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9–13 March 2015

Bergen, Norway

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CIEM Conseil International pour l'Exploration de la Mer

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Executive summary

The Working Group on the Biological Effects of Contaminants (WGBEC), chaired by Bjørn Einar Grøsvik (NO) and Ketil Hylland (NO), met at IMR, Bergen, Norway, 9–13 March 2015. There were 17 attendees through the week representing 10 countries.

This was the third year of the 3-year Terms of Reference for WGBEC. There were 14 items on the agenda. Presentations and discussions took place in plenary, with rapporteur responsibility shared by members of the group. All items on the agenda were completed and are reported.

Respond to requests for advice from the Regional Seas Conventions. WGBEC had received no official requests for advice from OSPAR via the ICES secretariat. WGBEC is aware of and welcomed the increased consideration of biological effects within OSPAR MIME and HASEC. Although widely used in European countries, most of the biological effects data has not been reported to the ICES database. The main reason is the complex reporting requirements and format, ensuring that only some institutions in each country has the capability to report. Future implementation of biological effects methods in OSPAR monitoring frameworks requires data in the database for assessment. WGBEC members will endeavour to submit as much data as possible during 2015 when the next assessment is due. The Chairs will facilitate this process.

WGBEC have been in contact with other expert groups, WGEEL, WGPDMO, MCWG and WGMS in relation to taking forward areas of common interest. WGBEC, WGMS and MCWG met concurrently at ICES HQ in 2014. The joint sessions focused on ocean acidification, marine litter and passive samplers/dosing. A Workshop has been suggested between WGBEC and WGEEL (WKBECEEL), to be held in Norway in January 2015.

Consider emerging issues of scientific merit and address knowledge gaps (in relation to the ICES Science Plan). Over the three year period, members have presented updates on oil toxicity to early life stages of fish, risk assessments, toxicity studies on nanoparticles, studies of effects of pharmaceuticals used to combat salmon lice in aquaculture, vitamin A interactions with contaminant effects and thiamine deficiency in wild life.

Review status of publications and consider requirements for new publications. WGBEC reviewed status with ICES TIMES manuscripts and revised deadlines as appropriate. New and partially finalised manuscripts were available as draft for the meeting to consider. The Chairs have communicated with the WGBEC TIMES editor Ricardo Beiras and ICES Secretariat on progress with the manuscripts whenever required.

AQC activities for biological effect methods including harmonisation activities initiated from WGBEC and within OSPAR, HELCOM and MEDPOL maritime areas. Results from an EROD and a PAH bile metabolite intercalibration exercise organised by Cefas, UK (analyses performed in 2013) were presented and discussed. The group discussed the necessity of keeping such exercises running on a regular basis and one of the Chairs (Ketil Hylland) assumed responsibility to follow up on this. No information was available concerning activities in the HELCOM area. A LMS neutral red exercise was held in Sweden in 2013 with predominantly Nordic participants, but also WGBEC member Concepcion Martinez-Gomez.

Respond to requests for advice from the ICES Data Centre. Requests from the data centre were addressed during meetings all three years. There is a continuing need for updating fields and data reporting requirements. Representatives from NMD (Norwegian Marine Data Centre, IMR) were invited to the 2015 meeting to discuss how they could assist on these matters. WGBEC members have been identified as responsible, i.e. contact persons, for different biological effects methods.

National monitoring programmes. Presentations and reports for the 2015 meeting were provided by Juan Bellas (ES), Jakob Strand (DK), Ulrike Kammann (GE), Thierry Burgeot (FR), Steven Brooks (NO) and Lars Förlin (SE). Regular reports have been provided for most countries with active WGBEC membership over the three-year period. An overview paper will be prepared for the 2016 meeting.

Development and harmonisation of methodologies for marine monitoring. Assessment Criteria for biological effect techniques within the OSPAR SGIMC approach have been reviewed and revised when and where deemed necessary.

Novel and emerging compounds (e.g.) pharmaceuticals, recreational drugs, biocides and discharges from mining. A review on pharmaceuticals in marine ecosystems involving former WGBEC members has been published. Reviews on early life stage toxicity from oil on fish and on immunotoxicological methods suitable for monitoring will be prepared prior to the 2016 meeting. Other issues that will be followed up in 2016 are toxicity of nanoparticles, as well as neurodevelopmental and behavioural effects.

To evaluate the results of marine litter monitoring and research activities, especially microparticles (plastic/non plastic) and associated chemicals. A joint meeting with MCWG was held on plastics in 2014. The 2015 call by JPI-Oceans on occurrence, analytical methods and ecological effects of microplastics was discussed during the 2015 meeting. Several of the participating members were involved in proposals for this call.

1 Administrative details

Working Group name

Working Group on the Biological Effects of Contaminants

Year of Appointment

2013

Reporting year concluding the current three-year cycle

3

Chair(s)

Bjørn Einar Grøsvik, Norway

Ketil Hylland, Norway

Meeting venue(s) and dates

10–15 March 2013, San Pedro del Pinatar, Spain

3–7 March 2014, ICES Headquarters, Copenhagen, Denmark

9–13 March 2015, Bergen, Norway

2 Terms of Reference and Summary of work plan

ToR	Description	Background	Science Plan topics addressed	Duration	Expected Deliverables
a	Respond to requests for advice from Regional Seas Conventions (e.g. OSPAR, EU) as required.	*Advisory requirement*. WGBEC has a history in its ToR of responding to requests from OSPAR and these have always been considered as a priority and importance by the EG. In addition, there is a wide breadth of knowledge and expertise which allows the EG to respond in an informed manner to these requests.	Advice to ICES	Annual 2012-2015	Each year advice is reported to ICES secreatariat for onward transmission e.g. to OSPAR
b	Consider emerging issues of scientific merit and address knowledge gaps (in relation to the ICES science plan). - Oil toxicity to early life stages of fish	*Science and advisory requirement* In reviews over the past three years WGBEC has considered emerging special scientific issues in relation to biological effects	112, 172, 241, 242	2012-2015	Review paper published in the peer review literature (2015)

	- Ocean Acidification - Immunotoxicity - Novel monitoring techniques (e.g. 'omics technology) - Thiamine defficiency in marine wildlife (2014 only)	and contaminants and also in relation to the ICES Science Plan These topics have been selected as of current concern.			
c	Review status of publications and consider requirements for new publications - ICES TIMES - Other ICES publications - peer review publications	*Science and advisory requirement.* It is important for WGBEC to keep track of publication progress with biological effects methods it has considered useful for monitoring. Protocols are needed for national and international programmes as well as monitoring to met OSPAR and EU MSFD obligations.	Advice to ICES	Annual 2012-2015	Publication of ICES TIMES methods for marine monitoring purposes
d	Conduct assessment of data as required -Quality assurance data from method intercomparison trials - Integrated assessment of monitoring data (and advise on procedures to other groups as appropriate)	*Science and advisory requirement* AQC is vital to support, report and assess data, particularly for cross maritime areas and developments and harmonization in this area need to be taken forward in a coordinated manner.	123, 241, 242, 244	2012-2015	Report each year via ICES secretariat to OSPAR on progress with AQC initaitives / schemes for biological effect methods. Report to ICES data centre on current AQC programmes.
e	Respond to requests for advice from the Data Centre	*Advisory requirement* Biological effect data are increasingly being submitted to the ICES database and technical queries arise. WGBEC can assist with answering queries from the ICES Data Centre.	Advice to ICES	Annual 2012-2015	Provide support and information to ICES data centre that can be used to facilitate submission of biological effects data to the ICES database
f	Development and harmonization of methodologies for marine monitoring and surveillance including: - Integrated assessments -Environmental risk assessment	*Science and advisory requirement* WGBEC has found it of value to discuss, feedback and support national monitoring programmes across the maritime areas and this is a valuable opportunity to	241	Annual 2012-2015	Report via ICES secretariat to OSPAR on annual review of assessment criteria for JAMP biological effects and progress with the application of

	-Review and develop assessment criteria for biological effects methods - Report on national monitoring programmes for biological effects	improve and harmonize programme designs and assessment of data (e.g. OSPAR / MEDPOL / WFD / HELCOM/ EU MSFD)			the OSPAR SGIMC integrated approach. Report to ICES data centre on current AQC programmes. Link up with MCWG and WGMS on integrated approach and assement of data.
g	Address issues in relation to novel and emerging contaminants (e.g. pharmaceuticals, nanoparticles, toxicity of mixtures etc.) -Pharmaceuticals and recreational drugs in the marine environment. -Biocides in the marine environment.	*Science requirement* These are two issues identified by WGBEC that are of value and special scientific interest to understanding the effects of contaminants in the marine environment. Information on environmental impacts is currently lacking.	123, 172, 242, 241	2012-2015	Provide report to ICES on these special scientific issues and publish in the peer reviewed literature (year 3)..
h	To evaluate the results of marine litter monitoring and research activities, especially microparticles (plastic/non plastic) and associated chemicals: -Status on monitoring protocols for marine litter in biota -Marine litter research outcomes and results of impact assessments on key marine organisms. Evidence of bioaccumulation, toxicity and adverse physical, biological and chemical effects of microplastics and associated contaminants on a range of marine organisms, populations and communities. -Evidence of transfer of microplastics and associated contaminants through marine food chains.	*Science and advisory requirement* There has been considerable interest over the past two years on the biological effects of plastic particles, particularly in relation to contaminants associated with plastic particles. It is important that this work area is reviewed and any reports and feedback from other Expert Groups considered by WGBEC.	241, 243, 344	2012-2015	Review and report to ICES on how this work area is developing and identify how this may be progressed and applied to marine monitoring programmes. Link up with other EGs with intetrest in this topic i.e. MCWG and WGMS (planned for 2014) Publish outputs in peer review literature (Year 3).

Year 1	Subgroups were identified to respond to requests to other expert groups and to develop scientific reports to answer ToR of the requested themes. Reporting of TOR will be completed as soon as possible in readiness for final 3 yr report.
Year 2	Requests for advice from ICES, OSPAR and requests for support from data centre will be addressed each year as appropriate. Time allocation is variable depending on the task and preparation required pre the meeting and reporting post meeting.
Year 3	Requests for advice from ICES, OSPAR and requests for support from data centre will be addressed each year as appropriate. Time allocation is variable depending on the task and preparation required pre the meeting and reporting post meeting. Complete and sign off 3 yr report and report on publication outputs.

3 Opening, agenda, rapporteurs

The ICES WGBEC 2016 meeting was hosted by IMR, Bergen, Norway. The meeting was opened by a 'tour de table' to introduce group members, their affiliations background and science interests. 17 participants were present at the meeting through the week, representing 10 countries: France, Germany, Norway, Spain, Sweden, Denmark, Iceland, USA, UK and Portugal. The list of attendees is given in Annex 1.

The provisional agenda for the 2015 meeting was approved by the group (Annex 2). The meeting started with a plenary presentation on thiamine deficiency by Lennart Balk (Sweden) for the group and IMR staff. He elaborated on this subject for the WG and the topic was discussed by the group.

On Thursday, WGBEC had an excursion to IMR's Research station at Austevoll where the group was informed on ongoing research activities. Sonnich Meier (IMR) described ongoing experiments on haddock egg and larvae exposure to crude oil. Caroline Durif (IMR) had a presentation for the group where she informed on ongoing work in WGEEL and on her own results with studies on magnetic field orientation in eel.

Rapporteurs for the agenda items were selected. This meeting was the last meeting with the present ToRs. The group expressed a wish to proceed for a new period and discussed themes and agenda points that are elaborated on and which subjects or questions that the group would recommend for the next ToRs (Appendix 3).

The group expressed interest to have a common workshop with members of WGEEL on how contaminants may explain the population decline of eel.

4 Respond to requests for advice from the Regional Seas Conventions (ToR a)

4.1 Review of the EU Marine Strategy Framework Directive Commission Decision document (2010/477/EU) for Descriptors 8 and 9

Craig Robinson (UK) informed WGBEC on ongoing work with descriptors 8 and 9 within MSFD. The aim of the MSFD is to achieve Good Environmental Status (GES) by 2020. It requires Members States to define GES, establish monitoring programs, set targets to achieve and measures by which those targets can be achieved. It is therefore necessary that when targets are not met, measures effective to obtain GES can be identified.

In 2014, the European Commission began a process to review the Commission Decision document on criteria and methodological standards on good environmental status of marine waters (2010/477/EU). For Descriptors 8 (*Concentrations of contaminants are at levels not giving rise to pollution effects*) and 9 (*Contaminants in fish and other seafood for human consumption do not exceed levels established by Community legislation or other relevant standards*) this review process is being led by the Joint Research Centre in Ispra, with support from the Expert Network on Contaminants (for members, see Annex 4). The output of the review should be a new Decision document that is clearer and simpler than the existing one, including minimum requirements, and is coherent with other Community Legislations.

Following the first meeting of the Expert Network (July 2014), draft report templates for D8 and D9 were circulated for Expert Network comments before the JRC presented progress to the EU Working Group on Good Environmental Status (WG-GES) in October 2014. A second Expert Network meeting was held in February 2015 and, following a further round of comments, the JRC will submit its final report to WG-GES by end March 2015. Progress is communicated within the Expert Network using CIRCABC and with the public via the MSFD Competence Centre website (mcc.jrc.ec.europa.eu). The public website hosts the MSFD Task Group reports, the in depth assessments and other related documents; it includes links to EU funded projects of relevance to each of the MSFD Descriptors. At the February network meeting the following points were agreed:

- The substances that are required to be monitored for D8 are the WFD Priority Substances, except any that are not relevant to the marine environment, with the possible addition of Marine Specific Pollutants, to be guided by the Regional Seas Conventions. A minimum list of substances should not be specified, rather a common de-selection mechanism is required in order to harmonise approaches between different Regions;
- Radionuclide assessments will be required for D8;
- Water is not recommended as a monitoring matrix; rather, monitoring should take place using sediment and biota;
- Biological Effects should be considered for D8, using the Regional Sea Conventions' approaches, e.g. OSPAR Common Indicators or HELCOM CORESET;
- The minimum requirement for monitoring under D9 are those substances listed in the EC. Food Regulations; pathogens (e.g. *E. coli*) are not considered under D9.

After the review, any revision of the Commission Decision document will have long-term implications, and will affect the next round of MSFD monitoring plans and not the current (2014–2020) reporting cycle.

Currently, only one biological effect is accepted as a Common Indicator by OSPAR, i.e. imposex in gastropods. HELCOM additionally include PAH metabolites in bile. Within OSPAR, several biological effects are considered as Candidate Indicators, including EROD, lysosomal membrane stability, external fish disease and bile PAH metabolites; HELCOM have a similar list of candidates, plus additional ones on fish (eelpout) and amphipod reproductive success. MEDPOL also consider Biological Effects as important, but their use is not widely implemented in MSFD monitoring (except by Spain).

Within OSPAR, the process to elevate techniques from Candidate to Common Indicators requires that they meet various OSPAR criteria: there must be protocols in the OSPAR system, QA/QC procedures in place (including inter-laboratory comparisons) and assessment criteria (background values and EQS-like harm-related EAC thresholds). To promote Candidate Indicators to Common Indicator status, the OSPAR MIME Working Group must be convinced to recommend to their parent committee (HASEC) that the effects methods meet the suitability criteria and then, at HASEC, a significant number of Contracting Parties must agree to conduct the monitoring.

In HELCOM, the criteria that indicators need to meet to be considered fully operational include a well-developed concept and GES-boundary, monitoring being in place and updating procedures having been agreed. For adaptation, indicator reports are being made available for national consultation by end March 2015. The indicators will be reviewed from a technical point of view by HELCOM WG State & Conservation and WG Pressure groups in mid-May, after which WG Gear will evaluate them from a strategic point of view. After this review, a list of indicators will be put up for adoption at the HELCOM Heads of Delegation meeting in June 2015.

WGBEC took note of the current status of biological effects within MSFD and highlighted the need to make data available for OSPAR MIME assessments (see section 4.2).

4.2 OSPAR

WGBEC have had continuous contact with OSPAR groups, particularly MIME and HASEC, directly and indirectly through the three-year reporting period concerning the use of biological effects in OSPAR programmes. WGBEC members Thierry Burgeot (FR) and Thomas Maes (UK) have attended MIME meetings throughout this period and Burgeot presented the integrated assessment of biological effects and contaminants in the North Sea (ICON programme) as a demonstration of the SGIMC integrated approach (2012) as well as a pilot activity including other data sets (2014).

WGBEC has reviewed OSPAR MIME reports and has welcomed the increased consideration of biological effects. OSPAR MIME had identified a need to reduce the size of the suite of biological effect techniques in the SGIMC integrated approach and had attempted to select some techniques as ‘Common indicators’. WGBEC is however of a strong opinion that reduction of the SGIMC suite of methods will severely reduce the possibility to detect effects of contaminants in marine ecosystems. WGBEC is also concerned about the process that has resulted in the selection of biological effects methods as ‘Common Indicators’. The methods that were selected by OSPAR have not been considered to represent a relevant and robust set of methods to detect effects of contaminants. Any reduction in the number of techniques used for monitoring purposes either within the SGIMC integrated approach or as choice indicator techniques should be evidence-based with clear rationale. In addition, there is a need for OSPAR to decide on a monitoring design strategy i.e. the application of the integrated approach, deployment of any techniques, with regard to targeted application, frequency of monitoring, statistical aspects of designing a monitoring programme and techniques for combining assessments across regional scales. WGBEC has offered its expert advice in this area should the need arise.

Although widely used for monitoring purposes in European countries (see section 9.4), a large proportion of the available biological effects data has not been reported to the ICES database. The main reason has been complex reporting requirements and format. Future implementation of biological effects methods in OSPAR monitoring frameworks requires an availability of data for assessment. The Chairs have facilitated a process through the reporting period to aid WGBEC members to submit biological effects data to ICES, but this is not completed.

During the 2015 WGBEC meeting, Thierry Burgeot (FR) presented a report on 'pilot activity' concerning testing the integrated guidelines and practical application of biological effects monitoring techniques, including the issue of enhanced access to biological effects measurement data and their use by Contracting Parties (CPs). France demonstrated the results of an application of the integrated monitoring to HASEC using a French case study with a theoretical example of the integrated process. Because of a limited submission of biological effects data to the ICES data-base, a roll-over could not be done during the MIME meeting in 2014. France suggested to OSPAR that inclusion of integrated monitoring in the roll-over could be partially reached during the MIME 2014. It is important to note that species- and contaminant-specific EACs and BACs are now available for the selected biological effects methods (SGIMC 2011) and allow a generalised interpretation of biological effects in the northeast Atlantic Ocean.

France will lead a trial of the integrated guidelines with whatever data were available. The intention would be to identify a way forward with available data. MIME agreed that France would provide an Excel template for Contracting Parties and other experts (e.g. WGBEC) to populate directly with biological effects data, with a view to a rapid application of the integrated approach for presentation to HASEC 2015. MIME also agreed that the UK would consider inclusion of integrated monitoring of contaminant and biological effects data in the online assessment tool.

At the WGBEC 2015 meeting, the report sent to OSPAR for the HASEC 2015 meeting was presented. The primary conclusion from the simple assessments is that it is clear that biological effects and associated contaminants datasets exist, are accessible and sufficiently homogeneous for the completion of an integrated assessment. CPs should be invited to implement harmonised biomarkers/bioassays/contaminant analyses according to a common set of core methods suggested by SGIMC (2011). Such harmonisation is already under way and is required for a spatial comparison of contaminant impacts in marine areas.

Conclusions/recommendations from the MIME report 2014 relevant to WGBEC were:

- A core set of biological effect techniques has been recommended by SGIMC following a comprehensive process. These recommendations were further taken forward by the HELCOM CORESET Programme.
- Although not all biological effect data can be uploaded to the ICES database due to issues with reporting formats, a wide range (spatial plus inshore versus offshore) data exists and can be included in an integrated approach.
- A simplification of the ICES format 3.2 during the initial phase of the integrated approach should presumably facilitate improved data submission.

- While a variety of data exist, not all core contaminants, biomarkers and bioassays put forward by SGIMC have been included in the monitoring programmes of different CPs.
- The SGIMC integrated approach can provide an overall ecological assessment based on a selection of contaminants, biomarkers and bioassays.

Action: All WGBEC members need to submit biological effects data to the ICES data centre, if required through the assistance of other institutions (contact the Chairs).

In the future, WGBEC should consider whether it is feasible to recommend a minimum set of effect methods.

4.3 Response to other expert groups

Joint workshop of the Working Group on eel and the working group on Biological Effects of Contaminants (WKBECEEL)

Area of mutual interest for WGBEC and WGEEL: are contaminants in eels contributing to their decline?

WGBEC and WGEEL in their 2012 reports identified the possibility to collaborate intersessionally with over-reaching questions as a ToR.

WGBEC identified the following over-reaching questions:

a) What are the concentrations of contaminants in eel populations and have they changed in recent years? To include “traditional” and/or “emerging” contaminants.

b) Are these contaminants at concentrations likely to cause harm and contribute to decreasing eel populations via impacts on reproduction and quality of offspring including endocrine disruption?

c) Are contaminants in conjunction with other factors (e.g. lipid metabolism) impairing the survival, fitness and reproductive capability of eels?

d) Are there tools that can be developed to measure the effects of contaminants in a non-destructive manner?

e) Can experiences / data from other species stand as a model for what goes on in the eel?

Recommendation: WGBEC have identified a sub-group who are prepared to contribute to this collaboration:

Dick Vethaak (NL), Ulrike Kammann (DE), Katja Broeg (DE) and Jakob Strand (DK), Sonnich Meier (NO), Lennart Balk (SE) and Ketil Hylland (NO).

Caroline Durif (WGEEL) and Bjørn Einar Grøsvik has been asked to chair a joint workshop of the Working Group on Eel and the Working Group on Biological Effects of Contaminants (WKBECEEL). The Workshop will be held at Solstrand outside Bergen, 25–27 January 2016. See Annex 6 for ToRs for the workshop.

5 Consider emerging issues of scientific merit and address knowledge gaps (in relation to the ICES Science Plan); (ToR b)

5.1 Oil toxicity to early life stages of fish

In 2013, WGBEC received a request from Tracy Collier (USA) to consider recent studies demonstrating phototoxicity of bunker fuel combined with the field work after the Cosco Busan spill. This may represent the strongest case yet for phototoxicity in a field setting. This appears to be associated with bunker fuel exposures, and not with crude oil. WGBEC was asked to address the findings that very low ppb levels of tricyclic PAHs, found in weathered oils, are embryotoxic and consider the implications for altering eg OSPAR EACs on the information available thus far, and what types of information would be useful to strengthen the case more.

Developing fish embryo and larvae are highly sensitive to different types of PAHs, and this toxicity is dependent on oil composition, weathering and photosensitization. Environmental Assessment Criteria for PAHs are presently scarce and a better resolved data set of EACs should be elaborated for the oil toxicity to early life stages of fish based on the recently published work. WGBEC appreciate the initiative from Tracy Collier and would be interested in suggestions for EACs of PAHs in water and egg/larvae from Collier and colleagues or other researchers in this field. Such data would be highly valuable for risk assessments of oil exposures to early life stages of fish.

Considering the information provided above and discussions WGBEC decided to follow the approach below to progress this ToR before the next 3-year period.

1) Review suitability of existing assessment criteria for hydrocarbons in light of new toxicity data to larval fish;
2) Identify uncertainties and knowledge gaps and place these in context of environmental risk assessment framework;
3) Account for photooxidation and risk factors relevant to life history and ecology of sensitive species such as exposure to surface micro-layers;
4) Produce a review with appropriate recommendations for environmental assessments.

Point 2 in the action list have been addressed in the published articles listed below and B. E. Grøsvik (NO) informed about these studies under agenda 9.2 (see below).

The other points from the action list will be discussed intersessionally by Bjørn Einar Grøsvik (NO), Ketil Hylland (NO), Ulrike Kamman (GE), Sonnich Meier (NO), and North American colleagues will be invited to contribute to this process.

One of the Chairs, Bjørn Einar Grøsvik (NO) informed on on-going research projects at IMR on effects of crude oil and PAHs on early life stages and juveniles of haddock, being performed at IMRs field station at Austevoll. A project financed by the Norwegian Research Council titled “Assessment of long-term effects of oil exposure on early life stages of Atlantic haddock using state-of-the-art genomics tools in combination with fitness observations” started in 2014. For this project, haddock eggs and larvae were exposed to dispersed crude oil continuously or by pulse. Another exposure was performed on juvenile haddock in 2014, funded by Statoil, titled ”Comparative DNA damage and long-term health effects in juvenile haddock after exposure to sediment or produced water associated PAHs." For this project PAHs were given through pellets with PAH profiles similar to produced water, weathered crude oil or pyrogenic PAH. Both projects aim to give more data related to effect parameters after exposures of haddock to oil and PAH components, in addition to more data for setting environmental assessment criteria. Some of the WGBEC attendees expressed interest in collaborating on effect studies on material from the lab exposures.

Action: Prepare a review paper on early life stage toxicity of oil on fish.

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5.2 Ocean Acidification

Kris Cooreman (BE) had participated in ICES SGOA (Study Group on Ocean Acidification) and provided an overview of the area for WGBEC.

Despite that ocean acidification has been recognized as a global problem that may threaten marine ecosystems and the services they provide over this century, research on impacts is still in its infancy.

In 2014, an ICES document was prepared following a special OSPAR request for advice on monitoring methodologies for ocean acidification (ICES, 2014). In summary, the measurements of two, and preferably three, of the following physical parameters should be monitored: total alkalinity (TA), dissolved inorganic carbon (DIC), partial pressure of CO2 (pCO_2) and pH. All waters from estuaries, shelf seas and ocean mode waters and abyssal waters where sensitive ecosystems may be, should be covered. Other programs also recommend additional measurements of temperature, salinity, oxygen and nutrients critical to primary production (NRC, 2010).

Despite the clear and concise knowledge and monitoring guidelines on the carbonate system, very little is known about the biological impacts on organisms, their populations, and communities. These knowledge gaps urged WGBEC to develop a strategy to strengthen the biological impact research and assessments.

There has been an exponential increase in the number of publications on biological effects of OA and several recent reviews have covered this topic. The importance of the combined, and frequently interactive impacts of multiple stressors (such as temperature, low oxygen and pollutants) is now recognised, also the potential for multi-generational adaptation. Experimental research confirms that survival, calcification, growth, development and abundance can all be negatively affected by acidification, but the scale of response can vary greatly for different life stages, between taxonomic groups and according to other environmental conditions, including food availability. Volcanic CO_2 vents can provide useful proxies of future OA conditions allowing studies of species responses and ecosystem interactions across CO_2 gradients. Studies at suitable vents in the Mediterranean and elsewhere show that benthic marine systems respond in consistent ways to locally-increased CO_2. At the shelf-edge, the ongoing shoaling of carbonate-corrosive waters (with high CO_2 and low pH) threatens cold-water corals, in particular *Lophelia pertusa*, in the North East Atlantic. These reefs are rich in biodiversity but we have a poor understanding of their functional ecology, and their responses to the combined effects of future ocean acidification, warming and other stressors.

The greatest effects of projected ocean acidification on zooplankton are likely to be on shelled pteropods ('sea butterflies') in the Arctic and sub-Arctic. Responses by phytoplankton may be positive, negative or neutral; some calcified species show evidence of multi-generational adaption under experimental conditions. Community-wide planktonic responses to future ocean acidification are likely to be strongly influenced by competitive interactions, that might involve nutrient/food availability and predation, as well as by other environmental changes in a high CO_2 world.

The identification of a limited range of species for monitoring the biological effects of OA (with associated description of appropriate morphological, biochemical or other metrics that can be used to document OA impacts) is currently considered premature. As a first step, SGOA prepared a table of potential indicator taxa and possible quantitative metrics for OA responses. Shell erosion in pteropods (planktonic molluscs) is a potential indicator for the occurrence of low saturation state and its biological consequences, but given the morphological diversity of pteropod shells, identification of suitable species for the OSPAR area and associated metrics are required. One species *Limacina helicina*, is particularly promising as a potential indicator because it has a broad distribution within the OSPAR region and a number of studies report its sensitivity to OA. In the absence of specific species guidelines at present, SGOA recommends that collections are made of a broad suite of species from taxonomic groups likely to be sensitive to OA, and that such samples are archived (without compromising carbonate structures). This archive will serve as a repository of specimens that can be retrospectively examined for evidence of OA responses once appropriate indicator metrics are developed.

ICES (2014) suggested criteria for selection of suitable species for such monitoring. While pteropods and foraminifera contain taxa that were considered potentially suitable, the responses of coccolithophores to OA, particularly *Emiliania huxleyi*, appear to be strain-specific, rendering them unsuitable as indicators at this time. Cold-water corals such as *Lophelia pertusa* are species of high conservation concern and identification of areas where they are currently abundant is important. To this end, new habitat suitability index models developed by NOAA for cold-water corals in the North West Atlantic and Gulf of Mexico may prove useful when adapted to the OSPAR area.

Most of the knowledge on ocean acidification is gathered in recent years. This is reflected by the growing number of reports, papers, reviews and integrated science strategies for OA monitoring, research, impact assessments and networking initiatives that emerged in the literature.

OA is recognized as a global problem covering a wide range of physical factors and all waters from estuaries, shelf seas, ocean mode waters and abyssal waters which grants the problem the status of extreme complexity. The research on the impacts of OA is expanding rapidly (hundreds of studies since 2010) but the global geographical (local and regional scale, water depth, upwelling regions) and temporal (diurnal and seasonal) conditions, physical and biological processes, evolutionary adaptation and terrestrial influences mostly led to case by case information on and substantial intra- and interspecies variability of effects of OA on responses such as impaired metabolism, calcification, ion transport balance, acid/base regulation, growth, reproduction and mortality in a wide range of taxa and population and community responses in the entire food chain. It is generally assumed that coastal ecosystems experience greater variability than open ocean ecosystems. Yet several excellent reviews (Fabry *et al.*, 2008; Kroeker *et al.*, 2010; Branch *et al.*, 2013; Nikinmaa, 2013) synthesized the results of hundreds of studies examining biological responses to OA and revealed differences in sensitivities of biological responses of a species or species within and across taxonomic groups. These variations in sensitivities of responses have been described comprehensively in the reviews. Several of these reviews made use of meta-analyses combining results of numerous individual experiments with the ultimate goal to identify key patterns in the impacts that may lead to a better understanding of the subject (Kroeker *et al.*, 2010; Kroeker *et al.*, 2013; Dupont *et al.*, 2010, Hendriks *et al.*, 2010). These achievements are crucial to improve understanding of the current variability and to develop models for straightforward and accurate impact assessments and future impact projections.

Altered carbonate chemistry directly affects the deposition and dissolution rates of the $CaCO_3$ used for structures in e.g. corals, plankton such as coccolithophores, foraminifera (Gattuso & Buddemeier 2000; Riebesell *et al.* 2000; Marubini *et al.* 2003; Royal Society, 2005; Ridgwell and Zeebe, 2005; Orr *et al.* 2005; Hoegh-Guldberg *et al.* 2007), bivalves (Michaelidis *et al.* 2005), echinoderms (Kurihara *et al.* 2004; Shirayama & Thornton 2005). Impairment of the calcification processes is seen as the primarily sensitive response and calcifying organisms to build shells, plates and skeletons are expected to be more sensitive than organisms that do not calcify. The sensitivity varies with the solubility of the mineral forms of their $CaCO_3$ skeletons. Calcifying organisms that produce the calcite form of $CaCO_3$ (coccolithophores, foraminifera, crustacean, echinoderms, some non-pteropods) are expected to be less vulnerable to OA than organisms that construct aragonite structures (e.g. warm and cold water corals, pteropods and other non-pteropods) (Royal Society, 2005). In between are e.g. mollusks that construct shells consisting of layers of either all aragonite or inter-layered aragonite and calcite and organisms such as sea urchins, sea stars and brittle stars that form magnesium-enriched calcite structures. Magnesium enriched-calcite at magnesium concentrations higher than 12% is more soluble and sensitive to OA than aragonite. Magnesium calcite at magnesium concentrations lower than 4% is less soluble than aragonite with calcite, being the least soluble of the three. There is thus a wide range of sensitivities of vulnerability of calcifying organisms to OA depending on their mineral forms of $CaCO_3$. Calcareous benthic algae precipitate either high magnesium calcite or aragonite and perform an important function in the construction of the skeletons of corals.

In addition, organisms that have no or a partial compensation mechanism to maintain the acid/base balance of their body fluids are potentially more vulnerable than organisms that have evolved a regulatory system. Examples of organisms that lack a developed regulatory system are mussel, crab and sea urchin.

Reduced calcification is demonstrated in response to increased pCO_2, decreased pH and carbonate ions and the $CaCO_3$ saturation state. It is, however, generally accepted that the calcification rate is controlled by the saturation statuses of the $CaCO_3$ sources aragonite (Ωarag) and calcite (Ωcal). In regions where the saturation status of aragonite or calcite is higher than 1.0, the formation of shells and skeletons is favoured. Seawater is corrosive to $CaCO_3$ at saturation values of less than 1.0. The saturation index might thus be useful as a suitable indicator to determine the calcification condition of an area and could serve consequently as a warning signal for safe or stressful situations. However, the knowledge on calcification processes in corals, coccolithophores and foraminifera suggest a reduction in calcification by 5–25% in response to a doubling of atmospheric CO_2 from pre-industrial values (from 280 to 560 ppm CO_2; Feely *et al.*, 2004) and this reduced calcification rate would also occur even when the carbonate saturation level was well above one (*Ibid*.).

Marine invertebrates like molluscs and crustaceans show negative responses to acidification at various life stages. Especially the early life stages of calcifying organisms are potentially more vulnerable. The severity of the impact is here again not universal across the taxonomic groups.

WGBEC developed the following strategy for the remainder of its 3-year ToR:

1) To review the existing literature for recommendations on suitable species/ endpoints for monitoring;
2) To focus efforts on those parameters relating to the expertise of WGBEC (endpoint measurements in individuals / populations rather than e.g. biogeographic trends etc.);
3) To account for in-combination effects with other climate change variables (e.g. carbonate chemistry changes and temperature).

Action: To summarise developments in annual reports.

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5.3 Immunotoxicity

Environmental immunotoxicology was discussed by WGBEC in 2011, 2012 and 2013, based on reviews by Tom Hutchinson (UK), Andrea Johnson (US) and a presentation by Ketil Hylland (NO), respectively.

The aim of the group over the 3-year period is to develop or identify methods by which to assess environmental immunotoxicity in marine ecosystems.

The 2013 WGBEC meeting identified Johan Aerts, Dick Vethaak and Ketil Hylland as members to be involved in the continuing work on immunotoxicology, in addition to members who have previously shown interest (Tom Hutchinson, Andrea Johnson). The 2014 WGBEC meeting discussed alternatives for developing this issue further and agreed on limiting our efforts to methods applicable for monitoring. Lack of progress by 2015 led WGBEC to decide to leave the issue for the time being.

5.4 Neurodevelopmental and behavioural effects

One of the Chairs, Ketil Hylland (NO), introduced the topic with a couple of papers showing the potential for early exposure to toxic substances, in this case methylmercury, to affect behaviour years later in a fish species (Fjeld *et al.*, 1998) and for a pharmaceutical to affect behaviour in perch at an environmentally relevant concentration (Brodin *et al.*, 2013).

5.5 Nanoparticles

Joachim Sturve (SE) introduced the topic and informed on experimental work with silver nanoparticles (AgNP) on fish compared with exposure to silver (Ag^+). Responses were studied with molecular methods, i.e. gene expression and proteomics.

Nanomaterials and nanoparticles (NPs) are rapidly becoming an important part of new technology. NPs are today used in a variety of products, from IT to pharmaceuticals and body care products. NPs may also be part of future intelligent solutions for new environmental friendly technologies. However, very little is known about the toxicity of NPs and their effects in the aquatic environment. Compared to molecular chemicals tested, the NPs are quite complex due to their physico-chemical properties, and new integrated strategies are necessary to elucidate their toxicity. NP behaviour builds primarily on physical forces between particles and not on thermodynamic equilibria. One factor that effects the particle interaction is the salinity of the media and higher salinity leads to higher rate of agglomeration of the NPs. It has therefore been suggested by several researchers that NPs are not of major concern in marine environments, since the NP will agglomerate and end up in the sediments. However, NPs are possible sources of toxic compounds in the marine environment.

Silver NPs are of main concern regarding ecotoxicological effects due to the known toxicity of silver ions, and silver ions are toxic to several aquatic organisms such as algae and daphnia. Silver is also toxic to fish. Recent studies show that the levels of silver ions in sewage treatment effluent recipients have increased and in some case reach levels close to LC_{50} for several algae species. It is believed that this increase is connected to an increase in the use of silver NPs. Silver NPs are today the most extensively used inorganic metallic nanoparticles and every fifth nano-product in the market is expected to contain Ag NPs.

A large part of these products have the potential to release silver into the aquatic environment, often through sewage treatment plants.

Even though NPs are considered not to be a major threat to the marine environment it is important to follow the development of the research field. NPs might still pose a threat, especially as a source of toxic metals and/or molecules.

5.6 Vitamin A status is a potential biomarker for contaminant effects

Jakob Strand (DK) presented a review of interactions between contaminants and vitamin A. Levels of retinoids (i.e. vitamin A and its derivates) has become a widely used biomarker of exposure to environmental pollutants, especially in studies on birds and mammals, but the same biomarker potential seems also to be present for fish (Rolland, 2000; Novak *et al.*, 2008).

Retinoids act as hormone-like signalling molecules since they are involved in essential processes such as morphogenesis, development, reproduction or apoptosis. Retinoids become available for the organisms by diary intake of either plant sources (with carotenoids as precursor for vitamin A) or as retinyl esters (REs) from animal sources. ROH is the main transport form of retinoids and has two major fates inside the organism: (1) esterification and tissue storage, which in fish is mainly liver and kidneys; and (2) oxidative metabolism to retinal (RAL), which is the direct precursor of retinoic acid (RA). RA is the active retinoid that regulates biological processes via binding to RA receptors and triggering the cascade molecular reactions, initiating gene transcription (Alsop *et al.*, 2005; Novak *et al.*, 2008; Clagett-Dame and Knutson, 2011)

Some environmental pollutants that affect embryo development and the immune system have also been shown to interfere with the vitamin status in the organisms. For instance organic pollutants like dioxins, PCBs and PAH have been linked to retinoid depletion caused my increased metabolism and thereby by also mobilisation of liver storages of RE. This suggests that CYP induction by contaminants can be an important mechanism for the interactions with retinoid system, since CYP systems also are involved in the retinoid metabolism by both transforming mobilised ROH and RAL into the receptor active form RA as well as in the conversion of RA to more polar metabolites for regulation of cellular levels of RA by excretion processes.

However, also other potential mechanisms for contaminants are related to effects caused their potential as also acting as ligand on retinoid receptors RXR and RAR, where retinoic acid is the ligand for activating the gene response (Murphy *et al.*, 2007). This can also include cross-talk with the AhR receptor, making the cause-effect relationship even more complex.

Both, long-term low-dose exposure to AhR ligands in nature (Brunström *et al.*, 1991; Doyon *et al.*, 1998; Jenssen, 2003; Murk *et al.*, 1998; Skaare *et al.*, 2001; Spear *et al.*, 1992) as well as short-term exposure to a single compound TCDD in the laboratory animals (Kransler *et al.*, 2007; Nilsson *et al.*, 2000; Schmidt *et al.*, 2003) have been shown to alter retinoid concentrations in liver, blood serum and kidney of different organisms. In Table 1, examples from the literature of retinoid changes induced by exposure to environmental pollutants in different animals are shown.

Table 5.1. Studies showing changes in vitamin A status induced by exposure to DLCs in different organisms, reviewed by Tairova (2014).

Compound	Animal	Effects	References
Birds			
TCDD, dlPCB	Herring gull	Reduced hepatic retinol and retinyl esters	Spear *et al.*, 1992
PCB	Black guillemot	Reduced hepatic retinol and retinyl esters	Kuzyk *et al.*, 2003
Mammals			
PCB	Mink	Reduced hepatic retinol	Brunström *et al.*, 1991
PCB	Otters	Reduced hepatic retinol and retinyl esters	Murk *et al.*, 1998
PCB	Polar bear	Reduced plasma retinol	Skaare *et al.*, 2001
PCB	Grey seal	Reduced plasma retinol	Jenssen, 2003
Organohalogens	Sled dog	Reduced hepatic retinol	Kirkegaard *et al.*, 2010
TCDD	Rat	Reduced hepatic retinol and retinyl esters, increased renal retinol and retinyl esters	Kransler *et al.*, 2007
TCDD	Rat	Reduced hepatic retinol and retinyl esters, increased renal retinol and retinyl esters, increased serum retinoic acid (RA)	Nilsson *et al.*, 2000
TCDD	Rat	Reduced hepatic retinol, retinyl esters and RA metabolites; increased hepatic RA; increased renal RA and serum RA	Schmidt *et al.*, 2003
PCB	Rat	Reduced hepatic retinol and retinyl esters; reduced serum retinol	Morse and Brouwer, 1995
Fish			
TCDD, dlPCB	White sucker	Reduced hepatic retinol, retynil esters and 3,4-dehydroretinyl ester	Spear *et al.*, 1992
PCB	Sturgeon	Reduced hepatic retinol and retinyl esters, dehydroretinol, dehyretinyl esters	Doyon *et al.*, 1998
PHAH, PAH	Flounder	Reduced hepatic and plasma retinol	Besselink *et al.* 1998
EE2	Arctic grayling	Reduced hepatic retynil esters	Palace *et al.* 2001
PAH, Cu	Zebrafish	Reduced whole body retinoid levels	Alsop *et al.*, 2007
PBDE	Zebrafish	Reduced levels of retinyl esters in intestines, Increased deposition of retinal in eggs	Chen *et al.*, 2012

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5.7 Periodic thiamine deficiency in aquatic ecosystems

Lennart Balk (SE) presented ongoing work on thiamine deficiency. Populations of many animal species in the Northern hemisphere are currently declining more rapidly than can be explained by known causes, such as habitat loss, climate change, overfishing, and classical environmental pollutants. In a worldwide perspective, these ongoing population declines and the loss of species have been described as the sixth mass extinction (Barnosky *et al.*, 2011), and naturalists have proposed this development as the most serious environmental threat to life on our planet (Rockström *et al*., 2009). For several species this negative development has been paralleled by an observed altered behaviour, paralysis, and excess mortality, especially during the last two decades.

Recent discoveries have resulted in a scientific hypothesis [Balk *et al.*, 2009], which implies that a wide range of wild animal species, belonging to different animal classes, are suffering from an anthropogenic thiamine (vitamin B1) deficiency, which causes a number of disorders and, as an ultimate consequence, substantial population declines. Affected animal classes include fish (Fitzsimons, 1995, Åkerman and Balk, 1998; Balk *et al.* 2015), birds (Balk *et al*., 2009, Paton *et al*., 1986), reptiles (Ross *et al*., 2000, Sepulveda et. al., 2004), and bivalves (Balk *et al.* 2015). In the aquatic environment, beside several Salmonids, the well-known negative global development (migration, reproduction, survival) of eel (*Anguilla anguilla*, *A. rostrata*, and *A. japonica*) (Dekker 2003) populations gives reason to assume widely, not to say globally, occurring thiamine deficiency (Fitzsimons *et al*., 2013; Balk *et al.* 2015).

In summary, lethal thiamine deficiency has by now been demonstrated in several species from different geographical areas and ecological niches, including terrestrial, freshwater, brackish, and marine environments.

Thiamine is a water-soluble vitamin essential for all vertebrates and invertebrates (Gothilf and Waites 1968). Here, essential means that it must be obtained from the food. Thiamine is present in four major forms in the tissues of the body, either non-phosphorylated or phosphorylated with 1–3 phosphate groups. It is mainly the non-phosphorylated form of thiamine (T) that is absorbed from the intestine. Inside the cells, T is phosphorylated in an enzymatic reaction, in which a kinase adds two phosphate groups so that thiamine diphosphate (TDP) is formed. TDP acts as an essential cofactor for several life-sustaining enzymes in basic metabolism, for example transketolase, pyruvate dehydrogenase, and α-ketoglutarate dehydrogenase. Transketolase (TK) is present in the cytosol and is part of the hexose monophosphate shunt, which produces building blocks for nucleic acid synthesis, reducing equivalents (NADPH), and 4-, 5-, and 7-carbon sugars for further metabolism in glycolysis. Pyruvate dehydrogenase (PDH) is present in the mitochondria and catalyses a reaction, which couples the glycolysis to the citric acid cycle. This reaction prevents the accumulation of lactate, which otherwise will result in acidosis, which in turn results in cell death and tissue necrosis. For example, the neurons in the brain have been considered to be particularly sensitive to thiamine deficiency-induced acidosis. α-Ketoglutarate dehydrogenase (KGDH) is present in the mitochondria and catalyses one of the reactions in the citric acid cycle. In this metabolic step, reducing equivalents for the respiratory chain are produced (NADH), among other things. A fourth thiamine dependent enzyme is branched-chain α-keto acid dehydrogenase (BCKDH). This enzyme is also present in the mitochondria and metabolizes derivatives of the branched amino acids valine, leucine, and isoleucine. The fifth known thiamine dependent enzyme is 2-hydroxyacyl-CoA-lyase, which is present in the peroxisomes and catalyses the α-oxidation of lipids (Sniekers *et al.*, 2006). In the light of these facts, it is easily realized that even minor changes of the thiamine status in animals may cause severe systemic effects and damage. Apart from the direct role of thiamine as a cofactor in thiamine dependent enzymes, it has another important function in the body. A fraction of the TDP is further phosphorylated to thiamine triphosphate (TTP), which is necessary for the functioning of nerves (Nakagawasai, 2005). Two central and direct consequences for individuals affected by thiamine deficiency are impairment of the immune system (Kumar and Axelrod 1978, Pletsity *et al.* 1979, Prasad *et al.* 1980, Pletsityi and Pletsity, 1987; Shoji *et al.* 1994; Shoji *et al.* 1998; Fattal-Valevski *et al.*, 2005) and damage to the blood-brain barrier (Harata and Iwasaki 1995, Calingasan and Gibson, 2000). These effects of thiamine deficiency result in increased susceptibility to pathogens and toxic substances, respectively. We have, in fact, already encountered numerous examples of infections in birds that most probably are secondary to thiamine deficiency (Balk *et al.*, 2009; Balk *et al.*, 2010; Balk *et al.*, 2015), yet mistakenly thought to be primary causes for the mortality. Thiamine deficiency results in increased formation of certain metabolites, raising them to toxic concentrations in the cells, due to perturbed metabolism. Such metabolites include pyruvate, lactate, glutamate, alanine, glyoxal, methyl glyoxal, γ-butyric acid, and phytanic acid.

As a consequence, the clinical and biochemical symptoms of thiamine deficiency are often expected to be more related to the concentration of toxic metabolites and their detoxification, than to the absolute thiamine concentration in the tissues. Finally, new research in mammals indicates that chronic and/or repeated thiamine deficiency is also implicated in the progression and onset of a number of chronic diseases, including diabetes (Karachalias *et al.*, 2005), atherosclerosis, heart disorders (Avena *et al.*, 2000), Alzheimer's disease (Rodriguez-Martin *et al.*, 2001, Gibson and Zhang, 2002), eye disorders (Jacques *et al.*, 2005) and some forms of cancer (Bruce *et al.*, 2003).

Thiamine deficiency as an environmental problem has also been recognized by the International Union for Conservation of Nature and Natural Resources (IUCN), an organization with the purpose to inform the general public, the scientific community, and responsible authorities about current threats against biodiversity (Vié *et al.*, 2009). They regularly publish a "red list" with endangered species, globally and nationally. The Swedish branch of IUCN (ArtDatabanken) has suggested two priority areas where further scientific knowledge is urgently needed (Gärdenfors *et al.* 2010). The first area is the severe mortality among trees that has reduced the populations of various species in Sweden during the last 20–25 years. The second priority area suggested by IUCN is the problem with thiamine deficiency in fish and birds, and possibly also other classes of animals. All thiamine circulating in the ecosystems (terrestrial and aquatic) is produced by the green plants (including algae), and the possibility that this natural process may be at risk makes it crucial to clarify the issue.

Thiamine deficiency has been observed in both primary and secondary consumers from several animal classes, birds (Balk *et al.* 2009), fish (Fitzsimons 1995) and bivalves (Balk *et al.*, 2015). A chemical substance causing thiamine deficiency may exert its effect either by decreasing the uptake of thiamine or by increasing its consumption (metabolic degradation and/or excretion), or both. Thiamine deficiency may also arise from insufficient amounts of thiamine in the food. Since the mid-1990s, when it was first realized that numerous fish species suffered from thiamine deficiency (Fitzsimons 1995, Åkerman and Balk, 1998), several hypotheses for the ultimate cause have been proposed, but so far none of them can alone account for the wide array of population level effects that have been observed in wildlife. The documented and widespread thiamine deficiency has certain characteristics that are very unusual for environmental effects of known toxic chemicals. Its geographical distribution, occurrence in many different types of animals, and temporal variation all indicate that classical environmental pollutants are not the underlying cause. Moreover, in contrast to many such classical pollutants, the causative agent(s) of thiamine deficiency appears not to be accumulated in the food web. Organisms at the top of the food web are not necessarily affected most severely; rather, thiamine deficiency impacts the ecosystem “sideways”, i.e., with approximately the same severity at all trophic levels in the food web. In addition to being more difficult to unravel, such an effect is indicative of an unknown toxic mechanism that differs radically from present dogma in the field. This fact probably explains, to a large extent, why the thiamine deficiency has occurred for a long time without attracting much attention. The observation that thiamine deficient animals are observed also at comparatively remote sites (fjords in Iceland, remote areas in the Baltic Sea, etc.) adds a new perspective to this environmental problem. In fact, it may suggest the involvement of atmospheric distribution, as well as atmospheric conversion, of the causative agent.

Recommendations

The following should be monitored in different species: transketolase (including percentage apoenzyme) and thiamine analysis (T, TMP, TDP), liver brain (ratio).

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6 Review status of publications and consider requirements for new publications (ToR c)

6.1 ICES TIMES

WGBEC reviewed status with ICES TIMES manuscripts and revised deadlines as appropriate. Several new manuscripts were produced in draft for the meeting to review. Status of the manuscripts was deemed satisfactory. The current status of WGBEC TIMES manuscripts is given in table 6.1 below.

Table 6.1 Current status of ICES TIMES publications.

Resolution Ref	Deadline	Description	Comment on status
2012/1/SSGHIE08	30/04/13	The report on the COMET assay for fish and mussels Author/Editor: Tim Bean (UK) and Farida Akcha (France).	Formatting and review required.
2012/1/SSGHIE09	30/04/13	The report on the Condition Index for fish and mussels. Author/Editor: John Thain (UK), Matthew Gubbins (UK), Concepcion Martinez Gomez (ES), and Lennart Balk (SE).	Formatting and text revision required prior to review.
2012/1/SSGHIE10	30/04/13	The report on the Stress On Stress assay for mussels. Author/editor: John Thain (UK) and Concepcion Martinez Gomez (Spain).	Formatting and final edits required.
2012/1/SSGHIE11	31/07/13	The report on the Lysosomal Membrane Stability in the Blue Mussel Author/editor: Concepcion Martinez Gomez (Spain), John Bignell (UK) and David Lowe (UK).	Reviewed version submitted to ICES.
2009/1/SCICOM08	30/11/12	The method for determining 'Reproductive Success in Eelpout' Author/Editor: Jakob Strand (Denmark) Reviewer: WGBEC/SSGHIE	Withdrawn - new request when ready.
2006/1/MHC06	08/10/12	The Protocol for Extraction Methods for Bioassays Author/Editor: Hans Klamer (NL), Steve Brooks (NO) and John Thain (UK) Reviewer: Chair of SSGHIE	Manuscript sent to ICES.
2006/1/MHC07	08/10/12	The protocol for conducting EROD determinations in flatfish Author/Editor: Compiled by M. Gubbins, WGBEC members Reviewer: Chair of SSGHIE	For publication as a minor revision to TIMES 23.
2007/1/MHC02		Blue Mussel Histopathology Author/Editor: John Bignell, Steve Feist, Dave Lowe and MirenCajaraville	Withdrawn -new request when ready.

Requirements for new TIMES manuscripts were discussed. The possibility of a method manuscript for litter monitoring was raised. This will be revisited after EU protocols have been defined.

6.2 Other publications

Ketil Hylland (NO) presented the ICON project publication plan as a series of peer review papers in a special edition of Marine Environmental Research. Authors have been contacted to confirm authorship, deadlines and length of publications. Final submission will be summer 2015. An overview of contributions can be found at Annex 5.

Action: Chairs to monitor progress of manuscripts to comply with deadlines.

7 Report progress on AQC activities for biological effect methods including harmonisation activities initiated from WGBEC and within OSPAR, HELCOM and MEDPOL maritime areas (ToR d)

Any biological effect method to be used for national or international monitoring programmes should be subject to appropriate internal and external AQC, particularly as this is a requirement for submitting data to the ICES database. It is likely that the role of AQC will take on an even greater importance with the use of biological effect methods for monitoring GES (Descriptor 8) in the EU MSFD.

At its meeting in 2012, WGBEC discussed AQC activities for biological effect methods and agreed to initiate a low cost programme for EROD and PAH bile metabolites, organised under BEQUALM. Cefas UK, collected samples of fish liver and bile from wild caught fish and distributed these to interested laboratories, 11 in total from Norway, Denmark, France, Germany, Spain, The Netherlands and the UK. Data from this exercise were made available to WGBEC during the meeting and discussed.

WGBEC are not aware of any current AQC activities underway in the HELCOM maritime area.

Action: Chairs to follow-up on the performed intercalibration with a view of repeating it, including AChE and vitellogenin in addition to EROD and PAH metabolites.

8 Respond to requests for advice from the ICES Data Centre (ToR e)

It was noted that reporting format issues had precluded WGBEC member institutions from submitting biological effects data to the ICES database. In addition, only specific institutions are allowed to submit data. This situation needs to be improved to ensure that biological effects data will be available through the ICES database for OSPAR assessments.

Recommendation: Appropriate formats should be made available for submission of biological effects data.

9 Development and harmonisation of methodologies for marine monitoring (ToR f)

9.1 Integrated assessments

With the interim adoption of the SGIMC approach by OSPAR on a 3-year trial basis there is a request to Contracting Parties to provide evidence of application and assessment of the value of the new approach (cf OSPAR MIME 2012). WGBEC therefore collated examples of national and international case studies either completed, planned or in progress across the ICES/OSPAR regions to keep track of progress with case studies.

The use of an integrated approach was reported by Ginevra Moltedo (IT) for the environmental monitoring carried out by ISPRA for offshore industrial facilities such as oil and gas activities or a Terminal LNG and for a shipwreck (Costa Concordia). Besides chemical analysis of water and sediment samples, biological effects of contaminants were evaluated through biological assays and biomarkers analysis. In particular, biomarker analyses were performed on mussels, clams, fishes and polychaetes. According to the species analysed specific set of biomarkers were chosen including responses at the whole organisms level (mortality, Condition index and stress on stress) and those at subcellular level such as biomarker of exposure (MT, AOX, AChE, EROD, VTG, bile metabolites), of genotoxicity (micronuclei, comet assay), of oxidative stress (CAT, GR, GSTs, GPx, totGSH level, TOSC-Assay, MDL levels) and lysosomal stability (NRRT, cytochemical). A special emphasis was given to the need of identify bioindicator species for evaluation of sediment contamination. To that respect ISPRA highlighted the use of a benthic invertebrate species, *Hediste diversicolor*. After testing the sensibility of the species to B[a]P and to Hg in dedicated experiments, ragworms were tested with sediment samples; biological effects were evaluated after *ex situ* sediment exposure. Comparisons were performed between biological responses after exposure to sediment samples collected in the proximity of source of potential contamination and after exposure to sediment samples collected in a reference site.

Brita Sundelin (SE) presented integrated assessment in the CORESET project. The HELCOM CORESET project started in 2010 and had the objective of developing and delivering a set of preliminary core indicators and targets to be forwarded to the national decision making processes by the end of September 2011. This HELCOM project was replaced by Coreset II project that will run from autumn 2013 to summer 2015. The core indicators will ultimately be placed on the HELCOM web page and kept annually/bi-annually updated with new data. In order for the core indicators to be clear, concise and informative, the final core indicators and their information sheets need a uniform format. It is of utmost importance that the HELCOM core indicators covering eutrophication, hazardous substances as well as biodiversity aspects of the ecosystem comprise a uniform set of indicators with identical layout and headings.

Two assessment criteria are used to assess biological effects: Background Assessment Criteria (BAC) and Environmental Assessment Criteria (EAC).

The assessment criteria were developed within the Oslo and Paris Commission (OSPAR) framework with scientific advice from the International Council for the Exploration of the Sea. Mean values below the BAC are said to be near background. Values below the EAC indicate no chronic effects on the organisms concerned. Full details can be found in Davies & Vethaak (2012), OSPAR (2013) or Vethaak *et al.* (in press).

It has been important to develop and apply tools for a science-based assessment and management with regard to the impact of anthropogenic contaminants on the Ecosystem Health of the Baltic Sea to further develop science-based guidelines, assessment and management of the impact of anthropogenic contaminants on the Ecosystem Health of the Baltic Sea. Presently, different methodologies are available or under development to monitor and assess pollution effects and ecosystem health in marine and coastal waters. A number of integrated indices and similar approaches based on the measurement of a set of biomarkers have recently been developed and tested in the field in the North Sea/Atlantic or the Mediterranean. So far their application for the specific conditions in the Baltic Sea is still missing. The ideas and concepts were taken into consideration into the further development and recommendations of a set of bioeffect indicators for the future HELCOM MONAS programme. The HELCOM core indicators should primarily be used to assess the effectiveness of the implementation of the Baltic Sea Action Plan (BSAP), but also as tool for implementation of the Marine Strategy Framework Directive (MSFD). In this role, the core indicators needed to be aligned with the EU MSFD descriptors and criteria and methodological standards of good ecological status (GES). In accordance with the HELCOM Monitoring and Assessment strategy the core indicators will in the future be used to update the Thematic Assessments and the Holistic Assessment. Core indicators are being developed for eutrophication in the EUTRO-OPER project and for hazardous substances and biodiversity in CORESET II.

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9.2 Environmental risk assessment

B. E. Grøsvik (NO) presented two papers on environmental risk assessment, which were also relevant to agenda point 5.1- Oil toxicity to early stages of fish. The papers are listed under 5.1 (Hauge *et al.*, 2014 and Blanchard *et al.*, 2014.).

9.3 Assessment criteria

Addition of BAC-value for EROD activity in microsomal fraction for eelpout

Lars Förlin (SE) and Jakob Strand (DK) suggested criteria for EROD in eelpout liver. Swedish monitoring data for EROD activity in microsomal fraction for eelpout has been used for deriving Background Assessment Criteria at 20 pmol min^{-1} mg protein^{-1} based on long time trend series using sample sizes at 15 – 25 individuals per year from the reference stations Kvädofjärden in the Baltic Sea and Fjällbacka in the Skagerrak (the North Sea); (Hansson *et al.* 2014).

Based on this work, a BAC-value for EROD activity in microsomal fraction at 20 pmol min^{-1} mg $protein^{-1}$ for eelpout has been added to the table for assessment criteria (Table 9.1).

Comparative studies on BEQUALM and other relevant samples on EROD activity in both microsomal fraction and S9-fraction of fish liver, shows that EROD activity in the microsomal fraction can be determined to be around factor 3.5 higher in average after adjustment to protein content (Förlin, pers. comm.). This factor has thereby potential for being used as conversion factor comparing EROD data for microsomal fractions and S9 fractions. Using this conversion factor will correspond to a BAC-value for S9 fraction at 5.7 pmol min^{-1} mg $protein^{-1}$, which are in the same range as the 10 pmol min^{-1} mg $protein^{-1}$, which previously has been proposed for EROD activity in S9-fraction for eelpout (WGBEC report 2012), and thereby indicate the consistency for comparison of the two BAC-values for based on microsomal fraction and S9 fractions for EROD activity in eelpout liver.

Table 9.1. New criteria for EROD (perch and eelpout) and vitellogenin for perch.

Method	Species	BAC
VTG in plasma; ng/ml	Perch (M)	115
EROD; pmol/min/mg protein	Eelpout (F) Perch (F)	20 50

The criteria for malformed embryos of amphipods are based on monitoring data during 20 years in the Bothnian Sea and Baltic proper: 199–2011. Fourteen stations were included in the analysis. Data are based on 8600 gravid females and 230 000 embryos. A sample size of 50 gravid females (about 1500 embryos) was used. This is the recommended sample size within the National Monitoring program. The background value was 3.8 % malformed embryos, resulting in a BAC of 3.8. Another way to assess the effect is to analyse the proportion of females carrying malformed embryos. We used data from contaminated areas (outside industries) and monitoring data and comparison of proportion of females with more than 1 malformed embryo facilitates detection of statistically differences between pristine and contaminated areas. The EAC for females with more than one malformed embryos was 15.4 % and the 90e percentile 22%, resulting in an EAC of 22.

An assessment tool for monitoring liver cancer in marine environment– a preliminary report

John Bignell and Allan Reese (UK) submitted a proposal for a new assessment tool for monitoring liver cancer in marine fish to the 2014 WGBEC meeting. The work has primarily arisen from previous concerns raised that liver cancer might simply be a surrogate for age and that contaminants may not be the sole cause. This is particular concerning when the age distribution of one of the OSPAR region's primary biomarker species, the common dab (*Limanda limanda*), is considered. Analysis of data from dab of the size range 20–24 cm, as recommended by ICES in biological effects monitoring programmes, revealed that age can vary considerably.

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Recommendation: ICES Secretariat to inform OSPAR of revised criteria.

9.4 National monitoring programmes

Denmark

Jakob Strand (DK) presented status on the biological effects monitoring in Denmark. The biological effects monitoring in Denmark has since 1998 been performed yearly within the frame of the contaminant monitoring in the Nationwide Monitoring and Assessment Programme for the Aquatic and Terrestrial Environment (NOVANA). The biological effects monitoring has since the beginning in 1998 included imposex and intersex in 5 different marine snail species (*Hinia, Buccinum, Neptunea, Nucella and Littorina*) and in addition since 2004 included lysosomal membrane stability (LMS) in blue mussels (*Mytilus edulis*), and PAH-metabolites, EROD and reproductive success in eelpout (*Zoarces viviparus*). Fully coordinated sampling between the contaminants and the effects monitoring have been performed on blue mussels since 2004, where mussel sampled at the same time and place also have been analysed for metals, organotins, PAHs, dioxins and PCBs. For eelpout, coordinated sampling between the contaminants and the effects monitoring have been performed since 2011, where fish sampled at the same time and place also have been analysed for metals, organotins, BDEs, PFCs, dioxins and PCBs.

Due to prioritisations within the monitoring programme, however, biological effects monitoring has from 2014 experienced a significant reduction on the programme going yearly from 12 to two for both the mussel and eelpout stations. In the imposex programme, all the coastal stations has been removed, and only open water monitoring stations along shipping lanes are now prioritised, at least until 2015, where after a new revision of the programme will take place.

France

The CEMP monitoring strategy in France was presented by Thierry Burgeot (FR). The monitoring activity is conducted under the national network (Rocch: monitoring network of chemical contamination in French) since 1974 and allows to assure the continuity of an expertise and the collection of data in long term. The list of the chemical contaminants analysed under the national network is the list recommended by OSPAR and for some of them by the Barcelona convention. The chemical contaminants are analysed annually in oysters and mussels. Contaminants are also analysed in the sediment every five years. The imposex is determined every year as a mandatory biomarker since 2003.

The integrated approach of chemistry and biology was adopted in the CEMP monitoring and was used on a research basis in the pilot site (Seine estuary) in the channel between 2008 and 2012. The dataset was sent in 2015 to the ICES database. The French ministry of ecology and energy and sustainable development published an order for the descriptor 8 of the MSFD (17 December 2012) with a list of chemical contaminants and biological effects : Lysosomal stability, genotoxicity (Micronuclei, comet assay) in fish and mussels, reprotoxicity (gonads histology in fish and oyster embryotoxicity), fish pathologies and supporting parameters (size, weight, LSI, GSI). From, 2015, these parameters will be analysed in the French MSFD workplan for monitoring in three pilot sites (Seine estuary, Loire estuary, Marseille and Rhone delta) and will be twice analysed each during a first cycle of six years.

Germany

The German national monitoring programme of biological effects was presented by Ulrike Kammann (GE). She gave an overview on the methods used by the German Thünen Institute to fulfil the requirements of MSFD D8 which comprise external visible fish diseases, liver histopathology and PAH metabolites in fish bile. Other methods like micronucleus test and lysosomal stability were also used in the lab but are not yet part of the monitoring programme. PAH metabolites and fish diseases data have been submitted to the ICES database for years and are ready to be included in international evaluations.

In addition to this in Germany data of imposex were generated by an institution of Lower Saxony (NLWKN

http://www.nlwkn.niedersachsen.de/portal/search.php?_psmand=26&q=imposex&searchMode=&searchType=&searchInst=www.nlwkn.niedersachsen.de).

The monitoring programme of the Thünen Institute will be keep on going for the next years. At the moment there are no plans to alter or stop biological effects monitoring in Germany. MSFD-D8 requirements have a high priority for the Thünen Institute especially for those parameters that are mandatory. PAH metabolites are part of the HELCOM CORESET and are therefore recommended parameters for fish in the Baltic Sea. The German monitoring positions are located in the North Sea and in the southern Baltic Sea region and are not restricted to the national waters. Most fish are sampled off shore or in the German Bight. During the last years fish diseases in the Baltic Sea were assessed in international research projects on possible adverse effects of dumped munitions (CHEMSEA, MODUM). However, also data from other regions have been generated: Fish from low polluted, distant areas (e.g. Iceland, Barents Sea) have been used to calculate background assessment criteria for PAH metabolites.

Thünen Institute investigates around 200 fish bile samples each year by high performance liquid chromatography with fluorescence detection. Ulrike Kammann stressed that continuous quality assurance is important for all methods applied in monitoring, including PAH metabolites. The last intercalibration has been published five years ago (Kammann *et al.*, 2010) and should be repeated. Of the different methods for PAH metabolite quantification three methods: HPLC-F, GC-MS and SFS (with conversion factor) have been shown before to produce comparable results (Kammann *et al.*, 2010).

As an example data from the routine monitoring cruise in summer 2013 were presented. Newer data will be available soon. This data set cover PAH metabolite concentration in different fish species: dab, flounder, cod, mackerel, gurnard and horse mackerel in 188 samples from North Sea and the southern Baltic Sea (Table 1). Concentrations of the main metabolite 1-hydroxypyrene varied from 8.8 to 818.8 ng/ml showing highest results in cod from the Baltic Sea and in one single dab from the German Bight. These fish exceed the EAC (environment assessment criteria) level for cod of 483 ng/ml. Low concentrations, even below BAC (background assessment criteria) level for cod of 21 ng/ml for cod were found in different fish species. The distribution of the results suggest that large-scale comparisons of PAH metabolite concentration can be based on different fish species.

Table 9.1. Concentrations of PAH metabolite 1-hydroxypyrene in bile of fish caught in the North Sea and in the Baltic Sea in summer 2013.

FISH	1-OHPYR mean [ng/ml]	N	Min	Max
dab	94.66	60	11.5	818.8
cod	215.22	61	13.6	697.9
mackerel	74.88	30	8.8	273.6
flounder	95.33	29	26.8	461.1
horse mackerel	232.41	4	71.5	286.0
gurnard	92.96	4	63.5	119.2
All	**133.62**	**188**	**8.8**	**818.8**

Reference

Kammann U, Askem C, Dabrowska H, Grung M, Kirby MF, Koivisto P, Lucas C, McKenzie M, Meier S, Robinson C, Tairova ZM, Tuvikene A, Vuorinen PJ, Strand J (2013) Interlaboratory proficiency testing for measurement of the PAH metabolite 1-hydroxypyrene in fish bile for marine environmental monitoring, Journal of AOAC International. 96(3) 635-641.

Norway

Offshore Monitoring

An overview of on-going, governmental-initiated environmental monitoring programs in Norway was presented by Steven Brooks (NO). The Norwegian offshore monitoring programme, which is funded by Norwegian Oil and Gas has been performed over the last 20 years and is divided into three parts: 1) water column monitoring (WCM); 2) condition monitoring; and 3) sediment monitoring. A suite of biological effects methods in fish are employed in both the WCM and the condition monitoring programmes, of which the WCM was presented. The current WCM deploys an integrative approach of chemical and biological effects methods in wild fish populations that reside around offshore oil and gas installations. The biomarkers used include PAH metabolites, DNA adducts, COMET, EROD, LMS, VTG, AChE, liver and gill histology. Until 2014, the WCM was performed annually. Future WCM activities will include a large-scale biomonitoring programme using integrative biological effects methods in caged mussels and wild fish populations every third year.

Spain

This update was presented by Juan Bellas (ES). The Spanish Oceanographic Institute (*Instituto Español de Oceanografía*, IEO) is the scientific institution in charge of carrying out the Spanish Marine Pollution Monitoring Program. The IEO is also designated by the Spanish Government, through the Law 41/2010 for the Protection of the Marine Environment (which is the transposition of the Marine Strategy Framework Directive into the national legislation), as the institution in charge of the scientific implementation of the MSFD. Following the recommendations of the OSPAR and Barcelona Conventions, sampling of wild mussels (*Mytilus galloprovincialis*) for the study temporal trends of pollutants and biological responses is being held on an annual basis in Atlantic and Mediterranean areas of special relevance and in reference areas. According to the OSPAR-JAMP guidelines, the selected season for sampling mussels in the Atlantic coast corresponds to the period in which these organisms are found in a more stable physiological state (OSPAR, 1999), the pre-spawning season, which in this coast corresponds to the period October-December. The recommended period for sampling in the MEDPOL program corresponds to the pre-spawning period of May-August. Regarding fish species, the red mullet (*Mullus barbatus*) is sampled in the Mediterranean region, whereas the studied fish species in the Atlantic coast are the hake (*Merluccius merluccius*) and the spotted dogfish (*Scyliorhinus canicula*). Fish sampling is conducted with research trawling vessels, and the campaigns are conducted in October-November, both in the Atlantic and Mediterranean coasts. Gastropods (*Nassarius reticulatus* and *Nucella lapillus*) are sampled in the Atlantic coast for Imposex measurements. CEMP components are measured in mussels, fish and gastropods from the Spanish Atlantic coast. Those components include: metals, PAHs, PCBs, organochlorine pesticides and BDEs in sediments, mussels and fish, and Imposex in gastropods. Also Scope for Growth is measured in mussels, and sediments are used to conduct sea-urchin embryo-larval bioassays (Tables 1 and 2). Currently, the technique for PAHs metabolites in fish bile is being optimized and the frequency of micronuclei is being measured in mussel hemolymph on a trial basis. In the Mediterranean coast, metals, PAHs, PCBs, organochlorine pesticides are measured in mussels, red mullet and sediments. The biological responses to be measured are: Sress on Stress, lysosomal membrane stability, frequency of micronuclei, acetylcholinesterase activity, metallothioneins and antioxidant enzymes, in mussels, and frequency of micronuclei, acetylcholinesterase activity, EROD activity, ALA-D activity, plasma vitellogenin and PAHs metabolites in bile, in red mullets (Tables 9.2 and 9.3).

Table 9.2. Biomarkers and physiological parameters to be determined in sediments, mussels and fish.

			Matrix	Level	Organism
ATLANTIC COAST	Biological responses	Scope for Growth	Whole organism	Individual	Mussels
		Frequency of micronuclei (pilot study)*	Hemolymph	Cellular	Mussels
		Antioxidant enzymes*	Digestive gland	Molecular	Mussels
		PAHs metabolites in bile (pilot study)	Bile	Molecular	Fish
		Imposex	Whole organism	Individual	Gastropods
		Larval growth	Sediment	Individual	Sea-Urchins
	Physiological parameters	Condition index Gonadal Index* Hepatosomatic Index* Branchial Index*		Individual	Mussels
MEDITERRANEAN COAST	Biological responses	Stress on Stress	Whole organism	Individual	Mussels
		Lysosome membrane stability	Hemolymph	Cellular	Mussels
		Frequency of micronuclei	Hemolymph	Cellular	Mussels and fish
		Acetylcholinesterase	Gills/Brain	Molecular	Mussels and fish
		Antioxidant enzymes	Digestive gland	Molecular	Mussels and fish
		EROD activity	Liver	Molecular	Fish
		ALA-D activity (pilot study)	Blood	Molecular	Fish
		Intersex (pilot study)			
		PAHs metabolites in bile (pilot study)	Bile	Molecular	Fish
	Physiological parameters	Condition index Gonadal Index Hepatosomatic Index Branchial Index		Individual	Mussels
		Condition Factor Lipid content of muscle tissue Hepatosomatic Index Gonadosomatic Index		Individual	Fish

***in coordination with BIOCOM research project.**

Table 9.3. Pollutants to be determined in sediments, mussels and fish.

Metals	Mercury, Lead, Cadmium, Copper, Zinc, Arsenic, Nickel Lithium, Aluminum	Mussels, fish and sediments
Polychlorinated biphenyls	CB28,CB52, CB101, CB105, CB118, CB138, CB153, CB156, CB180	Mussels, fish and sediments
Organochlorinated pesticides	γ-Hexachlorocyclohexane, α-Hexachlorocyclohexane Hexachlorobenzene, p,p´DDE; p,p´DDT; p,p´DDD; o,p DDT DieldrIn, AldrIn	Mussels, fish and sediments
Polybrominated biphenyl ethers (only in the Atlantic coast)	BDE28, BDE47, BDE66, BDE85, BDE99, BDE100, BDE153, BDE154, BDE183 y BDE 209	Mussels, fish and sediments
Polycyclic Aromatic Hydrocarbons	Fluorene, Anthracene, Phenanthrene, Fluoranthene, Pyrene, Benzo [a] anthracene, chrysene Benzo [b] fluoranthene, benzo [k] fluoranthene, Benzo [a] pyrene, benzo [e] pyrene, Benzo [g, h, i] perylene Dibenzo (ah) anthracene, Indene [1-2-3-cd] pyrene	Mussels and sediments

Compliance with MSFD descriptor 8

This monitoring program (CONT) aims to evaluate spatial and temporal trends of pollution, to investigate new problems that may arise in this field, to control pollution incidents, and to identify inputs and sources of pollutants. It is subdivided and structured through 5 subprograms: Pollution in coastal waters (CONT1) and offshore (CONT2); radionuclides pollution (CONT3), episodes of accidental pollution (CONT4) and microbial pathogens in water (CONT5).

Complementary to those subprograms, CONT monitoring program also includes 3 subprograms to quantify the contributions of pollutants from various anthropogenic sources (PRES1: riverine inputs, PRES2: atmospheric deposition, PRES3: point sources of pollution), and 4 subprograms to characterize activities that can potentially cause pollution (ACT3: aquaculture, ACT4: port activities, ACT5: Navigation and ACT8: exploration and exploitation of hydrocarbons).

Monitoring marine pollution in coastal waters

The subprogram CONT1 will focus on monitoring pollution and its possible biological effects in the coastal area (up to one nautical mile offshore from the straight baseline). This area is the most exposed to direct or diffuse pollution from land area, and therefore also the most vulnerable. CONT1 will cover the main 'hot spots' of each marine demarcation, according to the risks at the local level on the basis of the known pressures and impacts, and areas far from pollution sources. This subprogram intends to integrate and harmonize data generated by the WFD and data sent to regional seas conventions. Spatial and temporal monitoring of the concentrations of pollutants in water, sediment and biota, as well as their effects on organisms will be carried out, integrating information of both pollution levels and the impact on the environment. The intensity and sampling effort will be greater in those areas most likely to be affected by chemical pollution. Annual or biennial sampling will be carried out for mussels and fish, whereas in the case of sediments sampling will be done every 2–5 years depending on the rate of sedimentation and pollution in the area.

Indicators covered within the subprogram:

Chemistry

- metals, PCBs, PAHs, PBDE, organochlorine pesticides (biota, sediment);
- organotins (sediment);
- HCBD (biota);
- concentration of priority substances and other pollutants in coastal waters (WFD).

Biological responses

- AChE, imposex, Intersex, Scope for Growth, Stress on Stress, Sea-urchin larval growth, Lysosome membrane stability, Micronuclei, PAHs metabolites in fish bile, EROD, Metallothioneins.

Monitoring marine pollution offshore

The subprogram CONT2 will focus on monitoring pollution and its possible biological effects in open waters (from one nautical mile offshore from the straight baseline to the outer limit of the Exclusive Economic Zone), which are less exposed to direct land-based pollution. Sampling areas will include reference areas, areas influenced by pollution sources and/or polluted areas (according to previous studies). Spatial and temporal monitoring of the concentrations of pollutants in sediment and biota, as well as their effects on organisms will be carried out, integrating information of both pollution levels and the impact on the environment. The intensity and sampling effort will be greater in those areas most likely to be affected by chemical pollution. Annual or biennial sampling will be carried out for mussels and fish, whereas in the case of sediments sampling will be done every 2-5 years depending on the rate of sedimentation and pollution in the area.

Indicators covered within the subprogram:

Chemistry

- metals, PCBs, PBDE, organochlorine pesticides (biota, sediment);
- PAHs, organotins (sediment).

Biological responses

- Sea-urchin larval growth, Micronuclei, PAHs metabolites in fish bile, AChE, EROD, Intersex.

Reference

Bellas J, Albentosa M, Vidal-Liñán L, VBesada V, M. Franco MA, Fumega J, González-Quijano A, Viñas L, Beiras R. 2014. Combined use of chemical, biochemical and physiological variables in mussels for the assessment of marine pollution along the N-NW Spanish coast. *Marine Environmental Research*, 96: 105-117.

Sweden

An overview of fish monitoring work in Sweden was given by Lars Förlin (SE). It was reported that the health status of two sentinel fish species perch (*Perca fluviatilis*) and the viviparous eelpout (*Zoarces viviparus*) have been regularly studied in four national reference Swedish coastal sites, one located in the Bay of Bothnia (Holmön), two in Baltic Proper (Kvädöfjärden and Torhamn) and one in Skagerrak (the North Sea); (Fjällbacka). In these coastal reference sites with no or small local point sources of contaminants, perch and eelpout health studies together with analytical chemistry work to measure anthropogenic chemicals and fish ecology studies, form an integrated fish monitoring program supported by the Swedish EPA. The fish health work has been run yearly for more than 25 years, and the integrated work for 15 years. Generally the fish health studies seem to indicate good status in the reference sites but an increasing number of the fish health parameters (i.e. biomarkers) clearly show significant time trends, which suggest changes of concern. For example in female perch from Baltic Sea coastal sites has been observed 20–30% reduction of gonad size, and a more than five times increase of the activity of the detoxification enzyme EROD. Other significant time trends include e.g. changes in blood plasma ions i.e. chlorine and calcium contents, increase lymphocyte number, and indications of oxidative stress.

It was also presented results from a recent project focused on the reference area Kvädöfjärden. The purpose of this project was to find possible explanation for the observed time trends. It was for example reported some details about co-variation over time for different biomarkers and some pollutants as well as that different biomarkers co-varied with different environmental factors e.g. temperature, precipitation, salinity as well as benthic fauna composition. It was concluded, based on current knowledge that it is not possible to find any simple explanation/causation for the observed changes of fish health in coastal fish from Kvädöfjärden. Instead the causes the changes seen in coastal fish health is to be found in combined impact of continuous and varied exposure to mixtures of chemicals and changes over time in different environmental factors such as temperature, salinity and food availability. The outcome of the project thus showed that there still are a number of questions that need to be clarified to elucidate causality. These questions are related for example to route of transport and exposure of pollutants including bioturbation in sediment, food preference and availability, land-sea gradient of pollutants and biomarker patterns. It was also stressed that more data is needed about degradation product of pollutant, and especially OH-PBDEs were mentioned, and that knowledge gaps with respect to ecotoxicity and time trends in biota must be filled for certain compounds groups such as organophosphate esters, adipates and siloxanes.

In addition, it was also presented that the Swedish monitoring biomarker data obtained in fish from the four national coastal areas has been used to set background or reference values. All the values were based on the variation in the average values each year, and set to represent 95% of all values from the reference sites. The established interval will then represent the normal variation from a reference area. It was finally shown how these reference values can be an aid in assessing the fish health impact in monitoring for example downstream point sources (e.g. in receiving areas of industry effluents).

Action: To prepare an overview paper for the 2016 meeting.

10 Address issues in relation to novel and emerging contaminants (e.g. pharmaceuticals, nanoparticles, toxicity of mixtures, discharges from mining activities, etc.)

- Pharmaceuticals and recreational drugs in the marine environment
- Nanoparticles in the marine environment
- Discharges from mining activities

Joachim Sturve (SE) showed results with monitoring and effects studies of pharmaceuticals and nanoparticles in the marine environment (Ref Chapter 5). Ketil Hylland (NO) introduced to discussions on pharmaceuticals and discharges of mining. Jacob Strand (DK), presented results from monitoring discharges of mining activities in Greenland.

Following discussion it was decided to follow the subsequent strategy for delivery of this ToR:

1) Continue to receive updates on inputs, concentrations and effects of emerging contaminants including: biocides, pharmaceuticals, nanoparticles and recreational drugs and in-combination effects.
2) Consider the above in the context of environmental risk assessment.
3) Produce a review document for each of these issues by 2015 highlighting advances made, continued knowledge gaps and recommendations for environmental monitoring.

Articles on emerging contaminants in marine ecosystems include nanoparticles (Munari *et al.*, 2014), mining flame retardants (Hutchinson *et al.*, 2013), Pharmaceuticals (Lillicrap *et al.*, 2015) and mining (Brooks *et al.*, 2015).

References

Brooks S, Harman, C, Hultman MT, Berge JA. 2015. Integrated biomarker assessment of the effects of tailing discharges from an iron ore mine using blue mussels (Mytilus spp.). Science of the Total Environment, 524: 104-114.

Hutchinson TH, Lyons BP, Thain JE, Law RJ. 2013. Evaluating legacy contaminants and emerging chemicals in marine environments using adverse outcome pathways and biological effects-directed analysis. Marine Pollution Bulletin, 74:517-525.

Lillicrap A, Macken A, Thomas KV. 2015. Recommendations for the inclusion of targeted testing to improve the regulatory environmental risk assessment of veterinary medicines used in aquaculture. Environment International, 85: 1-4.

Munari M, Sturve J, Frenzilli G, Sanders MB, Christian P, Nigro M, Lyons BP. 2014. Genotoxic effects of Ag2S and CdS nanoparticles in blue mussel (Mytilus edulis) haemocytes. Chemistry and Ecology, 30: 719-725.

11 Update on plastics (ToR h)

This agenda point was addressed in a joint session with MCWG and WGMS in 2014 (see also http://www.ices.dk/news-and-events/news-archive/news/Pages/43-scientists,-three-expert-groups,-one-overriding-theme.aspx).

The joint session was chaired by WGBEC who also provided the majority of presentations on marine litter and microplastics, see section 5.4.1. Furthermore, Marilynn Sørensen of the ICES Data Centre presented the Data Centre's work on a draft format for litter reporting, as described above.

Thomas Maes (UK) presented a comprehensive review of several aspects of the marine litter issue. The term "Marine Litter" has been introduced to describe "any persistent, manufactured or processed solid material discarded, disposed of, or abandoned in the marine and coastal environment". It consists of articles that have been made or used by people and, subsequently, deliberately discarded or accidentally lost. They originate from ocean-based or land-based sources and can be found in marine environments around the globe. Most sources of marine pollution are land based. Marine litter, mainly plastic, poses a serious environmental threat to marine organisms, as well as a series of economic and social problems. The majority of marine debris is comprised of plastic materials (60-80% overall and 90% of floating debris).

All marine litter particles smaller than 5 mm are considered microparticles. Most microparticles consist of microplastics, although other types exist. The abundance and global distribution of microplastics in the oceans has steadily increased over the decades to around the year 2000 following the rising plastic consumption worldwide since the 1940s. However, there has been a decrease in the average size of plastic litter over this time.

Primary microplastics are produced either for direct use, such as for exfoliants, cosmetics, industrial abrasives or for indirect use as precursors (nurdles or virgin resin pellets) for the production of multiple plastic consumer products. Secondary microplastics formed in the environment as a consequence of the breakdown of larger plastic material, especially marine debris, into smaller and smaller fragments (so called "secondary microplastics"). The breakdown is caused by mechanical forces (e.g. waves) and/or photochemical processes triggered by sunlight (especially UVB). Other types of microparticles are synthetic fibres shedding of textiles by domestic clothes washing, rubber fragments from tires rubbing tarmac, fly ash fine particles that rise with the flue gases after combustion.

The potential impacts of litter span both economic and ecological dimensions. The following section highlights the different aspects that are considered relevant.

Economic: losses to fishing and shipping industry; clean up costs on beaches; loss of tourist revenues; aesthetic disturbance.

Ecological: ingestion; entanglement; introduction of invasive species; bioavailability and transfer due to sorbing/leaching; smothering; disturbance.

Marine litter comes from a variety of land-based and sea-based sources and is essentially a consequence of poor waste management. However, the main sources can be grouped as follows:

The main land-based sources of marine litter are:

- Discharge of untreated municipal sewage, including storm water discharges and overflows
- Tourism (recreational visitors to the coast; beach-goers)
- Riverine transport of waste from landfills or other sources along rivers and other inland waterways and canals

- Industrial facilities: Solid waste from landfills, and untreated waste water
- Municipal landfills (waste dumps) located on the coast or inland
- Direct littering

The main sea/ocean-based sources of marine litter are:

- Fishing vessels
- Merchant shipping, ferries and cruise liners
- Military fleets and research vessels
- Pleasure craft
- Offshore oil and gas platforms
- Fish farming installations

The MSFD requires member states to manage their seas to achieve Good Environmental Status (GES) by 2020. MSFD Descriptor 10 requires litter to be at levels where the '*properties and quantities of marine litter do not cause harm to the coastal and marine environments*'.

MSFD criteria and indicators require understanding and monitoring of (JRC, 2013):

The characteristics of litter in the marine and coastal environment – including:

- Trends in the amount of litter washed ashore and/or deposited on coastlines, including analysis of its composition, spatial distribution and, where possible.
- Trends in the amount of litter in the water column (including floating at the surface) and deposited on the sea- floor, including analysis of its composition, spatial distribution and, where possible.
- Trends in the amount, distribution and, where possible, composition of micro-particles (in particular micro- plastics).

The MICRO EU Interreg project is monitoring microplastics (MP) within the 2 Seas Region and will provide a risk assessment based on field observations, lab experiments and mathematical models. MICRO is a cross border cooperation to prevent environmental, technological and human risks attributed to MP. Furthermore the project will contribute to establish common strategies for environmental risk assessment by modelling the potential impacts on the environment, and by proposing follow-up tools and mitigation measures. The three main pillars of the project are:

Scientific: a risk assessment of the current situation by combining distribution data, modelling and biological effect measurements with socio-economic endpoints.

Educational/knowledge exchange: establishing good practices for adequate monitoring or impact determination across Europe.

Public/scientific awareness: increase awareness of human behaviour in relation to waste production and management by creating co-responsibility among the different actors.

The EU FP7 MARLISCO project activities take place in the four European Regional Seas: North-East Atlantic, Baltic, Mediterranean and Black Sea, by a consortium with members located in 15 coastal countries. MARLISCO' s overarching goal is to raise public awareness, facilitate dialogue and promote co-responsibility among the different actors towards a joint vision for the sustainable management of marine litter across all European seas. It will do this by developing innovative mechanisms and tools. MARLISCO aims to effectively engage, inform and empower society, reaching the widest possible audience.

Bavo de Witte and Lisa Devries (BE) presented recent results of their research framed by the European projects MICRO and CleanSea into the associations between litter and contaminants. A quantitative GC-MS screening was performed on marine litter, present within benthos beam trawl nets during fishing activities. No clear indication of chemical contamination was found on blue synthetic rope. None of the OSPAR-7 indicator PCBs were found at concentrations > 0.1 ng/g. The origin of determined PAHs, alkylated PAHs, alkanes, alkenes and alkylated aromatic compounds may be pyrogenic/petrogenic pollution as well as plastic production. Phenols and specific antioxidants and UV-absorbers can also be related to plastic production.

Little data is available on the role of microplastics as a vector for PCBs through the marine trophic levels and impact studies are required under controlled conditions. Benthic marine organisms such as the common shore crab and Norway lobster were exposed to PCB loaded microplastics under controlled laboratory conditions. In these experiments, 500–600 μm diameter polyethylene or polystyrene spheres were loaded with PCBs. The microspheres will pass the digestive tract without accumulation in the organism and egestion of the spheres was observed within two days after uptake. Within this research, it was shown that PCBs could desorb from the microspheres during the short period in the digestive system, but only a very small uptake of PCBs was observed for Norway lobster. No additional effect caused by the microspheres could be observed.

Work on plastic litter as a vector for bacteria had been carried out by Lisa Devriese, Caroline de Tender and Sara Maes (ILVO, BE). The possibility for microplastics and litter to act as a vector for bacteria and pathogens was suggested based on a bacterial screening on beach pellets, marine plastic litter and plastic beach litter. Diverse methods such as Next Generation Sequencing (NGS), TOPO TA cloning, PCR-DGGE were used to identify the bacterial communities of the different types of plastic.

Michiel Kotterman (NL) presented research on plastics. Next to monitoring the presence of plastics in the environment (as monitored by trawling; bottom and egg surveys), in biota (fish, fulmars and seals) the main research topic is to determine the role of microplastics with regard to contaminants. Are they a vector of contaminants, enhancing the uptake of contaminants by biota, or are they a sink for some contaminants due to their high affinity for some contaminants, lowering the exposure.

This is being investigated with lugworms under realistic conditions, micro-PS in contaminated sediments, and models for effects of plastic ingestions have been made. So far, plastics can be vector as well as sink, the effects under natural conditions are, from of risk assessment perspective, generally small. More data is required for proof and to improve the models. Therefore, research will be focussed on the net effects of plastic on the uptake of contaminants under natural conditions.

Jakob Strand (DK) presented the relationships between microplastic particles, sediment characteristics and contaminants in sediments from Danish waters based on a study on distribution of microplastic particles (38 μm – 5mm) in sediment in the Danish waters from the Baltic Sea towards the North Sea. The results indicate that normalisation of microplastic abundances to adequate sediment characters can reduce the variability caused by natural heterogeneity between samples and thereby increase the power of identifying more or less affected areas. Strong relationships between the content of microplastics in sediments and both %TOC and fine sediment fraction (<6 3µm) were found throughout the area supporting that microplastics will accumulate in sedimentary depositional areas – i.e. with parallels to organic pollutants sorbed to organic materials. Positive correlations were also established to contaminants, especially PAHs and to lesser extent to alkylphenols and phthalates in sediments. It could be due to co-variation with sources and TOC rather than due to chemical extraction of microplastic particles. However, at least antifouling agents like TBT in paint flakes from ship lanes and harbours can be one exception.

Bjørn Einar Grøsvik (NO) presented a collaboration project with the Polar Research Institute of Marine Fisheries and Oceanography (PINRO) in Russia. Co-workers on this study were Elena Eriksen (IMR, Norway) and Tatiana Prokhorova and Pavel Krivosheya (both PINRO, Russia). Since 2004 these institutes have collaborated on ecosystem based surveys in the Barents Sea. From 2010 registration of marine litter has been a part of this collaboration. Surface investigations and trawl catches have demonstrated highest occurrence of litter in the areas of intensive fishery and navigation. Plastic prevailed among observed litter. Other types of litter (metal, paper, rubber, textile, glass) were sporadically observed.

Taking into account the presentations given during the marine litter session as well as the available literature, the groups remark that there is currently insufficient information to assume that the uptake of chemical contaminants by marine biota through digestion of microplastics is significant. In some cases, enhanced uptake of plastic additives can occur, if these are not yet in equilibrium with the surrounding environment. More plastic uptake might also occur at locations where marine litter accumulates by marine gyres. Major problems of marine plastic pollution seem to be related to obstruction by and/or uptake of large amounts of plastics.

WGBEC as well as MCWG stress their interest to work further on the field of marine litter as well as microplastics. Both groups would be particularly be interested in further information on desorption studies in gastrointestinal tracts and work on uptake of chemical contaminants by organisms from marine litter. MCWG recommended to WGBEC to share new information with MCWG identifying plastics as a vector of enhanced contaminant transfer to biota.

The large amount of information provided through the presentations did not leave enough time to work on a comprehensive problem description. Activities in the field of marine litter have increased significantly, including a number of national and EU research projects and work in several fora towards marine litter monitoring in relation to MSFD. As described above, a separate ICES working group dedicated to marine litter has been proposed by members of WGBEC.

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Koelmans, A.A.; Besseling, E.; Foekema, E.M. 2014. Leaching of plastic additives to marine organisms. Environ. Pollut. 187, 49-54.

www.marlisco.eu

www.ilvo.vlaanderen.be/micro

http://www.cleansea-project.eu/drupal/index.php

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Annex 2: Agenda

1. Opening of the meeting;

2. Adoption of the agenda;

3. Timetable and appointment of rapporteurs;

4. (ToR a) Respond to requests for advice from Regional Seas Conventions as required;

5. (ToR b) Consider emerging issues of scientific merit and address knowledge gaps;

a. Oil toxicity to early life stages of fish

b. Ocean Acidification

c. Immunotoxicity

d. Novel monitoring techniques

e. Thiamine deficiency and vitamin interactions

6. (ToR c) Review status of publications and consider requirements for new publications;

a. ICES TIMES

b. Other ICES publications

c. Peer review publications

7. (ToR d) Conduct assessment of data as required;

a. Quality assurance data from method intercomparison trials

b. Integrated assessment of monitoring data

8. (ToR e) Respond to requests for advice from the Data Centre;

9. (ToR f) Development and harmonisation of methodologies for marine monitoring and surveillance including:

a. Integrated assessments

b. Environmental risk assessment

c. Review and develop assessment criteria for biological effects methods

d. Report on national monitoring programmes for biological effects

10. (ToR g) Address issues in relation to novel and emerging contaminants (e.g. pharmaceuticals, nanoparticles, toxicity of mixtures etc)

a. Pharmaceuticals and recreational drugs in the marine environment

b. Biocides in the marine environment

c. Nanoparticles in the marine environment

11. (ToR h) To evaluate the results of monitoring and research activities on plastic litter, especially microplastics and associated chemical contaminants in the marine environment abroad in regard to:

a. Status on development of tools to quantify and qualify (micro) plastics in marine organisms, e.g. fish, turtles, crustaceans, marine mammals, and sea birds

b. Results of impact assessment surveys and research projects of microplastics and non-plastic microparticles in marine organisms from all trophic levels

c. Evidence of bioaccumulation, toxicity and of adverse physical and chemical effects of microplastics and associated contaminants on marine organisms, populations and communities. This would include the full range of marine organisms from bacteria to turtles, marine mammals and sea birds

d. Evidence of microplastics and associated contaminants to transfer through marine food chains

12. Any other business;

13. Recommendations and action list;

14. Adoption of the report and closure of the meeting

WGBEC will report on the activities of 2015 (the final year) by 30 April 2015 to SSGHIE.

Annex 3: WGBEC Self-Evaluation and draft Multi-Annual Resolution

WGBEC Working Group self-evaluation 2015

1) Working group of Biological Effects of Contaminants (WGBEC)

2) Year of appointment: 2013

3) Current Chairs:

Bjørn Einar Grøsvik (IMR) and Ketil Hylland (UiO)

4) Venues, dates and number of participants per meeting:

- San Pedro del Pinatar, Spain, 10-15 March 2013, 15 participants.
- ICES Headquarters, Copenhagen, Denmark, 3-7 March 2014, 15 participants.
- Bergen, Norway, 9-13 March 2015, 17 participants.

5) If applicable, please indicate the research priorities (and sub priorities) of the Science Plan to which the WG make a significant contribution.

-9. Identify indicators of ecosystem state and function for use in the assessment and management of ecosystem goods and services.- How contaminants affect ecosystem goods and services.

-11. Develop methods to quantify multiple direct and indirect impacts from fisheries as well as from mineral extraction, energy generation, aquaculture and other anthropogenic activities and estimate the vulnerability of ecosystems to such impacts. - With a focus on sublethal effects of stressors.

-13. Develop indicators of pressure on populations and ecosystems from human activities such as eutrophication, contaminants and litter release, introduction of alien species and generation of underwater noise. - Key activity of WGBEC - integrated chemical and biological monitoring.

-16. Quantify and map biological, ecological and environmental values with an aim to optimize ecosystem use and minimize environmental impacts in relation to ecosystem carrying capacity. - Related to contaminants.

-17. Develop science in support of advisory needs in marine aquaculture systems, minimizing environmental impacts and integrating other marine sectors. - Related to pharmaceuticals and biocides used.

- 19. Identify issue based ecosystem questions relevant to science and management needs that can be addressed by developing IEA's. - Related to contaminant impacts.

-20. Provide priorities and specifications for data collection frameworks supporting IEA's. - Contaminant-related.

-27. Identify knowledge and methodological monitoring gaps and develop strategies to fill these gaps. -The group continuously assesses and contributes to the development of monitoring strategies for contaminants.

-28. Promote new technologies and opportunities for observation and monitoring and assess their capabilities in the ICES context. - Related to stressors, particularly contaminants.

6) In bullet form, list the main outcomes and achievements of the WG since their last evaluation. Outcomes including publications, advisory products, modelling outputs, methodological developments, etc. *

-Publications:

-Balk L, Hylland K, Hansson T, Berntssen MH, Beyer J, Jonsson G, Melbye A, Grung M, Torstensen BE, Børseth JF, Skarphedinsdottir H, Klungsøyr J. 2011. Biomarkers in natural fish populations indicate adverse biological effects of offshore oil production. *PLoS One*. 6(5):e19735.

-Bakke T, Klungsøyr J, Sanni S. 2013. Environmental impacts of produced water and drilling waste discharges from the Norwegian offshore petroleum industry. *Mar Environ Res*, 92, 154-169.

-Blanchard A, Hauge KH, Gisle Andersen G, Fosså JH, Grøsvik BE, Handegard NO, Kaiser M, Meier S, Olsen E, Vikebø F. 2014. Harmful routines? Uncertainty in science and conflicting views on routine petroleum operations in Norway. Marine Policy, 43: 313-320.

-Guzzo, MM, Eckbo, NH, Gabrielsen, GW, Fisk, AT, Hylland, K, Borgå, K. 2014. Persistent organic pollutant concentrations in fledglings of two arctic seabird species, Environ Pollut, 184, 414-418.

-Hauge KH, Blanchard A, Andersen G, Boland R, Grøsvik BE, Daniel Howell D, Meier S, Olsen E, Vikebø F. 2014. Inadequate Risk Assessments – A Study on Worst-Case Scenarios Related to Petroleum Exploitation in the Lofoten Area. Marine Policy. 44:82-89.

-Holth, TF, Eidsvoll, DP, Farmen, F, Sanders, MB, Martínez-Gómez, C, Budzinski, H, Burgeot, T, Guilhermino, L, Hylland, K. 2014. Effects of water accommodated fractions of crude oils and diesel on a suite of biomarkers in Atlantic cod (*Gadus morhua*), Aquat Toxicol, 154, 240-252.

-ICES/OSPAR 2011. Report of the Study Group on Integrated Monitoring of Contaminants and Biological Effects (SGIMC). ICES CM 2911/ACOM 30. pp 265.

-Jörundsdóttir, HO, Jensen, S, Hylland, K, Holth, TF, Gunnlaugsdóttir, H, Svavarsson, J, Ólafsdóttir, A, El-Taliawy, H, Rigét, F, Strand, J, Nyberg, E, Bignert, A, Hoydal, KS, Halldórsson, HP. 2014. Pristine Arctic: Background mapping of PAHs, PAH metabolites and inorganic trace elements in the North-Atlantic Arctic and sub-Arctic coastal environment, Sci Total Environ, 493, 719-728.

-MAGRAMA. 2012. Estrategias Marinas- Evaluación inicial, buen estado ambiental y objetivos ambientales. DEMARCACIÓN MARINA NORATLANTICA. DESCRIPTORES DEL BUEN ESTADO AMBIENTAL 8. CONTAMINANTES Y SUS EFECTOS BIOLOGICOS. Autores (IEO): Lucía Viñas, Juan Bellas, Mª Victoria Besada, Mª Ángeles Franco, José Fumega, Amelia González-Quijano.Unidad solicitante: Dirección General de Sostenibili-dad de la Costa y del Mar. Ministerio de Agricultura, Alimentación y Medio Ambiente. http://www.magrama.gob.es/es/costas/temas/estrategias-marinas/IV_D8_Noratlantica_tcm7-207277.pdf

-MAGRAMA. 2012. Estrategias Marinas- Evaluación inicial, buen estado ambiental y objetivos ambientales. DEMARCACIÓN MARINA DEL ESTRECHO Y ALBORÁN. DESCRIPTO-RES DEL BUEN ESTADO AMBIENTAL 8. CONTAMINANTES Y SUS EFECTOS BIOLO-GICOS. Autores (IEO): José Benedicto, Juan Antonio Campillo, Beatriz Fernández, Concepción Martínez-Gómez, Víctor M. León. Unidad solicitante:

Dirección General de Sostenibilidad de la Costa y del Mar. Ministerio de Agricultura, Alimentación y Medio Ambiente. http://www.magrama.gob.es/es/costas/temas/estrategias-marinas/IV_D8_Estrecho_y_Alboran_tcm7-207246.pdf

-MAGRAMA. 2012. Estrategias Marinas- Evaluación inicial, buen estado ambiental y objetivos ambientales. DEMARCACIÓN MARINA LEVANTINO-BALEAR. DESCRIPTORES DEL BUEN ESTADO AMBIENTAL 8. CONTAMINANTES Y SUS EFECTOS BIOLÓGICOS. Autores (IEO): José Benedicto, Juan Antonio Campillo, Beatriz Fernández, Concepción Martínez-Gómez, Víctor M. León. Unidad solicitante: Dirección General de Sostenibilidad de la Costa y del Mar. Ministerio de Agricultura, Alimentación y Medio Ambiente. http://www.magrama.gob.es/es/costas/temas/estrategias-marinas/IV_D8_Levantino-Balear_tcm7-207261.pdf

-Oliveira, M, Ribeiro, A, Hylland, K, Guilhermino, L. 2013 Single and combined effects of microplastics and pyrene on juveniles (0+ group) of the common goby *Pomatoschistus microps* (Teleostei, Gobiidae), Ecol Ind, 34, 641-647.

-Olsen GH, Klok C, Hendriks J, Geraudie P, De Hoop L, De Laender F, Farmen E, Grøsvik BE , Hansen BH, Hjorth M, Jansen CR, Nordtug T, Ravagnan E, Viaene K, Carroll J. 2013. Toxicity data for modeling impacts of oil components in an Arctic ecosystem. Marine Environmental Research, 90; 9-17.

-Sørhus E, Edvardsen RB, Karlsen Ø, Nordtug T. Meeren, T, Thorsen A, Harman C, Jentoft S, Meier, S. 2015. Unexpected interaction with dispersed crude oil droplets drives severe toxicity in Atlantic haddock embryos. *PLoS ONE*. DOI:10.137/journal.pone.0124376. pp21.

-Vethaak, A.D., Pieters, J., Jol, J.G. (2009). Long-term trends in the prevalence of cancer and ma-jor diseases among flatfish in the S.E. North Sea as indicators of changing ecosystem health. Env. Sci. Techn. 43: 2151–2158.

-Vikebø F, Rønningen P, Lien VS, Meier S, Kristiansen T. 2013. Spatio-temporal overlap of oil spills and early life stages of fish. ICES Journal of Marine Science. Doi:10.1093/icesjms/fst131. pp 12.

-Vikebø F, Rønningen P, Meier S, Grøsvik BE, Lien V. 2015. Dispersants have limited effects on exposure rates of oil spills on fish eggs and larvae in shelf seas. Environmental Science & Technology. 49: 6061-6069. http://dx.doi.org/10.1021/acs.est.5b00016.

-**Advisory products:**

To OSCAR MIME and HASEC on how integrated assessments of effects of contaminants (SGIMC approach, ICES/OSPAR 2011) can be used in monitoring programmes.

ICON integrated assessment demonstration programme (Integrated assessment of contaminant impacts on the North Sea (ICON). Interim final assessment for consideration by MIME 2012: doc OPSAR MIME 12/3/5-E (L).)

Integrated assessment of the Firth of Forth area of Scotland using the SGIMC framework (Robinson, C.D., Gubbins, M.J., Lyons, B.P., Bignell, J., Bean, T., MacNeish, K., Dymond, P., Dobson, J., and Thain, J.E. Assessing Good Environmental Status for Descriptor 8 – an integrated assessment of contaminants and their biological effects across multiple matrices in the Firth of Forth, Scotland ICES CM2012:G).

Integated assessment of the Humber estuary, UK
http://berlin.setac.eu/embed/Berlin/ET_extended_abstracts_Part2.pdf

To ICES Data Centre on data submission issues:

-Methodological quality assessments:

- o Interlaboratory method calibration of neutral red retention time assay (2013)
- o Interlabiratory method calibration of PAH metabolites in bile (2014)
- o Interlaboratory method calibration of EROD activity (2014)

7) Has the WG contributed to Advisory needs? If so, please list when, to whom, and what was the essence of the advice.

To OSCAR MIME and HASEC on how integrated assessments of effects of contaminants (SGIMC approach, ICES/OSPAR 2011) can be used in monitoring programmes.

-ICON integrated assessment demonstration programme (Integrated assessment of contaminant impacts on the North Sea (ICON). Interim final assessment for consideration by MIME 2012: doc OPSAR MIME 12/3/5-E (L).)

8) Please list any specific outreach activities of the WG outside the ICES network (unless listed in question 6). For example, EC projects directly emanating from the WG discussions, representation of the WG in meetings of outside organizations, contributions to other agencies' activities.

Some of the WGBEC members are members of a consortium receiving grant for studies on ecological effects microplastics from the call from JPI Oceans in 2015.

Grøsvik has participated in the EU project STAGES (Science and Technology Advancing Governance on Good Environmental Status).

Grøsvik has been appointed as member of Pool of Experts for the United Nations World Ocean Assessment. Member of the writing team on Chapter 21-Offshore hydrocarbon industries.

9) Please indicate what difficulties, if any, have been encountered in achieving the workplan.

-The group has experienced that the format needed to submit data to the ICES Data Centre seems complicated and express that a simplification of this format or process would be helpful.

Future plans

10) Does the group think that a continuation of the WG beyond its current term is required? (If yes, please list the reasons)

Yes, we need a continuous work to assess and quality assure the biological effect methods already suggested by the working group and to develop new methods to better describe if contaminant levels in the marine environment pose a risk to organisms.

It is important to be informed on, and discuss national monitoring programmes in order to better assess whether Good Environmental Status has been achieved according to descriptor 8 of MSFD.

A Workshop under the subject "Are contaminants in eels contributing to their decline?" has been suggested between WGBEC and WGEEL (WKBECEEL) planned to be held on 25–27 January 2016 (resolution approved in 2014).

In the suggested next multiannual ToRs we have suggested to obtain better knowledge on the following themes:

Review effecs of chronic oil exposure
Review marine seabird ecotoxicology
Review marine mammal ecotoxicology
Review effects of contaminants on community composition
Develop methods to evaluate effects of acute spills
Develop methods to evaluate effects of ocean acidification
Review interactions between essential nutrients or vitamins and contaminants in marine organisms
Review progress with marine plastic ecotoxicity
Review and update knowledge of environmental interactions and combined stressors
Review effects of emerging contaminants
Review the use of passive samplers and dosing

11) If you are not requesting an extension, does the group consider that a new WG is required to further develop the science previously addressed by the existing WG.

(*If you answered YES to question 10 or 11, it is expected that a new Category 2 draft resolution will be submitted through the relevant SSG Chair or Secretariat*.)

12) What additional expertise would improve the ability of the new (or in case of renewal, existing) WG to fulfil its ToR?

13) Which conclusions/or knowledge acquired of the WG do you think should be used in the Advisory process, if not already used? (please be specific)

-Improved methods on how to assess that concentrations of contaminants are at levels not giving rise to pollution effects (Descriptor 8, MSFD). This is a continuing process and needs the work of WGBEC for discussions on assessment criteria and advisory work.

WGBEC draft multi-annual Resolution 2016–2018

The **Working Group on Biological Effects of Contaminants** (WGBEC), chaired by Bjørn Einar Grøsvik, Norway, and Ketil Hylland, Norway, will work on ToRs and generate deliverables as listed in the Table below.

	Meeting dates	Venue	Reporting details	Comments (change in Chair, etc.)
Year 2016	7-11 March	Lisbon, Portugal	Interim report by 15 May to SSGEPI	
Year 2017			Interim report by	
Year 2018			Final report by	

ToR descriptors

ToR	Description	Background	Science Plan topics addressed	Duration	Expected Deliverables
a	Review effecs of chronic oil exposure on marine organisms (desk study)	Oil pollution is one of the major chemical and physical challenges for coastal (and to some extent offshore) marine ecosystems worldwide. WGBEC has a unique competence in this field, having contributed to developing guidelines for OSPAR and individual countries concerning monitoring, as well as performing research (see reference list for the group. Although has been shown that early life stages of fish are particularly susceptible to oil pollution, there is limited understanding of the extent to which such pollution impacts natural fish populations and virtually no knowledge of effects on invertebrate early life stages.	11, 13	year 2	Manuscript submitted to a peer-reviewed scientific journal
b	Review available studies on marine seabird ecotoxicology (desk study)	Seabird populations are decreasing worldwide and there is concern that chemicals may be at least partly involved. WGBEC has members that have been and are involved in research on effects of contaminants on seabirds. There is no parallel activity elsewhere in the scientific community and there is certainly a need to summarise what we know, future challenges and research directions to understand how chemicals may affect seabirds.	11, 13	year 2	Summarise in annual report (yr 1) Manuscript submitted to a peer-reviewed scientific journal (yr 2)
c	Review available studies on marine	WGBEC has previously not been heavily involved in research on marine mammals,	11, 13	year 3	Summarise in annual report

	mammal ecotoxicology (desk study)	but new members to the group (and some old) have ongoing projects in this area. There is a need to evaluate effects of toxic chemicals on marine mammals worldwide, not only in the Arctic or Antarctic.			(yr 1) Manuscript submitted to a peer-reviewed scientific journal (yr 2)
d	Review effects of contaminants on community composition	There has been a long discussion as to whether community composition is sufficiently contaminant-specific to merit inclusion on a list of recommended methods for contaminant-oriented assessment. There is a clear need for further discussion on this topic, which should also involve other ICES working groups (e.g. benthic ecology).	11, 13	year 3	Manuscript submitted to a peer-reviewed scientific journal
e	Develop methods to evaluate effects of acute spills on marine organisms (desk study)	Acute spills have conceivably different patterns of effects than diffuse, chronic inputs. This is a fundamental question in environmental science, but very important for environmental assessment. WGBEC is well placed to contribute a review and advice in this area.	11	year 2	Manuscript submitted to a peer-reviewed scientific journal
f	Develop methods to evaluate effects of ocean acidification on marine organisms	Other working groups and research activities address this issue, but WGBEC is of the opinion that the group can contribute in developing a monitoring methodology.	11	year 1, 2, 3	Summarise in annual report
g	Review interactions between essential nutrients or vitamins and contaminants in marine organisms (desk study)	This is an issue which has surfaced the past decade and which may conceivably be of major importance in how contaminants affect marine organisms. WGBEC have members that are at the forefront of research in this area and hence well placed to contribute.	11	year 1	Summarise in annual report
h	Review progress with marine plastic ecotoxicity to marine organisms	This is another issue which is a hot research topic and with much international interest. WGBEC members are involved nationally and internationally. The group has competence on relevant issues and should keep abreast of developments.	11	year 2	Summarise in annual report
i	Review and update knowledge of environmental interactions and combined stressors in marine ecosystems (desk study)	Environmental interactions and combined effects are crucial issues in the assessment of contaminant effects. Members of WGBEC have addressed the issues over the past two decades and are in a position to provide significant contributions to the scientific community and environmental managers.	11	year 3	Summarise in annual report (yr 1) Manuscript submitted to a peer-reviewed scientific journal (yr 2)
j	Review effects of emerging contaminants on marine organisms	This issue has been discussed at every WGBEC meeting the past decade (or more). It should be on the work plan to ensure the group is kept abreast of current	11	year 1	Summarise in annual report

	(desk study)	developments in the field.			
k	Review the use of passive samplers and dosing in marine ecotoxicity studies	An issue more directly relevant to MCWG, this item is important for the development of effect-directed monitoring and testing of chemicals and hence of relevance to WGBEC. Members of the group are involved.	28	year 3	Summarise in annual report

Summary of the Work Plan

Year 1	The development of methods to assess effects of acidification is an ongoing issue, to be reported each year. Effects of emerging contaminants will be finalised (there has been activity on this issue over the last 3-year period). The group will finalise recently initialised work on interaction between contaminants and vitamins. Work will also focus on items to be reported in year 2 with status updates this year (a, b, c).
Year 2	This is an important reporting year during this 3-year cycle with a final reporting, i.e. review papers, on items a, b (chronic oil exposure, seabird toxicity), in addition to status updates for items f and h (effects of acidification and plastics).
Year 3	Final reporting (i.e. review papers) on items c, d and i (marine mammal ecotoxicology, effects on communities and interactions/combined effects) as well as status reports for ocean acidification.

Supporting information

Priority	The current activities of this Group will lead ICES into issues related to the ecosystem effects of fisheries, especially with regard to the application of the Precautionary Approach. Consequently, these activities are considered to have a very high priority.
Resource requirements	The research programmes which provide the main input to this group are already underway, and resources are already committed. The additional resource required to undertake additional activities in the framework of this group is negligible.
Participants	The Group is normally attended by some 15-20 members and guests.
Secretariat facilities	None.
Financial	No financial implications.
Linkages to ACOM and groups under ACOM	There are no obvious direct linkages.
Linkages to other committees or groups	There is a very close working relationship with all the groups of SSGEPI. It is also relevant to the Marine Chemistry Working Group, Working Group on Marine Sediments, Working Group on Seabirds and Working Group on Marine Mammal Ecology.
Linkages to other organizations	MIME and HASEC, HELCOM, MED POL

Annex 4: Contact list

Contacts from EU Members State nominees to the Expert Network on contaminants (and from the Regional Seas Conventions) invited to respond to the review of the MSFD Commission Decision document (EU/477/2010) by the Joint Research Centre, Ispra, Italy. Milieu Ltd. was employed to review the MS responses to the Commission regarding their activities under MSFD Articles 8, 9, 10 and 11. *attendee at one or more of the Ispra meetings.

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Estonia		
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Annex 5: List of manuscript submitted for ICON special issue in Mar Environ Res

Hylland *et al.*, Assessing contaminant impacts in European marine ecosystems: the ICON workshop

Robinson *et al.*, Assessment of contaminant concentrations in marine sediments, fish and mussels sampled from the North Atlantic and European regional seas within the ICON project

Lang *et al.*, Methylmercury in dab (*Limanda limanda*) from the North Sea, Baltic Sea and Icelandic waters: relationship to host-specific variables

Kammann *et al.*, PAH metabolites in fish bile: from the Seine Estuary to Iceland

Vethaak *et al.*, *In vitro* and *in vivo* toxicity profiling of marine sediments from the ICON survey

Broeg *et al.*, Lysosomal membrane stability in the liver of dab (*Limanda limanda*) – Applicability and reliability of assessment criteria under concrete contaminant-related monitoring conditions of coastal, estuarine and offshore locations

Carney Almroth *et al.*, Is oxidative stress evident in dab (Limanda limanda) in the North Sea?

Hylland *et al.*, Genotoxicity in dab (*Limanda limanda*) and haddock (*Melanogrammus aeglefinus*) from European seas

Lang *et al.*, Diseases of dab (*Limanda limanda*): analysis and assessment of data on externally visible diseases, macroscopic liver neoplasms and liver histopathology at offshore sites in the North Sea, Baltic Sea and off Iceland

Burgeot *et al.*, Integrated assessment of contaminant impacts in the Seine estuary

Lyons *et al.*, Determining Good Environmental Status under the Marine Strategy Framework Directive: case study for descriptor 8 (chemical contaminants)

Hylland *et al.*, Impacts of contaminants in European marine areas: an integrated assessment

Annex 6: ToRs for Joint Workshop of the Working Group on Eel and the Working Group on Biological Effects of Contaminants (WKBECEEL)

2014/2/SSGEPD07 A **Workshop** of the **Working Group on Eel** and the **Working Group on Biological Effects of Contaminants** (WKBECEEL) will be established under the subject "Are contaminants in eels contributing to their decline?". WKBECEEL will be chaired by Caroline Durif*, Norway, and Bjørn Einar Grøsvik*, Norway, and will meet in Os, Norway, 25–27 January 2016 to:

a) Describe the spatial and temporal trends in concentrations of "traditional" and/or "emerging" contaminants in eel (but mainly refer to figures available from WGEEL 2008–2013).

b) Describe the potential impacts of contaminants on reproduction in the European eel, based on science of eel and what can be learned from other species models (including endocrine disruption, effect on sex ratio, maternal transfer of bioaccumulated contaminants toward the eggs and effects on the larvae).

c) Describe the potential impacts of contaminants on lipid metabolism and migration in the European eel based on eel science and what can be learned from other species

d) Review the impacts of contaminants on the genetics of the European eel.

e) Explore whether there is experience with assessing/qualifying the bioaccumulation + fitness status in other species, which can be helpful for the eel's quality assessment (Eel Quality Index) and to quantify the impact of eel quality.

WKBECEEL will report by DATE (via SSGEPD) for the attention of WGEEL, WGRECORDS and SCICOM.

Supporting information

Priority	During previous meetings WGEEL (2008-2013) made considerable progress i understanding and describing the potential impact of contaminants on th European eel stock. During the last sessions WGEEL 2012 and WGEEL 2013 indicated that the W would clearly benefit from a joint cooperation with experts from other ICE WGs, and specifically WGBEC. The experience and knowledge concerning th effect of contaminants in other species, as present within WGBEC, is anticipate to be very beneficial to make further progress in understanding the role o contaminants in the eel stock decline.
Scientific justification	The stock of the European eel *Anguilla anguilla* is in decline and there is a increasing awareness that poor health status due to contaminants might be a key element in this decline and might be a hindrance to recovery. Sever studies have recently been initiated to study the degree and the effects o pollution on the eel, resulting in an increasing quantity of information tha demonstrates the negative impact of pollution on eel. These advances in the science of the effects of contaminants on the eel have bee reviewed recently (e.g. Geeraerts *et al.*, 2010; by Elie and Gerard, 2009, and WGEEL 2008-2012). However, essential issues to assess the importance of eel quality for reproductive success, such as to evaluate the effect of specif

	contaminants on the ability for eel to migrate and to reproduce have still to be developed. The joint workshop will review all sources information (including work on other species) to better understand ho contaminants in eels contribute to their decline.
Resource requirements	
Participants	WGEEL and WGBEC Working Group Participants, and other experts. Th Workshop is anticipated to be attended by some 15-20 members and guests.
Secretariat facilities	Sharepoint
Financial	
Linkages to advisor committees	WGEEL,WGBEC and ACOM
Linkages to othe committees or groups	WGRECORDS, SSGEPD, SCICOM
Linkages to othe organizations	FAO EIFAAC, GFCM, EU DG MARE, EU DG ENV

Annex 7: An assessment tool for monitoring liver cancer in marine environment (a preliminary report)

John Bignell and Allan Reese (UK) submitted a proposal for a new assessment tool for monitoring liver cancer in marine fish. The work has primarily arisen from previous concerns raised that liver cancer might simply be a surrogate for age and that contaminants may not be the sole cause. This is particular concerning when the age distribution of one of the OSPAR region's primary biomarker species, the common dab (*Limanda limanda*), is considered. Analysis of data from dab of the size range 20–24 cm, as recommended by ICES in biological effects monitoring programmes (Feist *et al.*, 2004), revealed that age can vary considerably. Box plots of length against otolith age for each sex confirms that growth is continuous at the population level but for individual fish is a poor estimator of age above 3 years (Figure 1). Subsequent work led by Cefas also revealed that although cancer certainly increases with age (which could be a result of continued contaminants exposure), the age of onset is accelerated at certain locations i.e. fish get cancer younger (Stentiford, *et al.*, 2010). Whilst this is meaningful concerning an individual i.e. the earliest age cancer was observed at a sampling site; it does not inform us a great deal about the population as a whole. As such there is a requirement for an assessment tool which is complementary to that used elsewhere i.e. the Fish Disease Index (FDI); that is able to consider the effects of age into an assessment. Cefas are currently working on a logistic regression model that incorporates a large histopathology dataset (with corresponding age determination) from 2004–2013. In its simplest form, it provides a national liver cancer model of England and Wales for the first time. The model also allows "site to site" and "year to year" comparisons for the assessment of liver cancer.

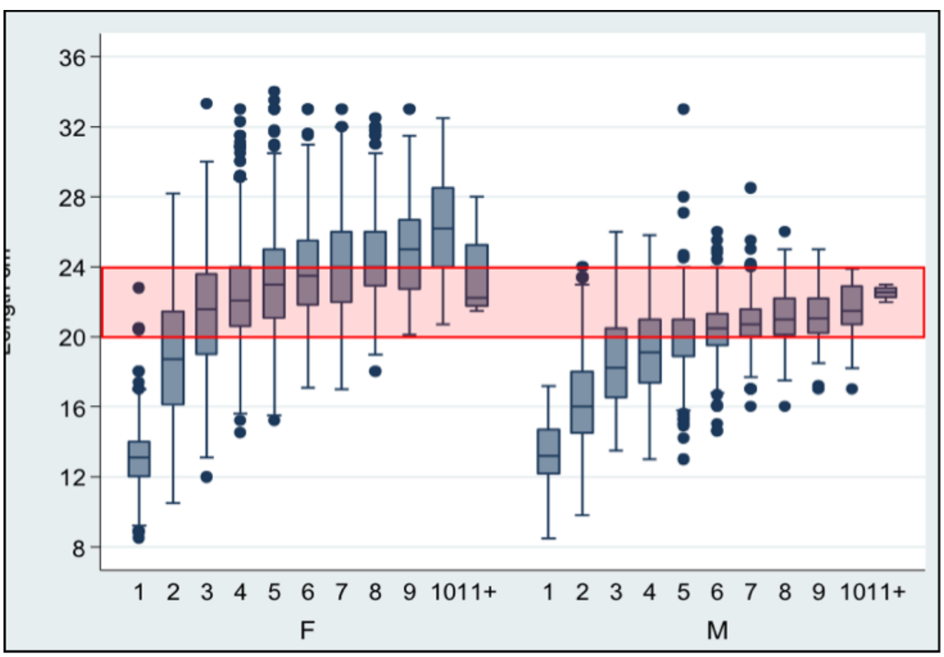

Figure 1. Age distribution of common dab (*Limanda limanda*) sampled between 2006 and 2013 (n= 7546) as part of the CSEMP in England and Wales.

Briefly, dab were sampled from CSEMP sampling stations in the Irish and North Sea during the summer from 2004–2013 (n=7546: currently awaiting incorporation of 2006 data). Following euthanasia, liver and gonad tissues were dissected and processed for histological analysis. Otoliths were also obtained from each fish and sectioned for age determination. Liver was analysed using light microscopy according to ICES TIMES protocols (Feist *et al.*, 2004). For assessment tool purposes, liver histopathology data concerning neoplasms were consolidated into the presence or absence of cancer (benign and malignant neoplasms). A standard logistic regression model was applied to data to produce a national model for liver cancer.

Logistic regression is an appropriate model to compare the cancer risks between sexes, locations, or time periods. The regression equation is a straight line on the logit scale and becomes S-shaped when transformed back to the probability (p) scale. The logistic model predicts the percentage of fish expected to have liver cancer at a specified age in England and Wales i.e. what is the risk of cancer. The model allows interrogation of the data at several hierarchy levels regarding geographical region. Initial observations demonstrate that fish from the MSFD Irish Sea region are adversely affected more than fish from the Greater North Sea MSFD region. This is demonstrated by the larger proportion of fish having liver cancer across the entire Irish Sea population sampled, compared to the North Sea. The model benefits from the ability to reveal which national MERMAN regions are potentially driving geographical differences i.e. hotspot areas such as Cardigan Bay (figure Xc). Furthermore, it allows geographical regions to be assessed for improvements regarding liver cancer prevalence. However, care should be exercised. For example, figure Xd appears to demonstrate that the North Sea population appears to be worsening regarding liver cancer prevalence when comparing the earliest (2004) and latest sampling events (2013). This is potentially the result of a random sampling event, although other parameters might be influencing this change i.e. sex ratios of sampled fish. Nonetheless, it is crucial that long term datasets are used to investigate trends of significance before drawing conclusions using this method.

This report describes preliminary results observed through the development of a new assessment tool for liver cancer. The sampling method and data show great promise for monitoring the health of the sea and comparing between regions and over time.

These results indicate that there is a significant but small difference in age distributions between the Western seas and the Greater North Sea. Subsequent annual samples from just one region can be compared with previous results from that region. It also suggests that year-to-year variation may be random but, if data collect continues, then subsequent years may confirm regional trends.

The power of the model can become compromised by ad hoc changes to the sampling scheme i.e. reduced frequency and numbers of samples per monitoring year. As a general principle, at least 6 or 7 years' data is required to demonstrate a trend that is gradual i.e. less than 20% annual rate change.

The next steps will be to discuss amongst experts how best to assess UK data and how this might compare to data across the OSPAR region when using the same approach. Several approaches are available including, but not limited to

(a) Assessment of all OSPAR data to the UK logistic model due to the wide ranging prevalence of liver cancer observed.

(b) Assessment against an OSPAR logistic model that incorporates ongoing age and cancer data from different regions i.e. Germany.

(c) Assessment between national logistic models, although regions may not be directly comparable or

(d) Assessment against a true reference baseline i.e. background response.

References

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