benefits, its latest version (2.3.3) includes the most up to date dissociation constants for K_1 and K_2 of Millero *et al.* (2006) and Millero (2010).

From the purely analytical point of view methods described by Dickson *et al.* (2007), are valid in estuaries for TA and DIC if used appropriately. Where pCO2, is measured directly using an equilibrator based systems problems have been identified in turbid estuarine waters, so that an appropriately designed equilibrator needs to be used (e.g. Frankignoulle *et al.*, 2001).

The major uncertainty arises with the choice of scales for pH and related buffers in estuaries. Frankignoulle and Borges (2001) have shown consistent data for pH can be achieved over the full range of estuarine salinities is sampled if the data are referenced to the NBS scale. The limitation of using TRIS buffer values and the Total Scale

is that values for TRIS buffers have only been determined for salinities > 5 (DelValls and Dickson, 1998). The data the AMP buffer value is only established for salinity 35. Hence, the Nernstian behaviour of pH electrodes can only be checked at this salinity.

9.2   Inconsistencies in the calculation of carbonate system variables

A discussion paper documents some of the concerns expressed at the meeting that in certain circumstances calculations of pH and pCO_2 from measurements of TA and DIC were not always consistent with direct measurements of pH and pCO_2. Hoppe *et al.* (2010) in their paper addressing carbonate chemistry in perturbation studies report in their abstract that: "Seawater carbonate chemistry is typically calculated from two measured parameters. Depending on the choice of these input parameters, discrepancies in calculated pCO_2 have been recognized by marine chemists, but the significance of this phenomenon for CO_2 perturbation experiments has so far not been determined. To mimic different pCO_2 scenarios, two common perturbation methods for seawater carbonate chemistry (changing either DIC or TA) were applied using state-of-the-art protocols and equipment. The carbonate system was over-constrained by measuring DIC, TA, pH, and pCO_2. Calculated pCO_2 matched measured pCO_2 if pH and TA or pH and DIC were chosen as input parameters, whereas pCO_2 calculated from TA and DIC was considerably lower than measured values. This has important implications for CO_2 perturbation experiments. First, calculated pCO_2 values may not be comparable if different input parameters were used. Second, responses of organisms to acidification may be overestimated when using TA and DIC for calculations. This is especially troublesome for experiments with calcifiers, as carbonate ion concentration and thus calcite or aragonite saturation state are overestimated. We suggest refraining from measuring TA and DIC only and rather include pH as input parameter for carbonate chemistry calculations."

Further consideration needs to be given to this issue. This needs to look at both: (i) Other potential sources of titratable alkalinity in samples not currently taken into account. For example Kim and Lee (2009) have pointed out that dissolved organic matter can potentially make a significant contribution to alkalinity. (ii) That the most appropriate dissociation constants are being used. Gattuso *et al.* (2010) addresses carbonate chemistry in perturbation studies in detail.

10   Related projects

Design of a long-term monitoring programme needs to take into account what can be learned from previous work and current and planned activities in the OSPAR area. It further needs to take into account relevant international research and observations programmes.

Key going projects in Europe are the **EU-EPOCA** project, and German **BIOACID** project and **UK-NERC Ocean Acidification** project.

On the observational side an important synergy will be linkage to the ICOS project looking at CO_2 fluxes.

For hydrographic work in deep waters much can be learned about existing data and its quality from the **CARINA** Project. The CARINA (CARbon dioxide IN the Atlantic Ocean) data synthesis project is an international collaborative effort of the EU IP CARBOOCEAN, and US partners. It has produced a merged internally consistent dataset of open ocean subsurface measurements for biogeochemical investigations, in particular, studies involving the carbon system. The original focus area was the

North Atlantic Ocean. The CARINA database includes data from 188 cruises. The salinity, oxygen, nutrient, inorganic carbon system and CFC data have been subjected to extensive quality control and adjustments have been made when necessary. The internally consistent data are available as three data products, one each for the Arctic Mediterranean Seas, the Atlantic and the Southern Oceans (CARINA Data Products). In addition, all of the individual cruise data files have been made available in WOCE exchange format in a single location (Cruise Summary Table) along with metadata and references. The CARINA effort is described in the CARINA special issue of Earth System Science Data (ESSD) Journal.

Other relevant former, ongoing and planned projects include:-

IMBER - Integrated Marine Biogeochemistry and Ecosystem Research Project; www.imber.info/

SOLAS - Surface ocean lower atmosphere study; www.solas-int.org

Note an IMBER/SOLAS Ocean Acidification Working Group (http://www.imber.info/C_WG_SubGroup3.html) which has the following tasks:

1- Coordinate international research efforts in ocean acidification;
2- Undertake synthesis activities in ocean acidification at the international level.

ANIMATE Atlantic Network of Interdisciplinary Moorings and Time-series for Europe; www.noc.soton.ac.uk/animate/

IOCCP International Ocean Carbon Coordination Project; www.ioc.unesco.org/ioccp/

OCB US Ocean Carbon and Biogeochemistry; www.us-ocb.org/

CARBON-OPS UK underway pCO_2 observations from Research Vessels; www.bodc.ac.uk/carbon-ops

CARBOOCEAN IP - Marine carbon sources and sinks assessment; www.carboocean.org

EPOCA European Project on Ocean Acidification; www.epoca-project.eu

BOOM Biodiversity of Open Ocean Microcalcifiers; www.sb-roscoff.fr/BOOM/

PEECE Pelagic Ecosystem CO_2 Enrichment Study; www.peece.ifm-geomar.de

MEECE Marine Ecosystem Evolution in a Changing Environment; www.meece.eu/

PEACE Role of Pelagic Calcification and Export of Carbonate Production in Climate Change; www.co2.ulg.ac.be/peace/

BIOACID Biological Impacts of Ocean Acidification; www.bioacid.ifm-geomar.de/

SOPRAN Surface Ocean Processes in the Anthropocene; www.sopran.pangaea.de/

CLIVAR Climate variability and predictability; https://webmail.ices.dk/Exchange/michala/emcgovern/Local Set-tings/emcgovern/Application Data/emcgovern/Local Set-

tings/Documents and Settings/menchu.ST/Configuración lo-cal/Temp/www.clivar.orgwww.clivar.org

SMHI-UGOT   Ocean acidification project 2010–2012; www.smhi.se

FERRYBOX   www.ferrybox.org

Acknowledgements

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Annex 9: Technical annex for dioxins/furans and dioxin-like PCBs in biota (revised version)

1. Introduction

Polychlorinated dibenzo-*p*-dioxins and polychlorinated dibenzofurans (dioxins/furans - PCDD/Fs) are ubiquitous in the environment, primarily as unintentional by-products of combustion and industrial processes. They enter the aquatic environment via several routes, including atmospheric deposition. Being strongly hydrophobic compounds, sediments are the eventual sink in the aquatic environment, providing a source of potential exposure to aquatic organisms (Hurst *et al.*, 2004). Generally highly resistant to metabolism, PCDD/Fs bioaccumulate and biomagnify and have reached high concentrations in e.g. fish from the Baltic Sea, which resulted in recommendations to restrict the use of those fish for human consumption (Verta *et al.*, 2007). This guideline only addresses the 17 tetra- through octa-chlorinated 2,3,7,8-substituted dioxin and furan congeners, and the non- and mono-*ortho* substituted polychlorinated biphenyls (PCBs), which are able to exhibit similar effects as the 2,3,7,8-substituted dioxins and furans. The general chemical structures of PCDD/Fs and PCB congeners are given in Figure 1.

In this guideline, the term "dioxin-like PCBs" (dl-PCBs) is used for the non-*ortho* and mono-*ortho* PCB congeners listed in Table 1. The coplanar structure of non-*ortho* substituted PCB congeners allows a configuration similar to that of PCDD/Fs. Mono-*ortho* substituted PCBs may take a steric position close to coplanarity and are consequently less toxic than non-*ortho* PCBs. Nevertheless, they have been considered due to their relatively high concentrations compared to those of non-*ortho* PCBs or dioxins/furans (Daelemans *et al.*, 1992). The exposure to dl-PCBs is mainly via the food chain, as the compounds are highly lipophilic and bioaccumulate and biomagnify in lipid-rich tissue (e.g. Dyke *et al.*, 2003).

Chlorinated dioxins/furans and dl-PCBs have been shown to produce various toxic responses, including immunotoxicity, developmental and reproductive effects, neurotoxicity and carcinogenesis (OSPAR, 2005). The initial mechanism of toxicity is via the aryl hydrocarbon receptor (AhR), leading to changes in gene expression, cell growth and cell differentiation (Nebert *et al.*, 1993; Hurst *et al.*, 2004). Due to their persistence, high toxicity, bioaccumulation potential and ability for long-range transport, they are controlled under the Stockholm Convention for Persistent Organic Pollutants (POPs). Their spatial and temporal monitoring in the aquatic environment is important to evaluate the risk to wildlife and human health (Hurst *et al.*, 2004). Due to the low concentrations at which adverse effects can be observed, the analytical methodology for the analysis of PCDD/Fs and dl-PCBs differs from those for other organochlorine compounds, as described in this guideline.
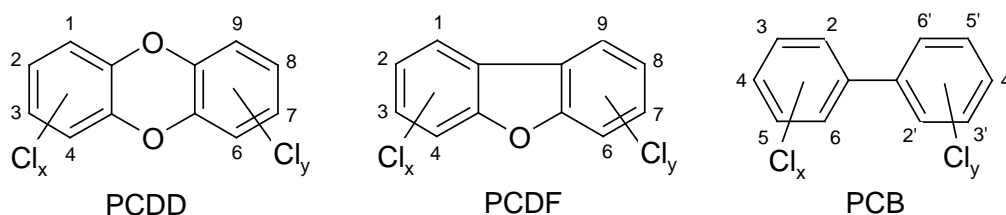
Figure 1. General formula of PCDDs, PCDFs and PCBs. The possible number of chlorine atoms results in 75 PCDD congeners, 135 PCDF congeners (x=1–4, y=0–4), and 209 PCB congeners (x=1–5, y=0–5).

2. Analytes and Toxicity Equivalent Factors (TEFs)

Environmental monitoring should include the 17 tetra- through octa-chlorinated 2,3,7,8-dibenzo-p-dioxins (CDDs) and dibenzofurans (CDFs) and the dl-PCBs listed in Table 1.

In the context of food and feed analysis and compliance checks with maximum residue limits, the concept of TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin) Toxicity Equivalency Factors (TEFs) is commonly used, to account for mixtures of several PCDD/Fs and other compounds with dioxin-like activity usually present in these samples. Each congener has been assigned a TEF relative to that of the most toxic dioxin congener, 2,3,7,8-TCDD that was given a TEF of 1.0. The concentrations of the individual congeners are multiplied with their respective TEFs, and the sum of this gives the total concentration of dioxin-like compounds, expressed in TCDD Equivalents (TEQs). Thus, concentrations of mixtures can be expressed in terms of their dioxin-like activity in TEQs, relative to the most potent 2,3,7,8-TCDD. Two parallel TEF systems are currently in use: TEFs established by the World Health Organization (WHO-TEF/TEQ) and TEFs developed by NATO/CCMS (International TEFs or I-TEFs/TEQ). The use of I-TEFs, however, is decreasing. The WHO-TEF-system is reviewed every five years, and Table 1 presents the most recent values, as of 2006 (Van den Berg *et al.*, 2006).

According to OSPAR (2005), the scientific relevance of using TEQs to express results is greater for human exposure than for evaluation of pollution sources and emissions, for which information on congener patterns can be of more importance. Furthermore, the system assumes additive effects of the individual congeners, while both synergistic and antagonistic effects have also been reported (OSPAR, 2005). It is therefore recommended for environmental monitoring to report concentrations of individual PCDD/Fs and dl-PCB congeners in biota as absolute concentrations, i.e. pg/g wet weight (ww), with additional information on dry matter and lipid content (see chapter "Data reporting"). The TEF concept can be applied in a subsequent risk assessment, if appropriate.

As part of the TEQ approach, there are different ways of handling results below limits of quantification (LoQ):

- The concept of *upper bound* requires using the limit of quantification for the contribution of each non-quantified congener to the TEQ.

- The concept of *lower bound* requires using zero for the contribution of each non-quantified congener to the TEQ.

- The concept of *medium bound* requires using half of the limit of quantification calculating the contribution of each non-quantified congener to the TEQ.

As mentioned above, results of environmental monitoring should preferably be reported for individual congeners, in absolute concentrations. However, OSPAR (2005) mentioned the ongoing food analysis programmes which might be complementary to environmental monitoring. In this context, information on the handling of concentrations below LoQ will be important. Thus, results expressed as TEQ values should be reported as both upper bound and lower bound values (at least, indication of which calculation mode was used should be given). COMMISSION DIRECTIVE 2002/69/EC specifies that, for samples containing 1 pg WHO TEQ/g fat, the difference between upper bound and lower bound level should not exceed 20%. For lower contamination levels, this difference may be in the range of 25 to 40%.

Table 1. Chlorinated dibenzo-p-dioxins (CDDs), chlorinated dibenzofurans (CDFs) and dl-PCBs with their Toxicity Equivalent Factors (TEFs) according to the systems developed by the World Health Organization (WHO2005-TEF, Van den Berg *et al.*, 2006) and NATO/CCMS (I-TEF).

HOMOLOGUE GROUP	CONGENER	I–TEF	WHO2005–TEF	IUPAC NO.
PCDDs				
TCDD	2,3,7,8	1	1	
PeCDD	1,2,3,7,8	0.5	1	
HxCDD	1,2,3,4,7,8	0.1	0.1	
	1,2,3,6,7,8	0.1	0.1	
	1,2,3,7,8,9	0.1	0.1	
HpCDD	1,2,3,4,6,7,8	0.01	0.01	
OCDD	1,2,3,4,6,7,8,9	0.001	0.0003	
PCDFs				
TCDF	2,3,7,8	0.1	0.1	
PeCDF	1,2,3,7,8	0.05	0.03	
	2,3,4,7,8	0.5	0.3	
HxCDF	1,2,3,4,7,8	0.1	0.1	
	1,2,3,6,7,8	0.1	0.1	
	1,2,3,7,8,9	0.1	0.1	
	2,3,4,6,7,8	0.1	0.1	
HpCDF	1,2,3,4,6,7,8	0.01	0.01	
	1,2,3,4,7,8,9	0.01	0.01	
OCDF	1,2,3,4,6,7,8,9	0.001	0.0003	
NON–ORTHO PCBs				
TeCB	3,3',4,4'		0.0001	77
TeCB	3,4,4',5		0.0003	81
PeCB	3,3',4,4',5		0.1	126
HxCB	3,3',4,4',5,5'		0.03	169
MONO–ORTHO PCBs				
PeCB	2,3,3',4,4'		0.00003	105
	2,3,4,4',5		0.00003	114
	2,3',4,4',5		0.00003	118
	2',3,4,4',5		0.00003	123

HxCB	2,3,3',4,4',5	0.00003	156
	2,3,3',4,4',5	0.00003	157
	2,3',4,4',5,5'	0.00003	167
HpCB	2,3,3',4,4',5,5'	0.00003	189

3. Biota samples

OSPAR (2005) presented a monitoring strategy for PCDD/Fs, which identified biota as one of the important matrices for environmental monitoring (the other one being marine sediments). Aquatic organisms can accumulate hydrophobic compounds like dioxins/furans and reach concentrations considerably above those of the surrounding waters. The ratio between the concentration in biota and in the water is the bioconcentration factor (BCF), which is between 2000 and 9000 for PCDD/Fs (OSPAR, 2005). As the BCF varies with species and compound, it is important to design a sampling programme which minimizes confounding factors, i.e. to choose the same species, sampling area and sampling period.

The species selected for monitoring should fulfil certain requirements:

- Reflect concentration changes in the sampling area, i.e. ensure a link between exposure and concentration in the organisms.
- Accumulate compounds without showing adverse effects.
- Representative of and abundant in the area (to ensure sufficient sample material for analysis).
- Relatively easy to handle.

Analogous to the monitoring of other organohalogen compounds, mussels and fish are suitable and commonly used for monitoring of PCDD/Fs and dl-PCBs (OSPAR, 2005). Highest dioxin concentrations are found in fish liver and muscle tissue of fatty fish such as herring and salmon. National food agencies often analyse PCDD/Fs and dl-PCBs in commercial fish and fish products, in order to monitor compliance with EU limit values. While different approaches will be necessary in environmental analyses, OSPAR (2005) recommends the monitoring of fish and shellfish as part of the monitoring strategy for dioxins.

In general, the same recommendations are valid as described for other organochlorine compounds, i.e. in the OSPAR JAMP guideline on organic contaminants (OCs) in biota, which also contains details on sample dissection and homogenization. It should be pointed out, however, that the risk of sample contamination is considerably higher, given the extremely low concentrations of PCDD/Fs and dl-PCBs in most biota samples. The staff collecting and handling the samples should be well-trained and properly instructed in how to avoid contamination.

For mussel samples, it is important to remove any sediment particles from their intestinal system, by depuration in a glass aquarium with filtered water from the sampling location for approximately 24 hours. Mussel samples must not be frozen prior to dissection, but should be transported at temperatures between 5 and 15°C, suitable for the area of origin, in a clean container. After dissection, all samples should be stored in the dark at < -20°C prior to analysis. Under these conditions, long-term storage of tissue samples is possible (De Boer and Smedes, 1997). More details on the practical aspects of sample handling and preparation are given in the OSPAR JAMP guideline on OCs in biota.

4. Analytical methods

An example of a suitable method for the analysis of biota samples is given in Figure 2.
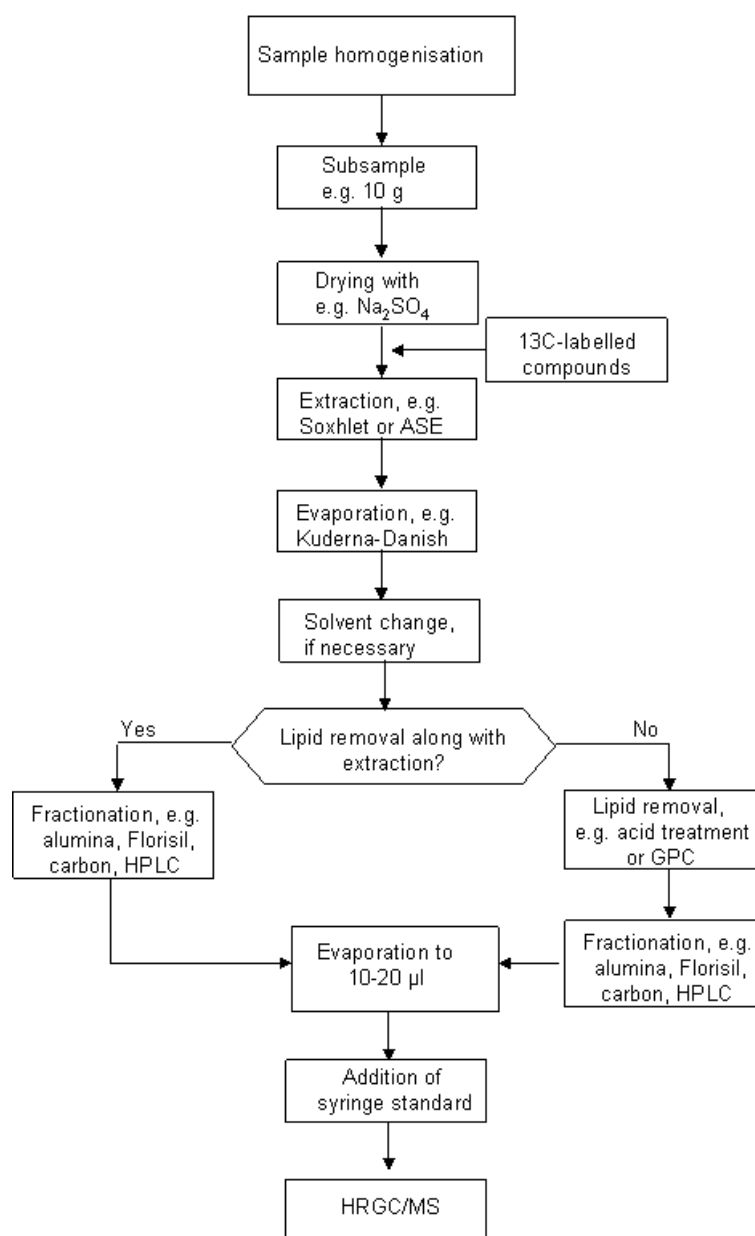

Figure 2. Analytical method recommended for analysis of PCDD/Fs and dl-PCBs in biota samples within environmental monitoring.

4.1 Preparatory steps

It is essential to avoid contamination during all analytical steps. Where possible, reagents should be of high purity or cleaned by extraction or solvent rinse. All solvents used must be checked for presence of residues of target or interfering compounds (e.g. polychlorinated diphenyl ethers). The purity of standards should be checked.

Reusable glassware should be rinsed with solvent, disassembled, washed with a detergent solution and further rinsed with ultrapure grade water and solvent. Baking glassware is common practice as part of the cleaning process, but the formation of active sites on the glass surface that may adsorb the target compounds has been reported (USEPA, 1994).

The preparation of stock solutions and standards can follow the guidelines developed for OCs in biota. However, care has to be taken to monitor and to avoid contamination. Furthermore, the high toxicity of the compounds might require a particularly careful handling; see comments under "Safety". Commercially available diluted stock solutions can be used to reduce safety issues. As valid for the entire analytical method, only trained personnel should perform these steps.

PCDD/Fs and dl-PCBs are normally determined by isotope dilution, using high resolution gas chromatography and high resolution mass spectrometry (HRGC/HRMS). ^{13}C-labelled standards of all congeners to be analysed are added prior to extraction of the samples. These internal standards are used to quantify the native PCDD/Fs and PCBs and to check the method performance in each sample (recovery surrogates). Table 2 provides a list of all $^{13}C_{12}$ labelled congeners available for use as internal standards while Table 3 provides the minimum number of internal standards to be used for the quantification of PCDD/Fs congeners.

Table 2. $^{13}C_{12}$ labelled congeners that can be used as the internal standards.

PCDD/F CONGENERS	PCB CONGENERS
2,3,7,8-$^{13}C_{12}$-TCDD	$^{13}C_{12}$-CB-77
1,2,3,7,8-$^{13}C_{12}$-PeCDD	$^{13}C_{12}$-CB-81
1,2,3,4,7,8-$^{13}C_{12}$-HxCDD	$^{13}C_{12}$-CB-126
1,2,3,6,7,8-$^{13}C_{12}$-HxCDD	$^{13}C_{12}$-CB-169
1,2,3,7,8,9-$^{13}C_{12}$-HxCDD	
1,2,3,4,6,7,8-$^{13}C_{12}$-HpCDD	$^{13}C_{12}$-CB-105
$^{13}C_{12}$-OCDD	$^{13}C_{12}$-CB-114
	$^{13}C_{12}$-CB-118
2,3,7,8-$^{13}C_{12}$-TCDF	$^{13}C_{12}$-CB-123
1,2,3,7,8-$^{13}C_{12}$-PeCDF	$^{13}C_{12}$-CB-156
2,3,4,7,8-$^{13}C_{12}$-PeCDF	$^{13}C_{12}$-CB-157
1,2,3,4,7,8-$^{13}C_{12}$-HxCDF	$^{13}C_{12}$-CB-167
1,2,3,6,7,8-$^{13}C_{12}$-HxCDF	$^{13}C_{12}$-CB-189
2,3,4,6,7,8-$^{13}C_{12}$-HxCDF	
1,2,3,7,8,9-$^{13}C_{12}$-HxCDF	
1,2,3,4,6,7,8-$^{13}C_{12}$-HpCDF	
1,2,3,4,7,8,9-$^{13}C_{12}$-HpCDF	
$^{13}C_{12}$-OCDF	

Table 3. Minimum number of internal standards to be used for calibration of PCDD and PCDF homologue groups.

SUBSTANCE	PCDD–HOMOLOGUES		PCDF–HOMOLOGUES	
	NATIVE	$^{13}C_{12}$–LABELLED	NATIVE	$^{13}C_{12}$–LABELLED
Tetrachloro homologues	2,3,7,8	2,3,7,8	2,3,7,8	2,3,7,8
Pentachloro homologues	1,2,3,7,8	1,2,3,7,8	2,3,4,7,8 1,2,3,7,8	2,3,4,7,8 1,2,3,7,8
Hexachloro homologues	1,2,3,4,7,8 1,2,3,6,7,8 1,2,3,7,8,9	1,2,3,7,8,9	1,2,3,4,7,8 1,2,3,6,7,8 1,2,3,7,8,9 2,3,4,6,7,8	2,3,4,6,7,8
Heptachloro homologues	1,2,3,4,6,7,8	1,2,3,4,6,7,8	1,2,3,4,6,7,8 1,2,3,4,7,8,9	1,2,3,4,6,7,8

4.2 Lipid determination

The total lipid content should be determined in all biota samples, using the method of Bligh and Dyer (1959) as modified by Hanson and Olley (1963) or an equivalent method such as Smedes (1999). For normalization purposes, the total lipid content is preferred to the extractable lipid content (De Boer, 1988).

4.3 Extraction

Soxhlet extraction is commonly used for biota samples. Immediately prior to use, the Soxhlet apparatus should be pre-extracted with e.g. dichloromethane:hexane (1:1) for approximately 3 hours (USEPA, 1994). According to the USEPA method, an adequate amount of tissue (e.g. 10 g of wet tissue) is spiked with the labelled compounds and mixed with sodium sulphate. The sample is allowed to dry for 12–24 hours and should be remixed prior to transfer to a glass Soxhlet thimble. Soxhlet extraction proceeds for 18–24 hours using e.g. fresh dichloromethane:hexane (1:1) (USEPA, 1994).

More recently, pressurized liquid extraction (PLE) has become a common and faster alternative to Soxhlet extraction (Focant *et al.*, 2004). PLE uses organic solvents at temperatures above their boiling point maintained in the liquid phase under high pressure. The extraction cell which contains the sample is heated (e.g. 100–150°C) and filled up with an appropriate solvent (e.g. toluene, DCM) up to a pressure of 140 bars. The minimum extraction time should be 10 minutes in static mode, and several extraction cycles are recommended (n = 2–3). To further reduce analysis time, PLE can be combined with in-line clean-up procedures using preferably sulphuric acid impregnated silica as fat retainer (Björklund *et al.*, 2006). Proper fat-fat retainer ratios are important to avoid fat remaining in the sample after extraction. Mixed (polar/non-polar) solvent combinations cannot be used with this technique. It is understood that the combination of pressure and temperature is sufficient to remove all dioxins, furans and dl-PCBs from the matrix.

4.4 Clean-up

The extracts are concentrated using suitable evaporation devices, e.g. rotary evaporation, Turbovap, Syncore, Kuderna-Danish. The risk of cross-contamination is fairly high for rotary evaporation, so the evaporator should be pre-cleaned, e.g. by 100 ml

of clean solvent. If the extracts are to be cleaned up by adsorption chromatography on e.g. silica gel, a solvent change to hexane is recommended. The purification procedures have two objectives: i) removal or destruction of lipids and ii) removal of interfering compounds. Due to the very low levels of PCDD/Fs in biota samples, the elimination of interferences is essential. Prior to column chromatography clean-up, the precision and recovery of this step should be assessed.

For the first part, addition of concentrated sulphuric acid is commonly applied, either in terms of a column chromatography clean-up or by direct addition of silica impregnated with sulphuric acid to the extracts. The column chromatography clean-up suggested by USEPA (1994) for lipid removal in biota extracts includes 2 g of silica gel, 2 g of potassium silicate, 2 g of anhydrous Na_2SO_4, 10 g of silica gel (impregnated with sulphuric acid) and another 2 g of anhydrous sodium sulphate, to be packed bottom-to-top into a column of 25 mm ID. The column is pre-eluted with 100 ml of hexane and after loading of the sample, eluted with 200 ml of hexane. Ready to use, multi-layer clean-up columns are also available commercially. Alternatively, approximately 30 -100 g of sulphuric acid impregnated silica gel can be added to the extract (200 ml), while stirring for 2–3 hours. The treatment with sulphuric acid impregnated silica requires strict safety procedures as the small particles can cause serious health damage after inhalation. Gel permeation chromatography (GPC) has also been applied for lipid removal, but often a series of GPC columns is needed to ensure a 100% fat removal. Alternatively, an additional clean-up step using concentrated sulphuric acid might be applied after GPC to remove residual lipids from samples with higher lipid content.

For removal of interferences, HPLC, GPC and column chromatography using alumina, silica gel, Florisil and activated carbon are possible alternatives. USEPA (1994) suggests adsorption chromatography on alumina or Florisil and carbon as minimum additional clean-up steps after lipid removal. Depending on whether acid or basic alumina is chosen, the eluents should be dichloromethane:hexane (1:4) or (1:1), respectively. The material for the carbon column can be e.g. Carbopack™-C. Interferences are removed in a washing step with e.g. hexane, dichloromethane:cyclohexane and dichloromethane:toluene. Then, the column is inverted and the analytes are eluted with toluene.

HPLC can also be used for purification and fractionation of the extracts. 2-(1-pyrenyl)ethyldimethylsilylated (PYE) silica columns and porous graphitised carbon are suitable columns for this purpose (Echols *et al.*, 1998). When coupled in series, nitrophenylpropylsilica column (Nucleosil, 5 μm particles, 250 x 4.6 mm) and PYE (Cosmosil, 5 μm particles, 150 x 4.6 mm) enables the separation of PCDD/Fs from dl-PCBs (Bandh *et al.*, 1996). Fully automated clean-up systems are also available commercially (e.g. PowerPrep™ system). The European research project DIFFERENCE recommended at least three clean up or fractionation steps to ensure sufficiently clean extracts (Van Leeuwen *et al.*, 2007).

4.5 Concentration and syringe standards

After clean-up, a keeper is added (e.g. iso-octane or nonane) and the extracts are concentrated to near dryness, i.e. 10–20 μl. A syringe standard mix should also be added to evaluate the recovery of labelled internal standards. For example $^{13}C_{12}$ -1,2,3,4-TCDD can be used for recovery determinations of TCDD/Fs and PeCDD/Fs internal standards while $^{13}C_{12}$ -1,2,3,7,8,9-HxCDD can be used for recovery determinations of HxCDD/Fs, HpCDD/Fs and OCDD/F internal standards.

5. Instrumental analysis

The dioxin/furan content in environmental samples is commonly monitored using high resolution gas chromatography (HRGC) and high resolution mass spectrometry (HRMS), but low resolution mass spectrometry (LRMS) may be a suitable and cost-effective alternative if the required minimum performance criteria are met (see "HRGC/LRMS").

5.1 GC-analysis

The GC analysis should be optimized with regard to separation and sensitivity. Fishman *et al.* (2007) provided a comprehensive review of GC columns available for dioxin analysis. Generally 50–60 m, 5% diphenyl 95% dimethylpolysiloxane columns are a common choice. However, these columns could exhibit multiple co-elutions for both PCBs and PCDD/Fs (Reiner *et al.*, 2006), depending on the matrix to be analysed. The use of RTx-Dioxin 2 column has been reported in the literature as a suitable alternative to DB-5 columns. Combining this phase with reduced inner diameter and phase thickness (for example a 40m x 0.18mm x 0.18μm) enables the analysis of the 17 PCDD/F congeners in 40 minutes, with data fulfilling QA/QC requirements and providing better selectivity, especially for 2,3,7,8-TCDD and 2,3,7,8-TCDF (Reiner *et al.*, 2006; Cochran *et al.*, 2007).

Potential interferences for dl-PCBs on common GC-columns are summarized in Table 4 (Reiner *et al.*, 2006). Complete separation can be achieved by multi-analysis on columns of different polarity. The GC separation of congener CB-123 from interferences is critical. Recent developments indicate possibilities of full separation of relevant PCB congeners on one column, e.g. on an SGE HT8-PCB capillary column. A full separation of all PCB congeners is also possible by using comprehensive multi-dimensional GC (GCxGC).

Table 4. Possible interferences for selected dl-PCBs using a 5% phenyl column (Reiner *et al.*, 2006).

PCB CONGENER	POTENTIAL INTERFERENCE
CB-81	CB-87
CB-77	CB-110
CB-123	CB-149
CB-126	CB-178 and CB-129
CB-156	CB-171
CB-157	CB-201

Various injection techniques are possible, e.g. on-column injection, splitless injection, pressure-pulsed splitless injection and programmed temperature vaporizing (PTV) injection. The most suitable injection volume depends on the dioxin concentrations in the sample and the sensitivity of the instrumental analysis. In HRGC/HRMS analysis, 1–2 μl are common injection volumes.

5.2 Compound identification

The HRMS system should be operated at a minimum of 10,000 resolving power throughout all the runs, and resolution should be checked regularly during the sequence of runs. The individual dl-PCBs, PCDD/Fs or labelled compounds are identified by comparing the GC retention time and ion abundance ratio of two exact

masses monitored (Tables 5 and 6) with the corresponding retention time of an au-thentic labelled internal standard and the theoretical or acquired ion abundance ratio of the two exact masses. The congeners for which there are no labelled analogues are identified when relative retention time and ion abundance ratios agree within prede-fined limits. The following criteria should be met for identification of an individual dl-PCB, PCDD/F or labelled compound in a standard, blank or sample:

- The signal for the two exact masses specified in Tables 5 and 6 should be present and within ± 2 s.

- The signal-to-noise ratio (S/N) for the GC peak at each exact mass has to be at least 3 for each congener detected in a sample extract, and at least 10 for all congeners in the calibration standard.

- The ratio of the integrated areas of the two exact masses specified in Tables 5 and 6 has to be within 15% of the theoretical shown in Table 7.

- The relative retention time of a native dl-PCB, PCDD/F has to be within a time window of ± 3 s based on the retention time of the corresponding $^{13}C_{12}$-labelled standard. The relative retention time of congeners for which there are no labelled analogues has to be within ± 0.002.

If interferences preclude identification, extract a new, further cleaned up aliquot and analyse again. If interferences cannot be removed flag the data to indicate results are maximum concentrations.

5.3 Compound quantification

Quantitative analysis is performed using selected ion monitoring (SIM) area, in one of the two following ways:

- For the dl-PCBs and PCDD/Fs for which labelled analogues have been added to the sample (Table 2), the GC/MS system is calibrated, and the concentration of each compound is determined using the isotope dilution technique.

- For the dl-PCBs and PCDD/Fs for which labelled analogues are not added to the sample (see Table 3 for PCDD/Fs), the GC/MS system is calibrated for each compound using a labelled isomer with the most similar structure and the concentration of each compound is determined using the internal standard technique.

Calibration curves should be based on a minimum of 5 calibration points. Mass drift correction is mandatory, usually based on a lock-mass m/z of perfluorokerosene (PFK) or perfluorotributylamine (PFTBA, FC43).

Table 5. Masses for the detection and quantification of PCDD/Fs.

SUBSTANCE	DIBENZOFURANS (M/Z)		DIBENZO–P–DIOXINS (M/Z)	
	NATIVE	¹³C₁₂–LABELLED	NATIVE	¹³C₁₂–LABELLED
Tetra-CDD/F	303.9016	315.9419	319.8965	331.9368
	305.8987	317.9389	321.8937	333.9339
Penta-CDD/F	339.8598	351.9000	355.8547	367.8949
	341.8569	353.8970	357.8518	369.8919
Hexa-CDD/F	373.8208	385.8610	389.8157	401.8559
	375.8179	387.8580	391.8128	403.8529
Hepta-CDD/F	407.7818	419.8220	423.7767	435.8169
	409.7789	421.8190	425.7738	437.8140
Octa-CDD/F	441.7428	453.7830	457.7377	469.7779
	443.7399	455.7801	459.7348	471.7750

Table 6. Masses for the detection and quantification of PCBs.

HOMOLOGUE GROUPS	NATIVE CBS (M/Z)	¹³C₁₂–LABELLED CBS (M/Z)
Tetrachlorobiphenyls	289.9223	301.9626
	291.9194	303.9597
Pentachlorobiphenyls	325.8804	337.9207
	327.8775	339.9177
Hexachlorobiphenyls	359.8415	371.8817
	361.8385	373.8788
Heptachlorobiphenyls	393.8025	405.8427
	395.7995	407.8398

The isotope ratio between the two ions of the molecular isotope cluster, which are recorded, has to match the theoretical value within ± 15% (see Table 7).

Table 7. Tolerance limits of isotope ratios for PCDD/Fs and dl-PCBs.

CHLORINE ATOMS	ISOTOPE RATIO LOWER LIMIT	ISOTOPE RATIO THEORETICAL VALUE	ISOTOPE RATIO UPPER LIMIT
4	0.65	0.77 (M/M+2)	0.89
5	0.55	0.64 (M+4/M+2)	0.75
6	0.69	0.81 (M+4/M+2)	0.94
7	0.83	0.96 (M+4/M+2)	1.10
8	0.76	0.89 (M+2/M+4)	1.02

5.4 HRGC/LRMS

Low resolution mass spectrometry (LRMS) has also been applied to the analysis of PCDD/Fs and/or dl-PCBs. Limits of detections are higher than those obtained with HRMS detectors, but can be compensated by e.g. larger injection volumes. A very efficient extract clean-up is of the utmost importance to exclude any interferences. A technique commonly applied is GC-LRMS using ion trap mass analysers working in tandem mode (Focant *et al.*, 2005; Malavia *et al.*, 2008, Eppe *et al.*, 2004). Table 8 pro-

vides information on precursor and product ions obtained by GC-ion trap MS. GC-LRMS (quadrupole) can be an option for dl-PCBs in particular.

Both HRMS and LRMS techniques have to demonstrate that they meet the requirements regarding separation and sensitivity described in the monitoring programme, see also comments under "Quality Assurance and Quality Control". When using LRMS the maintenance of the instrument is crucial and could be time consuming (e.g. frequent cleaning of the ion source). The sensitivity for 2,3,7,8-TCDD may be critical.

Table 8. Precursor ions and product ions for the determination of PCDD/Fs and dl-PCBs by HRGC-ion trap tandem MS.

TARGET COMPOUNDS	NATIVE		$^{13}C_{12}$–LABELLED	
	PRECURSOR ION (M/Z)	PRODUCT IONS (M/Z)	PRECURSOR ION (M/Z)	PRODUCT IONS (M/Z)
TCDD	322 (M+2)	257 + 259	334 (M+2)	268 + 270
PeCDD	356 (M+2)	291 + 293	368 (M+2)	302 + 304
HxCDD	390 (M+2)	325 + 327	402 (M+2)	336 + 338
HpCDD	424 (M+2)	359 + 361	436 (M+2)	370 + 372
OCDD	460 (M+4)	395 + 397	472 (M+4)	406 + 408
TCDF	306 (M+2)	241 + 243	318 (M+2)	252 + 254
PeCDF	340 (M+2)	275 + 277	352 (M+2)	286 + 288
HxCDF	374 (M+2)	309 + 311	386 (M+2)	320 + 322
HpCDF	408 (M+2)	343 + 345	420 (M+2)	354 + 356
OCDF	444 (M+4)	379 + 381		
CB-81, 77	292 (M+2)	220 + 222	304 (M+2)	232 + 234
CB-123, 118, 114, 105, 126	326 (M+2)	254 + 256	338 (M+2)	266 + 268
CB-167, 156, 157, 169	360 (M+2)	288 + 290	372 (M+2)	300 + 302
CB-189	394 (M+2)	322 + 324	406 (M+2)	334 + 336

6. Quality Assurance and Quality Control

The laboratory is required to operate a formal quality assurance programme. Indicative values for accuracy and precision are given under "Verification method" in Table 9. An example of a comprehensive QA/QC approach is described in method 1613 by USEPA (1994).

The analytical method requires high sensitivity and low detection limits, usually in the pg-range, for both dioxins/furans and dl-PCB congeners (OSPAR, 2005), and should meet the requirements for LoQ specified in the monitoring programme. For individual PCDD/Fs, LoQ of 0.3 pg/g wet weight should be achievable, with the exception of OCDD (1 pg/g wet weight). For non-*ortho* PCBs, LoQ should be as low as 5 pg/g wet weight, while for mono-*ortho* PCBs requirements on LoQ are less strict as their concentrations in biota samples are usually higher, in particular concentrations of congeners CB-105, CB-118 and CB-156. The selectivity of the method should be sufficient to avoid interfering compounds, i.e. the individual congeners should be separated from each other and any interferences present.

All sample series should include procedural blanks and measurements of certified/laboratory reference materials. Certified reference materials should be analysed

regularly, although only few are available for the determination of PCDD/Fs and dl-PCBs in biota, for example from NIST (cod liver oil), Cambridge Isotope Laboratories (fish) and the National Research Council Canada (Fish) (De Boer and McGovern, 2001). The laboratory should further prove its competence by regular participation in relevant laboratory proficiency tests. It is essential that the matrix and concentration range of the proficiency testing samples are comparable with the samples routinely analysed within the monitoring programme (De Boer, 2001; Wells and de Boer, 2006).

The recovery of the individual internal standards added prior to extraction should be between 60–120%. Blanks should be as low as possible, at least below 20% of the lowest concentration of interest.

7. Screening methods based on bioassays

Bioassays are not currently applied in the monitoring under the OSPAR Coordinated Environmental Monitoring Programme (CEMP), but have been suggested as screening tools for monitoring PCDD/F and dl-PCB in foodstuffs (COMMISSION DIRECTIVE 2002/69/EC), with the requirement to meet the criteria given in Table 9. Screening tools might be useful in, for instance, choice of sampling sites, and will therefore be briefly discussed in this guideline. Hurst *et al.* (2004) also emphasized that monitoring programmes were moving towards effect-based monitoring, with biological relevance becoming more important. The authors list the following requirements for bioassays to be included in monitoring programmes: The tool must be capable of rapid, inexpensive and high-throughput screening producing interpretable and meaningful results (Hurst *et al.*, 2004).

Table 9. Quality criteria for screening and verification methods (COMMISSION DIRECTIVE 2002/69/EC).

	SCREENING METHOD	VERIFICATION METHOD
False negatives	< 1%	
Accuracy		± 20%
Precision (expressed as the coefficient of variation between repeated measurements)	< 30%	< 15%

The dioxin responsive chemically activated luciferase expression (DR-CALUX) assay is mechanism specific and utilizes the interaction of compounds with the AhR. However, it is not compound specific and produces a response with all compounds capable of interactions with the AhR. While COMMISSION DIRECTIVE 2002/69/EC demands that the TEQ-values determined by bioassays should be the sum of PCDD/Fs and dl-PCBs in the sample, Hurst *et al.* (2004) and Van Leeuwen *et al.* (2007) demonstrated some disagreement between the results of the bioassay and the conventional targeted HRGC/HRMS analysis. The differences may be caused by unknown compounds producing a dioxin-like response in the CALUX assay (e.g. brominated or mixed halogenated dioxin analogues, polychlorinated naphthalenes, PAHs etc.) – or compounds antagonising the AhR (e.g. di-*ortho*-substituted PCBs).

These deviations from results of chemical analysis were also considered as an advantage by Hurst *et al.* (2004), as the assay allows a more accurate assessment of the true potency of dioxin-like compounds present in the samples. In order to obtain specific responses to PCDD/Fs and dl-PCBs in the sample, the extracts require specific clean-up methods to exclude interferences from other dioxin-like compounds. As men-

tioned above, environmental monitoring aims at presenting concentrations of individual compounds in the respective samples, rather than toxicity assessments.

8. Safety

The chemical compounds dealt with in this guideline are hazardous and must only be handled by trained personnel familiar with handling of PCDD/F and dl-PCBs, and associated risks as well as precautionary measures. USEPA (1994) recommends that laboratories purchase diluted standard solutions instead of preparing primary solutions.

9. Data reporting

Results are reported in pg/g ww. The lipid content and water content of the samples should be reported as well. For normalizing purposes the total lipid content should be determined, rather than the extractable lipid content (De Boer, 1988). Concentrations are reported to two significant figures. Minimum performance criteria such as LoQ and measurement uncertainty along with information on blanks and reference materials should be included in the report.

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Acknowledgements

Annex 10: Technical annex for polychlorinated dibenzo–*p*–dioxins, polychlorinated dibenzofurans and dioxin–like polychlorinated biphenyls in sediments

1. Introduction

Polychlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDD/Fs) are ubiquitous in the environment, primarily as unintentional by-products of combustion and industrial processes. They enter the aquatic environment via several routes, including atmospheric deposition. As PCDD/Fs are strongly hydrophobic compounds, sediments are the eventual sink in the aquatic environment, providing a source of potential exposure to aquatic organisms (Hurst *et al.*, 2004). This guideline only addresses the 17 tetra- through octa-2,3,7,8-substituted PCDD/F congeners, and the non- and mono-*ortho* substituted polychlorinated biphenyls (PCBs), which exhibit similar effects as the 2,3,7,8-substituted PCDD/Fs. The general chemical structures of PCDD/Fs and PCB congeners are given in Figure 1.

In this guideline, the term "dioxin-like PCBs" (dl-PCBs) is used for the non-*ortho* and mono-*ortho* PCB congeners listed in Table 1. The coplanar structure of non-*ortho* PCB congeners allows a configuration similar to that of PCDD/Fs. Mono-*ortho* substituted PCBs may take a steric position close to coplanarity and are consequently less toxic than non-*ortho* PCBs. Nevertheless, they have been considered in this guideline due to their relatively high concentrations in sediments compared to those of non-*ortho* PCBs or PCDD/Fs (e.g. Castro-Jiménez *et al.*, 2008; Helm *et al.*, 2008).

PCDD/Fs and dl-PCBs have been shown to produce various toxic responses, including immunotoxicity, developmental and reproductive effects and carcinogenesis (OSPAR, 2005). The initial mechanism of toxicity is via the aryl hydrocarbon receptor (AhR), leading to changes in gene expression, cell growth and cell differentiation (Nebert *et al.*, 1993; Hurst *et al.*, 2004). Due to their persistence, high toxicity, bioaccumulation potential and ability for long-range transport, they are controlled under the Stockholm Convention for Persistent Organic Pollutants (POPs). Spatial and temporal trend monitoring of PCDD/Fs in the aquatic environment is important for the evaluation of risks to wildlife and human health (Hurst *et al.*, 2004). Due to the low concentrations at which adverse effects can be observed, the analytical methodology for the analysis of PCDD/Fs and dl-PCBs differs from that for other organochlorine compounds.
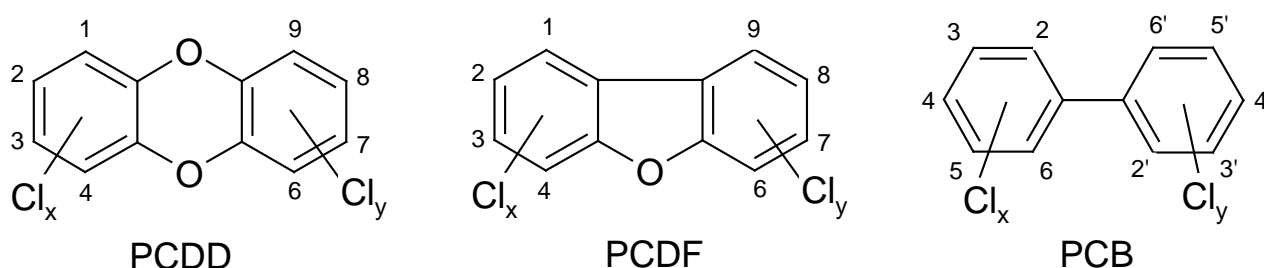

Figure 1. General structure of PCDDs, PCDFs and PCBs. The possible number of chlorine atoms results in 75 PCDD congeners, 135 PCDF congeners (x=1–4, y=0–4), and 209 PCB congeners (x=1–5, y=0–5).

The objective of this guideline is to give general advice on the analysis of PCDD/Fs and dl-PCBs in marine sediments. It is not a complete review of possible methods or an in-depth discussion of specific methods.

2. Analytes

Environmental monitoring should include the 17 tetra- through octa-chlorinated 2,3,7,8-PCDD/F congeners and the dl-PCBs listed in Table 1. Concentrations of individual PCDD/Fs and dl-PCB congeners in sediment are normally reported in pg/g dry weight (dw), with additional information on dry matter and organic carbon content (see chapter "Data reporting"). To assess overall toxicity, concentrations may additionally be expressed as toxicity equivalents (TEQs). However, although common practice to compare contamination levels, the use of TEQs derived from WHO TEFs is of limited relevance in abiotic environmental matrices (van den Berg *et al.*, 2006) and is therefore not recommended. The ICES guideline on PCDD/Fs and dl-PCBs in biota gives further information on the TEF systems in use (ICES, 2010).

Table 1. Chlorinated dibenzo-p-dioxins (PCDDs), chlorinated dibenzofurans (PCDFs) and dl-PCBs recommended for environmental monitoring.

HOMOLOGOUS GROUP	CONGENER	IUPAC NO.
PCDDs		
TCDD	2,3,7,8	
PeCDD	1,2,3,7,8	
HxCDD	1,2,3,4,7,8	
	1,2,3,6,7,8	
	1,2,3,7,8,9	
HpCDD	1,2,3,4,6,7,8	
OCDD	1,2,3,4,6,7,8,9	
PCDFs		
TCDF	2,3,7,8	
PeCDF	1,2,3,7,8	
	2,3,4,7,8	
HxCDF	1,2,3,4,7,8	
	1,2,3,6,7,8	
	1,2,3,7,8,9	
	2,3,4,6,7,8	
HpCDF	1,2,3,4,6,7,8	
	1,2,3,4,7,8,9	
OCDF	1,2,3,4,6,7,8,9	
NON−ORTHO PCBs		
TeCB	3,3',4,4'	77
TeCB	3,4,4',5	81
PeCB	3,3',4,4',5	126
HxCB	3,3',4,4',5,5'	169
MONO−ORTHO PCBs		
PeCB	2,3,3',4,4'	105
	2,3,4,4',5	114

HOMOLOGOUS GROUP	CONGENER	IUPAC NO.
	2,3',4,4',5	118
	2',3,4,4',5	123
HxCB	2,3,3',4,4',5	156
	2,3,3',4,4',5	157
	2,3',4,4',5,5'	167
HpCB	2,3,3',4,4',5,5'	189

3.   Sediment samples

OSPAR (2005) presented a monitoring strategy for PCDD/Fs in the environment, which identified marine sediments as one of the most important matrices (the other one being biota). Marine sediments, in particular those with a high organic carbon content, may accumulate hydrophobic compounds like PCDD/Fs and dl-PCBs to considerably higher levels than those of the surrounding waters. The sampling strategy depends on the purpose of the monitoring programme and the natural conditions of the region to be monitored and typically includes fixed-station sampling, stratified random sampling or stratified fixed sampling. Muddy sediments, i.e. containing a large proportion of fine material, are preferable for organic contaminant monitoring although sieving of sediments may be an alternative (OSPAR, 2002).

In general, the same recommendations are valid as described for other organochlorine compounds, i.e. in the OSPAR JAMP guideline on organic contaminants (OCs) in marine sediments (OSPAR, 2002), which also contains details on sample handling. It should be pointed out, however, that the risk of sample contamination is considerably higher, given the extremely low concentrations of PCDD/Fs and dl-PCBs in most environmental samples. The staff collecting and handling the samples should be well-trained and properly instructed in how to avoid any contamination.

4.   Analytical methods

Prior to systematic monitoring, it will be meaningful to perform an initial screening of the area under consideration, e.g. to check the relevance of monitoring stations. For this purpose, several suitable methods are available. In particular, bioassays and fast semi-quantitative GC-MS screening methods can give an indication of the extent of the contamination (e.g. Schrock *et al.*, 2009).

For a full quantitative analysis of individual congeners, state-of-the-art GC-MS is the preferred analytical method. Multidimensional gas chromatography (GCxGC) could be an alternative for PCDD/F and dl-PCB analysis, even in combination with ECD, as this will offer enough sensitivity and the selectivity will exceed that of single-column GC analysis provided a proper orthogonal combination of columns is selected (Haglund *et al.*, 2008). GCxGC can also be used in combination with LRMS Time-of-Flight (ToF)-MS (Skoczynska *et al.*, 2008).

An example of a suitable method for the analysis of sediment samples is summarized in Figure 2.

4.1   Preparatory steps

It is essential to avoid contamination during all analytical steps. Reagents should be of high purity, and where appropriate be cleaned by e.g. extraction or solvent rinse. All solvents used must be checked for the presence of residues of target or interfering compounds (e.g. polychlorinated diphenyl ethers). The purity of standards should be checked. Reusable glassware should be rinsed with solvent, disassembled, washed with a detergent solution and further rinsed with ultrapure water and solvent. Baking glassware at > 400°C is common practice as part of the cleaning process, but the formation of active sites on the glass surface that may adsorb the target compounds has been reported (USEPA, 1994). New glassware should be used from time to time as scratched glassware has more active sites.

The preparation of stock solutions and standards can follow the guidelines developed for OCs in sediment. However, care has to be taken to monitor and to avoid contamination. Furthermore, the high toxicity of the compounds might require a particularly careful handling; see comments under "Safety". Commercially available diluted stock solutions can be used to reduce safety issues. As valid for the entire analytical method, only trained personnel should perform these steps.

PCDD/Fs and dl-PCBs are normally determined by isotope dilution, using high resolution gas chromatography and high resolution mass spectrometry (HRGC/HRMS). ^{13}C-labelled standards of all the congeners to be analysed are added prior to extraction of the samples. These internal standards correct for errors and recovery losses, to some extent. ^{13}C-labelled standards are commercially available of all individual congeners and should preferably be used. The minimum number of internal standards to be used for the quantification of PCDD/Fs congeners is given in Table 2. Details on spike procedures are given by e.g. USEPA (1994).

Table 2. Minimum number of internal standards to be used for calibration of PCDD and PCDF homologue groups.

SUBSTANCE	PCDD–HOMOLOGUES		PCDF–HOMOLOGUES	
	NATIVE	13C12–LABELLED	NATIVE	13C12–LABELLED
Tetrachloro homologues	2,3,7,8	2,3,7,8	2,3,7,8	2,3,7,8
Pentachloro homologues	1,2,3,7,8	1,2,3,7,8	1,2,3,7,8 2,3,4,7,8	1,2,3,7,8 2,3,4,7,8
Hexachloro homologues	1,2,3,4,7,8 1,2,3,6,7,8 1,2,3,7,8,9	1,2,3,7,8,9	1,2,3,4,7,8 1,2,3,6,7,8 1,2,3,7,8,9 2,3,4,6,7,8	2,3,4,6,7,8
Heptachloro homologues	1,2,3,4,6,7,8	1,2,3,4,6,7,8	1,2,3,4,6,7,8 1,2,3,4,7,8,9	1,2,3,4,6,7,8

4.2   Extraction

The sample weight used for the extraction must be sufficient to fulfil the requirements with respect to sensitivity. 10 g of dried sediment are typically used to accurately measure PCDD/Fs and dl-PCBs by gas chromatography-high resolution mass spectrometry (GC-HRMS; USEPA, 1994). The samples are spiked with the ^{13}C-labelled standards as described above.

For extraction of solid samples, Soxhlet is widely accepted as a robust liquid–solid extraction technique. Immediately prior to use, the Soxhlet apparatus should be pre-extracted with e.g. dichloromethane:hexane (1:1) for approximately 3 hours (USEPA, 1994). The EPA method 1613B recommends to Soxhlet extract the sediment samples in toluene for 16–24 hours. Other typical solvents for Soxhlet extraction in the litera-ture are dichloromethane (Koh *et al.*, 2004), dichloromethane:hexane 1:1 (Liu *et al.*, 2006) and toluene:acetone 4:1 (Stern *et al.*, 2005) with extraction times between 16–24h. To remove sulphur compounds, copper granules can be either mixed with sedi-ment during extraction or added to the extract.

More recently, pressurised liquid extraction (PLE) has become a common and faster alternative to Soxhlet extraction (Antunes *et al.*, 2008; Kishida *et al.*, 2010). PLE uses organic solvents at temperatures above their boiling point maintained in the liquid phase under high pressure. The extraction cell which contains the sample is heated (ex: 100°C-150°C) and filled up with an appropriate solvent (example: toluene, di-chloromethane) up to a pressure of 140 bars. The minimum extraction time should be 10 minutes in static mode, and several extraction cycles are recommended (n = 2–3). To further reduce analysis time, PLE can be combined with in-line clean-up proce-dures (Chuang *et al.*, 2009). Mixed (polar/non-polar) solvent combinations cannot be used with this technique because of their different boiling points. Provided the proper conditions are chosen, the combination of pressure and temperature is suffi-cient to extract all PCDD/Fs and dl-PCBs from the matrix.

4.3   Clean-up

The extracts are concentrated using suitable evaporation devices, e.g. rotary evapora-tion, Turbo-Vap, Syncore, Kuderna-Danish. The risk of cross-contamination is fairly high for rotary evaporation, so the evaporator should be pre-cleaned, e.g. by 100 mL of clean solvent.

Due to the very low levels of PCDD/Fs in sediment samples, the elimination of inter-ferences is essential, both with regard to matrix effects and co-eluting compounds. Often, a combination of clean-up steps will be required, including e.g. gel permeation chromatography (GPC) and adsorption chromatographic techniques (preferably with a Florisil and/or acid silica, alumina and/or carbon column). The most common sol-vents used in this step are hexane and/or dichloromethane and toluene. If the extracts are to be cleaned up by adsorption chromatography on e.g. silica gel, a solvent change to hexane is essential.

Another possibility is the use of two-dimensional HPLC with two columns coupled in series. Nitrophenylpropylsilica (Nucleosil, 5 µm particles, 250 x 4.6 mm) and PYE (Cosmosil, 5 µm particles, 150 x 4.6 mm) columns enable the separation according to the number of aromatic rings and planarity, i.e. non-*ortho* and mono-*ortho* PCBs can be separated from each other as well as from PCDD/Fs (Bandh *et al.*, 1996). The eluting solvent is a non-polar solvent such as *iso*-hexane. Coupled to a fraction collec-tor, the use of a HPLC system allows the automatic clean-up of a considerable num-ber of samples. Alternatively, HPLC systems equipped with porous graphitised carbon can be used. Column sizes are in the order of 50 x 4.7 mm and care has to be taken that the column is not overloaded. Similarly to PYE columns, they will separate non-*ortho* PCBs from other PCBs and from PCDD/Fs. Fully automated clean-up sys-tems are also available commercially (e.g. PowerPrep™ system).

The European research project DIFFERENCE recommended at least three clean-up or fractionation steps to ensure sufficiently clean extracts (Van Loco *et al.*, 2004).

4.4   Concentration and injection standards

After clean-up, a keeper is added (e.g. iso-octane or nonane) and the extracts are concentrated to near dryness, i.e. 10–20 μl. An injection standard mix should also be added to evaluate the recovery of labelled internal standards. For example $^{13}C_{12}$ - 1,2,3,4-TCDD can be used for recovery determinations of TCDD/Fs and PeCDD/Fs internal standards while $^{13}C_{12}$-1,2,3,7,8,9-HxCDD can be used for recovery determinations of HxCDD/Fs, HpCDD/Fs and OCDD/F internal standards.

5.   HRGC/HRMS

The PCDD/F levels in environmental samples are commonly monitored using high resolution gas chromatography (HRGC) and high resolution mass spectrometry (HRMS), but low resolution mass spectrometry (LRMS) may be a suitable and cost-effective alternative if the required minimum performance criteria are met (see "HRGC/LRMS").

5.1   GC–analysis

The GC analysis should be optimized with regard to separation and sensitivity. Gas chromatographic separation of isomers must be sufficient (< 25% peak to peak between 1,2,3,4,7,8-HxCDF and 1,2,3,6,7,8-HxCDF). Fishman *et al*. (2007) provided a comprehensive review of GC columns available for dioxin analysis. Generally, 50–60 m, 5% diphenyl 95% dimethylpolysiloxane columns are a common choice. However, these columns could exhibit multiple co-elutions for both PCBs and PCDD/Fs (Reiner *et al*., 2006), depending on the matrix to be analysed. The use of RTx-Dioxin 2 columns has been reported in the literature as a suitable alternative to DB-5 columns. Combining this phase with reduced inner diameter and phase thickness (for example a 40m x 0.18mm x 0.18μm) enables the analysis of the 17 PCDD/F congeners in 40 minutes, with data fulfilling QA/QC requirements and providing better selectivity, especially for 2,3,7,8-TCDD and 2,3,7,8-TCDF (Robinson *et al*., 2004; Reiner *et al*., 2006).

Potential interferences for dl-PCBs on common GC-columns are summarized in Table 3 (Reiner *et al*., 2006). Complete separation can be achieved by multi-analysis on columns of different polarity. The GC separation of congener CB-123 from interferences is critical. Possibilities of full separation of relevant PCB congeners on one column have been reported, e.g. on an SGE HT8-PCB capillary column (Larsen *et al*., 1995).

Table 3. Possible interferences for selected dl-PCBs using a 5% phenyl column (Reiner *et al.* 2006).

PCB CONGENER	POTENTIAL INTERFERENCE
CB-81	CB-87
CB-77	CB-110
CB-123	CB-149
CB-126	CB-178 and CB-129
CB-156	CB-171
CB-157	CB-201

Various injection techniques are possible, e.g. on-column injection, splitless injection, pressure-pulsed splitless injection and programmed temperature vaporizing (PTV) injection. The most suitable injection volume depends on the target concentrations in

the sample and the sensitivity of the instrumental analysis. In HRGC/HRMS analysis, 1–2 μl are common injection volumes. With PTV injection volumes of up to 50 μl can be achieved. This has the potential to significantly lower detection limits, which is particularly helpful for low resolution MS techniques. Helium is normally used as carrier gas.

Regarding temperature programmes, the USEPA method recommends to start the GC oven programme at 200°C, but other methodologies in the literature start from 90, 140 or 160°C (Zhang *et al.*, 2008; Robinson *et al.*, 2004; Loganathan *et al.*, 2008). However, the initial oven temperature is linked to the keeper solvent and the injection technique and should ensure that no solvent effects and peak discrimination occur.

5.2   Compound identification

The HRMS system should be operated at a minimum of 10,000 resolving power, and resolution should be checked regularly during the sequence of runs. The individual dl-PCBs, PCDD/Fs or labelled compounds are identified by comparing the GC retention time and ion abundance ratio of two exact masses monitored (Tables 4 and 5) with the corresponding retention time of an authentic labelled internal standard and the theoretical or acquired ion abundance ratio of the two exact masses. The congeners for which there are no labelled analogues are identified when relative retention time and ion abundance ratios agree within predefined limits. The following criteria should be met for identification of an individual dl-PCB, PCDD/F or labelled compound in a standard, blank or sample:

- The signal for the two exact masses specified in Tables 4 and 5 should be present and within ± 2 s.
- The signal-to-noise ratio (S/N) for the GC peak at each exact mass has to be at least 3 for each congener detected in a sample extract, and at least 10 for all congeners in the calibration standard.
- The ratio of the integrated areas of the two exact masses specified in Tables 4 and 5 has to be within 15% of the theoretical one shown in Table 6.
- The relative retention time of a native dl-PCB, PCDD/F has to be within a time window of ± 0.003 based on the retention time of the corresponding $^{13}C_{12}$-labelled standard. The relative retention time of congeners for which there are no labelled analogues has to be within ± 0.002.

If interferences preclude identification, extract a new, further cleaned up aliquot and analyse again. If interferences cannot be removed flag the data to indicate results are maximum concentrations.

5.3   Compound quantification

Quantitative analysis is performed using selected ion monitoring (SIM), in one of the two following ways:

- For the PCDD/Fs and dl-PCBs for which labelled analogues have been added to the sample, the GC/MS system is calibrated, and the concentration of each compound is determined using the isotope dilution technique.
- For the PCDD/Fs and dl-PCBs for which labelled analogues are not added to the sample (Table 2), the GC/MS system is calibrated for each compound using a labelled isomer with the most similar structure and the concentration of each compound is determined using the internal standard technique.

Calibration curves should be based on a minimum of 5 calibration points. Mass drift correction is mandatory, usually based on a lock-mass m/z of perfluorokerosene (PFK) or perfluorotributylamine (PFTBA, FC43).

Table 4. Masses m/z for the detection and quantification of PCDD/Fs.

SUBSTANCE	DIBENZOFURANS (M/Z)		DIBENZO−P−DIOXINS (M/Z)	
	NATIVE	13C12−LABELLED	NATIVE	13C12−LABELLED
Tetra-CDD/F	303.9016	315.9419	319.8965	331.9368
	305.8987	317.9389	321.8937	333.9339
Penta-CDD/F	339.8598	351.9000	355.8547	367.8949
	341.8569	353.8970	357.8518	369.8919
Hexa-CDD/F	373.8208	385.8610	389.8157	401.8559
	375.8179	387.8580	391.8128	403.8529
Hepta-CDD/F	407.7818	419.8220	423.7767	435.8169
	409.7789	421.8190	425.7738	437.8140
Octa-CDD/F	441.7428	453.7830	457.7377	469.7779
	443.7399	455.7801	459.7348	471.7750

Table 5. Masses *m/z* for the detection and quantification of PCBs.

HOMOLOGUE GROUPS	NATIVE CBS (M/Z)	13C12−LABELLED CBS (M/Z)
Tetrachlorobiphenyls	289.9223	301.9626
	291.9194	303.9597
Pentachlorobiphenyls	325.8804	337.9207
	327.8775	339.9177
Hexachlorobiphenyls	359.8415	371.8817
	361.8385	373.8788
Heptachlorobiphenyls	393.8025	405.8427
	395.7995	407.8398

The isotope ratio between the two ions of the molecular isotope cluster, which are recorded, has to match the theoretical value within ± 15% (see Table 6; USEPA, 1994).

Table 6. Tolerance limits of isotope ratios for PCDD/Fs and dl-PCBs.

CHLORINE ATOMS	ISOTOPE RATIO LOWER LIMIT	ISOTOPE RATIO THEORETICAL VALUE	ISOTOPE RATIO UPPER LIMIT
4	0.65	0.77 (M/M+2)	0.89
5	0.55	0.64 (M+4/M+2)	0.75
6	0.69	0.81 (M+4/M+2)	0.94
7	0.83	0.96 (M+4/M+2)	1.10
8	0.76	0.89 (M+2/M+4)	1.02

6.   HRGC/LRMS

Low resolution mass spectrometry (LRMS) has also been applied to the analysis of PCDD/Fs and/or dl-PCBs. Limits of detections are higher than those obtained with HRMS detectors, but can be compensated by e.g. larger injection volumes (see

above). A very efficient extract clean-up is of the utmost importance to exclude any interferences. A technique commonly applied is GC-LRMS using ion trap mass analysers working in tandem mode (Eppe *et al.*, 2004; Focant *et al.*, 2005; Malavia *et al.*, 2008). Table 7 provides information on precursor and product ions obtained by GC-ion trap MS. GC-LRMS (quadrupole) can be an option for dl-PCBs in particular.

Schrock *et al.* (2009) implemented a method capable of detecting the lowest calibration point of the EPA Method 1613B corresponding to sample concentrations of 1, 5 and 10 pg/g for tetra-, penta-hepta, and octa-chlorinated congeners, respectively, for a sample intake of 10 g of sediments when samples were sufficiently cleaned. Chromatograms generated by LRMS were quantified using the same identification and quantification criteria, achieving a resolution of 45% between 2,3,7,8-TCDD and its closest eluting isomer.

The USEPA method 8280A is also focused on the analysis of PCDD/Fs by low resolution MS. This method is appropriate to the determination in environmental samples when the expected concentrations of the PCDD/Fs in soil samples are above 1, 2.5 or 5.0 µg/kg for tetra, penta-hexa-hepta, and octa-chlorinated congeners, respectively. The maintenance of the instrument is crucial and could be time consuming (e.g. frequent cleaning of the ion source). The sensitivity for 2,3,7,8-TCDD may be critical.

Table 7.Precursor ions *(m/z)* **and product ions** *(m/z)* **for the determination of PCDD/Fs and dl-PCBs by HRGC-ion trap tandem MS.**

TARGET COMPOUNDS	NATIVE		13C12–LABELLED	
	PRECURSOR ION (M/Z)	PRODUCT IONS (M/Z)	PRECURSOR ION (M/Z)	PRODUCT IONS (M/Z)
TCDD	322 (M+2)	257 + 259	334 (M+2)	268 + 270
PeCDD	356 (M+2)	291 + 293	368 (M+2)	302 + 304
HxCDD	390 (M+2)	325 + 327	402 (M+2)	336 + 338
HpCDD	424 (M+2)	359 + 361	436 (M+2)	370 + 372
OCDD	460 (M+4)	395 + 397	472 (M+4)	406 + 408
TCDF	306 (M+2)	241 + 243	318 (M+2)	252 + 254
PeCDF	340 (M+2)	275 + 277	352 (M+2)	286 + 288
HxCDF	374 (M+2)	309 + 311	386 (M+2)	320 + 322
HpCDF	408 (M+2)	343 + 345	420 (M+2)	354 + 356
OCDF	444 (M+4)	379 + 381		
CB-81, 77	292 (M+2)	220 + 222	304 (M+2)	232 + 234
CB-123, 118, 114, 105, 126	326 (M+2)	254 + 256	338 (M+2)	266 + 268
CB-167, 156, 157, 169	360 (M+2)	288 + 290	372 (M+2)	300 + 302
CB-189	394 (M+2)	322 + 324	406 (M+2)	334 + 336

ToF-MS and Orbitrap MS fill a gap between LR and HRMS and often offer an even higher resolution than HRMS instruments (Orbitrap: up to 100,000). This resolution is, however, mass dependent in contrast to HR sector instruments which normally offer a resolution of 10,000 or more, independent of the mass. To compare the resolution of HRMS and that of ToF and Orbitrap instruments, the resolution of the latter two should be reduced by ca. a factor 2, as they are measured at 50% peak height. Nonetheless, they require much less investment than an HRMS sector instrument

does and are likely to offer enough sensitivity and selectivity (full scan options and high resolution) for monitoring of PCDD/Fs and dl-PCBs in sediments.

7.   Quality Assurance and Quality Control

The laboratory is required to operate a formal quality assurance programme. An example of a comprehensive QA/QC approach is described in method 1613 by USEPA (1994).

The analytical method requires high sensitivity and low detection limits for both PCDD/Fs and dl-PCB congeners and should meet the requirements for limits of quantification (LoQ) specified in the monitoring programme. The selectivity of the method should be sufficient to avoid interfering compounds, i.e. the individual congeners should be separated from each other and any interferences present. The recovery of the individual internal standards added prior to extraction should be between 60–120%.

All sample series should include procedural blanks and measurements of reference materials. Blanks should be as low as possible, at least below 20% of the lowest concentration of interest. The correction for blanks is not recommended. Certified reference materials should be analysed regularly, e.g. DX-1 and DX-2 from NWRI, Canada (de Boer and McGovern, 2001) or WMS-01 from Wellington Laboratories, Canada, lake sediment with lower contamination levels. The laboratory should further prove its competence by regular participation in relevant laboratory proficiency tests. It is essential that the matrix and concentration range of the proficiency testing samples are comparable with the samples routinely analysed within the monitoring programme.

8.   Safety

The chemical compounds dealt with in this guideline are hazardous and must only be handled by trained personnel familiar with the handling of PCDD/F and dl-PCBs, and associated risks as well as precautionary measures. USEPA (1994) recommends that laboratories purchase diluted standard solutions instead of preparing primary solutions. The laboratory staff should be aware that other matrices might contain considerably higher levels of PCDD/Fs and dl-PCBs than marine sediments.

9.   Data reporting

Results are reported in pg/g dw (ng/kg dw). The water and organic carbon content of the samples should be reported as well, the latter being used for normalizing purposes. Concentrations are reported to two significant figures. Minimum performance criteria such as LoQ and measurement uncertainty along with information on blanks and reference materials should be included in the report.

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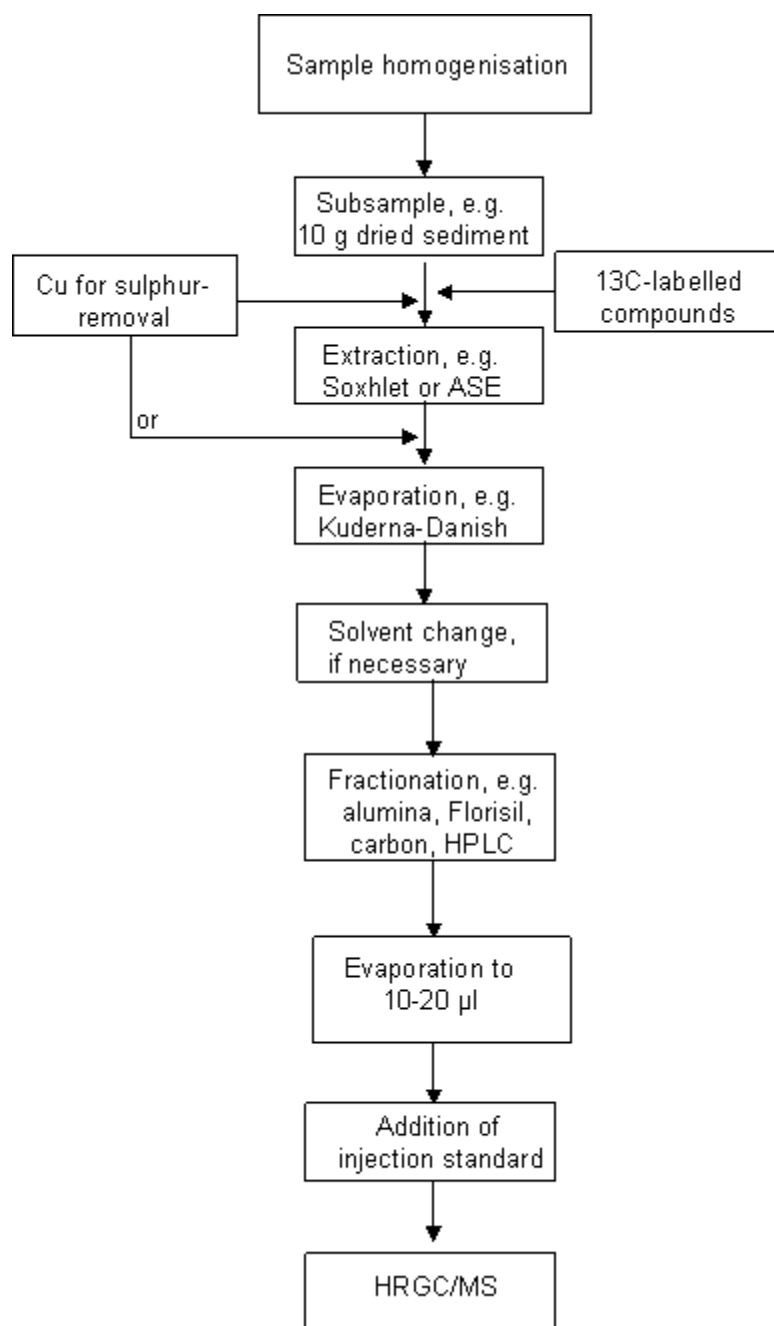
Acknowledgements

Figure 2. Analytical method suitable for analysis of sediment samples within environmental monitoring.

Annex 11: Technical minutes of the Review Group MON1 2010

Review Group: Jarle Klungsøyr (Chair), Jacob de Boer, Michiel Kotterman, Colin O'Dowd*, Pia Andersson*

*Not present at the meeting

Introduction

RGMON1 worked by correspondence and met at ICES in Copenhagen May 3th 2010 to review the work done by four ICES working groups answering requests by OSPAR on:

1. Monitoring methodologies for ocean acidification (2010/2)

2. Atmospheric monitoring of PFOS (2010/6)

3. Tools for coordinated monitoring of dioxins, planar CBs and PFOS (2008/6, 2010/6)

Expert Groups Reports

- Marine Chemistry Working Group Report 2010 (MCWG2010)
- Working Group on Marine Sediments in Relation to Pollution (WGMS2010)
- Working Group on Deep Water Ecology Report 2010 (WGDEC2010)
- Working Group on Zooplankton Ecology Report 2010 (WGZE2010)

2010/2.   Monitoring methodologies for ocean acidification

OSPAR Request

To provide, on the basis of a review of existing methodologies and experience, recommendations for cost efficient methods for monitoring ocean acidification (OA) and its impacts, including possibilities for integrated chemical and biological monitoring. Specifically this should provide:

    a.   advice on appropriate parameters, protocols and quality assurance for monitoring changes in pH and inorganic carbon chemistry in the OSPAR maritime area and other ancillary parameters that should be included in monitoring programmes

    b.   advice on the status of current knowledge of spatial and temporal variability of pH and inorganic carbon chemistry in the OSPAR maritime area

    c.   advice on appropriate spatial and temporal coverage for monitoring, considering different oceanographic features and conditions and key habitats/ecosystems at risk from OA in the OSPAR maritime area,

    d.   advice on the status and maturity of potential indicators of OA impacts on species, habitats and ecosystems that could be considered for inclusion in OSPAR monitoring programmes.

RG Comments

The MCWG 2010 Report with Annex 8 gives general advice on the inorganic carbon chemistry and other physical and biological factors that must be taken into account when studying ocean acidification.

The text is relevant and of generally good quality and gives an overview of existing methods for the chemical analysis of ocean acidification. The report gives general advice to most questions under bullet point a), b) and c) and can be used by the ADG to prepare advice. The language in Annex 8 is of variable quality/standard and some technical editing is needed, particularly so for Section 9.

A general comment by the RG is that natural variation in the euphotic part of the water column is an obstacle/challenge to obtain a sufficient number of data from ship based on discrete sampling to resolve the present trend in ocean acidification. The winter season is less influenced by biological activity, and data from this period might therefore be well suited to determine the present development in long-term trends in ocean acidification. Further development of modelling tools can be useful to better predict future development in ocean acidification.

2010/6 Atmospheric monitoring of PFOS

OSPAR Request

To provide advice on whether it is appropriate to include PFOS in atmospheric monitoring programmes and if other perfluorinated compounds should be included in such monitoring to support assessments of inputs of PFOS to the marine environment.

RG Comments

The MCWG 2010 Report (par. 5.13) is technically correct and describes the current status and expertise of atmospheric monitoring of PFOS and other PFCs. The report is rather straightforward and cautious; the information and analysis techniques are not at a level to give other advice than to improve the knowledge and expertise before adding PFCs to atmospheric monitoring programmes. The scope and depth is appropriate to the request and the text can be used by the ADG. The advice given does answer the request; as for now it is not appropriate to include PFOS or other perfluorinated compounds in atmospheric monitoring programmes.

The RG has some few comments / questions. The report states "At present, no scientific consensus exists whether the atmospheric transport by volatile precursor compounds or the transport by oceanic currents or by means of sea-spray is the dominant transport pathway". Relating to sea-spray, supermicron spray does not travel that far so it is unlikely to be a viable transport mechanism; however, submicron spray, where PFOS may be more enriched as is organic matter in the spray aerosol, has the potential to travel long distances. Given that deposition velocities and transport lifetimes vary by orders of magnitude depending on the aerosol size, knowledge of the potential carrier size is critical. In terms of volatile precursors, they are perhaps of low water solubility and as a result would have low deposition velocities if no reactive chemical sink is present in oceanic waters. It seems that the aerosol-gas phase partitioning is uncertain and, therefore, also the deposition route is not clear.

Prior to proceeding with a measurement programme, the report recommends further method development and capitalizing on research programmes to improve the

knowledge. One suggestion along these lines is to explore PFC data availability from current marine atmospheric monitoring stations such as Mace Head (Ireland), Cape Grim (Tasmania), or Cape Verde. There are likely to be some gas phase PFC measurements at these stations which could support initial budget, transport and deposition estimations. However, it is unlikely that a suitable aerosol dataset from these stations is available for post analysis.

Atmospheric deposition has been measured and modelled in some remote areas (Schenker, 2008) and PFC concentrations in seawater and biota are increasing. However, it is unclear if the observed/ calculated atmospheric deposition (within an order of magnitude) can account for the observed concentrations of PFCs at remote areas. It would be interesting to compare the contribution of PFCs in air with those by ocean currents.

2008/4.   Tools for coordinated monitoring of dioxins, planar CBs and PFOS

OSPAR Request

To prepare the following tools to support the coordinated monitoring of dioxins, planar CBs and PFOS under the OSPAR CEMP:

   a.   technical annexes to the JAMP Guidelines for monitoring Contaminants in Sediments (OSPAR agreement 2002–16) and JAMP Guidelines for monitoring Contaminants in Biota (OSPAR agreement 1992–2) according to the structure of the existing technical annexes covering the following:

   (i)   monitoring of dioxins in biota in sediments, taking into account advice from SIME 2007 that monitoring of dioxins in sediments should only be carried out in specific areas (such as sedimentation areas or estuaries) because of time-lag (10 – 12 years) in deposition of quantities required for sampling;

   (ii)   monitoring of PFOS in sediments, biota and water;

   b.   to review the existing technical annexes on monitoring of chlorinated biphenyls in biota and sediment and propose revisions so that they are adequate for monitoring of planar CBs in these compartments, taking into account advice from SIME that monitoring in sediments should be undertaken only if levels of marker PCBs are e.g. 100 times higher than the BACs and that for biota monitoring of concentrations in seabird eggs could provide an alternative matrix;

   c. to develop background concentrations for dioxins.

RG Comments

Most of the questions in this request have been addressed previous years. The remaining issues are reflected in ToRs for MCWG2010 (c) Finalize technical annex for monitoring PCDD/Fs and "dioxin like" PCBs in sediments (with WGMS).

a (i) *Monitoring of dl-PCB and PCDD/Fs in sediments*

The draft guidelines for monitoring of polychlorinated dibenzo-*p*-dioxins(PCDDs), dibenzofurans (PCDFs) and dioxin-like PCBs (dl-PCBs) in sediment give an updated overview of requirements and possibilities for PCB and PCDD/F analysis in sediment. This is useful, as many developments in technologies have taken place in this

field. The document includes a lot of useful information and it may well serve to guide marine laboratories for analysing dl-PCBs and PCDD/Fs in sediments.

The guideline prepared is of good quality. The RG has some general and some more specific comments which are listed below.

According to SIME (2007) monitoring of dl-PCBs and PCDD/Fs in sediments should take place in specific areas such as sedimentation areas and estuaries. Monitoring dl-PCBs and PCDD/Fs in sediments differs from that in biota because other PCDD/Fs are present in sediment, in addition to the 2,3,7,8-substituted PCDD/Fs that are found in biota. Therefore, more selectivity is needed for sediment analysis. In contrast to biota, the PCDD/F levels – certainly in sedimentation areas – are normally higher. Consequently, the requirements for sensitivity are less stringent for sediments than for biota monitoring. This implies that laboratories do not necessarily need high-resolution mass spectrometry (HRMS). Low-resolution (LR) MS will probably do in most cases. The guidelines should mention this upfront, so laboratories should not be directed to investments which are not essential.

Recently, there has been a substantial development in bench-top mass spectrometers. Not only have LRMS instruments become more sensitive, but new types of mass spectrometers have entered the market. These instruments, Time-of-Flight (ToF) MS and Orbitrap MS fill a gap between LR and HRMS. Often they offer an even higher resolution than HRMS instruments. Orbitrap machines offer a resolution of up to 100 000. This resolution is, however, mass dependent in contrast to HR sector instruments, which normally offer a resolution of 10 000 or more, independent of the mass. A resolution of 10 000 (measured at 10% of the peak height) is needed for a proper dioxin analysis. To compare the resolution of HRMS sector instruments and that of ToF and Orbitrap instruments, the resolution of the latter two should be reduced by ca. a factor 2, as they are measured at 50% peak height. The guidelines should mention the options that ToF and Orbitrap MS offer. They require much less investment than a sector instrument does, while certainly for PCDD/F monitoring in sediments they will offer enough sensitivity and selectivity (full-scan options and high resolution). For more in-depth, academic studies, sector instruments may still be the optimum choice, but they are no doubt much more expensive.

The guidelines should also mention comprehensive-multidimensional gas chromatography (GCxGC) as a new technique for dl-PCBs and PCDD/F analysis. For dl-PCBs and PCDD/Fs in sediments GCxGC can even be used in combination with ECD, as this will offer enough sensitivity, and selectivity is much better than on single-column GC provided a proper orthogonal combination of columns is selected. GCxGC can, however, also be used in combination with LRMS and, even better, in combination with ToF-MS as the latter offers a rapid response, which is ideal for the narrow peaks that come out of GCxGC.

New stationary phases have entered the market, such as liquid crystalline phases and ionic liquids. These are particularly useful to combine with e.g. non-polar columns in GCxGC to achieve sufficient orthogonality. The guidelines should mention polar that are often used in dioxin laboratories, and particularly for sediments, as more congeners are present than the 2,3,7,8-congeners that are present in biota. Table 4 should be extended as there is much more information on possible co-elutants of PCBs, e.g. in the papers of B. Larssen and S. Bøwadt form the 1990s.The section on temperature programmes is very limited. This should be extended. The 'solvent effect' is e.g. not

mentioned at all. This is essential in the selection of the temperature program of the oven. Discrimination effects should also be mentioned.

Nowadays, standards for all dl-PCBs and PCDD/Fs, including all ^{13}C labelled compounds are available. Therefore, the Table 2 is actually redundant. One sentence in the text mentioning these standards are available is sufficient.

The description of the fractionation is very short. This should be improved because as it is now, the text does not give information on the proper order of columns to be used. It should be mentioned that a fat separation e.g. on alumina columns (or GPC but that is less efficient, even when more columns are used in series) is needed first. Then, a fractionation of PCBs, and OCPs is required on e.g. silica gel or Florisil. An extra cleaning step by sulphuric acid may be added (or the acid can be added to the silica column). Finally, the separation of the planar compounds (dl-PCBs and PCDD/Fs from the non-planar PCBs and other non-planar compounds) on a HPLC PYE or graphitized carbon column should take place. For the solvent reduction/evaporation steps the option of a Turbo-Vap should be mentioned.

The various additions should be made with reference to the appropriate literature. The paper can further be improved according to the suggestions given in Annex I.

b. *Review guidelines for PCBs in sediments and biota to propose revisions to make them adequate for dl-PCB and PCDD/F monitoring*

Monitoring planar (dl-) PCBs and PCDD/Fs in biota or sediments require specific guidelines. Guidelines for monitoring non-dl- ('regular') PCBs cannot be used for that. Guidelines for monitoring dl-PCBs and PCDD/Fs in biota have been prepared and are already accepted (ICES Advice report 2009, p.82–99). The guidelines for dl-PCB and PCDD/F monitoring in sediments have been discussed above under a.

c. *Developing background concentrations for dioxins*

PCDDs occur in nature, e.g. due to forest fires. Because these are also unintentionally produced by man, e.g. in waste incinerators, they are transported around the globe. Therefore, nowadays it is difficult to distinguish natural PCDDs from man-made PCCDs. The best option for finding background concentrations of PCDDs is to analyse sediment cores. The sediment layers that date back ca. 100 years ago should present background PCDD levels. MCWG in 2009 has tried to identify pristine areas, but these are difficult to find and little information is available (ICES MCWG Report Annex 17, p. 141). MSWG in 2009 has asked for additional work to collect data. It is suggested that MCWG may be asked to identify literature in which PCDD concentrations in deeper sediment layers (cores) are reported. A few useful papers could be added:

L.O. Kjeller, C. Rappe (1995). Time trends in levels, patterns and profiles for polychlorinated dibenzo-p-dioxins, dibenzofuranes and polychlorinated biphenyls in a sediment core from the Baltic proper. Environ. Sci. Technol., 29: 346–355.

R.E. Alcock, K.C. Jones (1996). Dioxins in the environment: a review of trend data. Environ. Sci. Technol., 30: 3133–3143.

J.M. Czuczwa, F. Niessen, R. Hites (1985). Historical review of polychlorinated dibenzo-p-dioxins and dibenzfurans in Swiss lake sediments. Chemosphere, 14: 1175–1179.

Annex 1: Detailed comments on Draft guidelines for monitoring dl-PCBs and PCDD/Fs in sediments

Title: Add" Guidelines for ….monitoring, etc.

Page 1

Line 3: Being… → As PCDD/Fs are ….strongly

Line 6: Use abbreviation PCDD/F instead of chlorinated dioxin and furans in full

Line 8: add ref. (e.g. WHO, Van den Berg)

Line 15: Daelemans ref. refers to biota, better take ref. referring to sediments, and of more recent date.

Line 17: Neurotoxicity is a typical effect caused by ortho-PCBs but for non-ortho's?

Line 21: Spatial and temporal trend monitoring

Under Figure 1: Add paragraph with 'Objectives'

Analytes: 17 congeners: give arguments and ref. (WHO/EU)

3rd line: should be → are normally; …dry matter and total organic carbon

4th line: last word: toxic → TCDD

5th line: However, although common practice …

Page 2

Line 1: ICES guidelines: unclear: these ones or ref.?

Table 1: Give ref.

Sediments: PCDD/Fs, add: and dl-PCBs

Page 3

Line 2: add ref. on OSPAR JAMP guidelines

Page 3, 3 lines above Table 2: Give a better description of the way of spiking

Anal. Methods., line 5: state-of-the-art

Prep. Steps: Give quantitative indications of purities of solvents and reagents, rather than general statements like 'high purity'.

Cleaning: add enzymatic solution to remove fat rests, even for sediments

Baking: at which temperatures? Also: age of glassware is an issue: when scratched, then more active sites. Use new glassware from time-to time.

Last paragraph above Table 2, line 4: These internal standards are used to correct for errors and recovery losses.

Table 2 can be left out: just mention in text that ^{13}C standards are available for all target compounds

Page 4.

Extraction, line 4: be consequent in use of abbreviations: PCDD/F, dl-PCB, HRMS

Line 5: add ref.

Last sentence: Provided the proper conditions are chosen, …

Page 5

Line 3: mL

Line 9: recommended: replace by: essential

clean up / clean-up? Be consequent in way of writing

Line 17: Ref. Van Leeuwen (2007) should be replaced by

> Loco, J. van, S. van Leeuwen, P. Roos, S. Carbonelle, J. de Boer, L. Goeyens, H. Beernaert (2004). The international validation of bio and chemical screening methods for dioxins and dioxin-like PCBs: The DIFFERENCE project round 1 and 2. *Talanta* 63, 1169–1182.

Page 7

Add comments on software issues, on calibration curve problems (linearity or not).

Above table 7: Provide ref. for ±15%

Page 8

Please give additional comments on problems with ion-suppression and need for extra clean up.

Page 9

Correction for blanks is not recommended.

Safety: do not use crystals for standards; do not analyse fly ash: if desired, an entirely new lab with specific safety precautions needs to be built.

Data reporting: add a comment on sieving of sediment with ref.