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Theme Session on Biological Effects of Contaminants in Marine Pelagic Ecosystems

An ICES workshop on biological effects in pelagic ecosystems (BECPELAG): summary of results and recommendations

K. Hylland*, G. Becker**, J. Klungsøyr***, T. Lang[^], A. McIntosh^{^^}, B. Serigstad***, J.E. Thain^{^^^}, K.V. Thomas^{^^^}, T.I.R. Utvik[□], D. Vethaak^{□□}, W. Wosniok^{□□□}

*NIVA, P.O.Box 173, Kjelsas, N-0411 Oslo, Norway [ketil.hylland@niva.no]

**BSH, Bernhard-Nocht-Str. 78, D-20359 Hamburg, Germany

***IMR, P.O.Box 1870, N-5817 Bergen, Norway

[^]BFA-Fi, Deichstrasse 12, D-27472 Cuxhaven, Germany

^{^^}FRS, P.O. Box 101, Victoria Road, Aberdeen AB1X 9DB, UK

^{^^^}CEFAS, Burnham-on-Crouch, Remembrance avenue, Essex CM0 8HA, UK

[□]Norsk Hydro, Environmental Section, Sandsliveien 90, N-5020 Bergen, Norway

^{□□}RIKZ, P.O.Box 8039, 4330 EA Middelburg, Netherlands

^{□□□}Universität Bremen, Postfach 330 440, D-28334 Bremen, Germany

Abstract

During and after three previous workshops, biological effects methods have been compared and refined. The first was held in Oslo in 1988, the second at Bermuda in 1990, and the third in Bremerhaven in 1992. All three workshops were held under the auspices of ICES and/or UN (GEEP, IOC). This activity is important insofar that it has stimulated research into the use of biological effects methods to monitor pollution impacts in marine ecosystems and contributed towards a framework for general and contaminant-specific monitoring.

The objective of the BECPELAG workshop was to bring together scientists involved in relevant work in a practical workshop in order to assess the ability of selected methods to detect biological effects of contaminants in pelagic ecosystems and this is described in the overview of the project (session X first paper). In addition to biological analyses being performed by participants, laboratories were contracted to perform the chemical analyses required and hydrographers involved to provide models of water movement prior to and during sampling. The development of methods to assess the effects of contaminants in pelagic systems will be important for the development of future monitoring programmes in coastal waters and to understand impacts of oil production in the North Sea.

This paper will consider the wide range (ca. 40) of methods used during the workshop and provide a view on the methodologies and science behind the BECPELAG and how the methods could be used to provide a platform to develop a strategy for future

monitoring and management of pelagic ecosystems. The results support an integrated chemical and biological approach. Furthermore, the collection of wild organisms should be supported by the use of caging due to the large variability in wild specimens. It may be possible to recommend single methods for cost effectiveness but added value may be achieved by using a combination of methods with little increase in cost. The added value of using an integrated array of tools will also be presented.

Introduction and objectives

There is a widely recognised need for methods to detect and quantify effects of contaminants in pelagic ecosystems, not least in relation to offshore oil- and gas-production activities. In the past few decades, the main focus of biological effect methods development has been on benthic organisms and systems. Progress with such methods has been advanced as the result of practical workshops. The first was held in Oslo/Langesund/Solbergstrand (Bayne et al., 1988), the second at Bermuda (Addison & Clarke, 1990) and the third in Bremerhaven (Stebbing & Dethlefsen, 1992). These three workshops stimulated research into the use of biological effects methods to monitor pollution impacts in marine ecosystems and contributed towards a framework for general and contaminant-specific monitoring (JAMP, 1998a,b).

Whereas there has been substantial progress in developing methods to assess pollution effects in benthic systems, there is still a lack of agreed methods to evaluate biological effects in the water column. There are many sources of contaminants to pelagic ecosystems including coastal inputs, dumping and long-range transport by ocean currents and the atmosphere. In areas of oil- or gas-production, there will be inputs of large volumes of produced water. The composition of produced water varies considerably from well to well, but generally contains mono- and polycyclic aromatic hydrocarbons, other organic contaminants and trace metals.

The objective of BECPELAG was to bring together scientists involved in relevant work in a practical workshop in order to assess the ability of selected methods to detect biological effects of contaminants in pelagic ecosystems under uniform and standardised conditions. The methods are being assessed for their applicability in monitoring programmes and the results from the BECPELAG workshop are currently being used to develop the water column monitoring programme for Norwegian offshore activities. This paper will give an overview of results from the chemistry programme, on biological effects in field-collected and caged organisms and bioassay results. In addition, recommendations and promising research directions will be presented.

A more extensive background for the workshop can be found in Hylland (2000), Hylland *et al.* (2002) or at the project web-site (<http://www.niva.no/pelagic/web>).

Hydrography

Hydrographical modelling was done for both study areas. Whereas the exposure situation appears to have been stable at locations in the Statfjord transect (at least during the cage deployment), the current regime was more variable at the German Bight locations. Especially the outer stations in the transect appeared to have been affected by different water masses. The main exposure at the German Bight stations would nevertheless be from the rivers Elbe and Weser.

The chemistry programme

A wide range of contaminants was analysed in the BECPELAG chemistry programme. The main focus was on biological matrices, i.e. fish (herring, saithe, mackerel), blue mussel, caged

cod and zooplankton, but extracts (SPMDs, produced water) were also analysed for e.g. PAHs and alkylphenols.

PAHs in caged mussels decreased along both transects, but overall concentrations were higher in the oil-rig transect (Figure 1). The somewhat elevated concentration at station G4 in the German Bight transect is probably related to the proximity of a Danish gas-field.

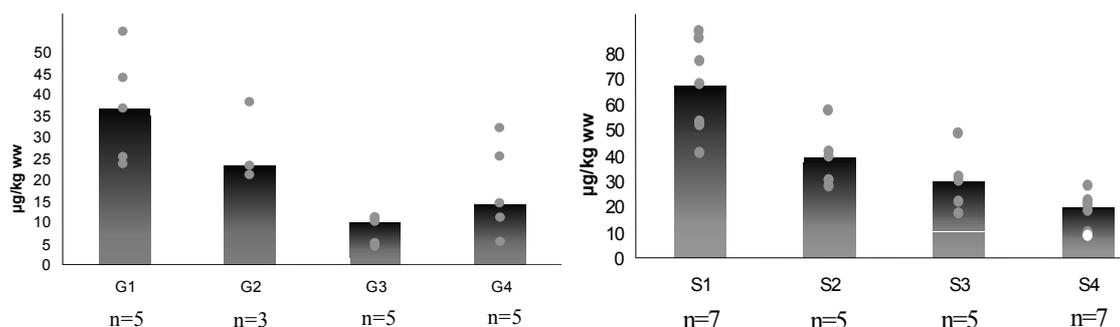


Figure 1. Sum PAH in blue mussels caged at the indicated locations; median and individual values shown. German Bight – left panel, Statfjord – right panel. G1 is closest to the Weser/Elbe estuaries, S1 closest to the platform. Data from NIVA.

In addition to PAHs, PCBs in caged mussels also decreased from G1 to G4 in the German Bight transect. Perhaps surprisingly, there were no clear trends for contaminants in caged cod. Also somewhat surprisingly, there was a significant decrease in organotin compounds in mackerel liver along the German Bight transect. Similar results were seen for herring liver, but sample sizes were too small for clear conclusions. PAHs in zooplankton samples from the Statfjord area were highest close to the platform, but otherwise levels varied with distance.

SPMDs accumulated organic contaminants in both areas and the highest concentrations of PAHs were found at the stations closest to the expected source (S1 and G1). There was however no clear gradient for total organic contaminants in any of the two areas. Alkylated PAHs dominated in SPMDs near Statfjord, but were not found in the German Bight. Alkylphenols were detected in SPMDs deployed in the German Bight, but not in SPMDs from Statfjord. The extraction method was not optimised for alkylphenols, so those results must be interpreted with caution.

Biological effects – field collected organisms

Studies on field collected organisms ranged from bacteria to fish. Bacterial diversity was assessed at some sites, but any site-dependent differences have not yet been quantified. Although there was apparent reduced grazing capacity by microzooplankton at one location in the German Bight, there was no obvious link to exposure. No effects were observed on primary production by algae cultured on board the vessel. Although there is an extensive database on fish embryo aberrations from the German Bight (Dethlefsen et al. 1996; von Westernhagen et al. 1980; von Westernhagen et al. 1988; von Westernhagen et al. 1989), there is only limited background from the northern North Sea. There were no obvious increases in fish larvae aberrations along any of the transects. There were no differences in PAH metabolite levels in bile from either saithe (Statfjord transect only), herring (mainly German Bight transect) or mackerel collected at different sites. Although there was no difference between stations in EROD (cytochrome P4501A activity), concentrations of cytochrome P4501A in herring liver differed between sites in the German Bight. DNA adducts were found in fish larvae (sandeel) in the German Bight, but there were no

differences between locations. One interesting finding was the differences between sites in liver tissue integrity (histopathology). Significant effects on herring and saithe liver tissue integrity were found in the inner German Bight (G1) and close to the Statfjord B platform (S1). There were no significant differences in plasma vitellogenin levels in either species in either area.

Biological effects – caged organisms

Various analyses were done on caged blue mussels and Atlantic cod. Clear differences along the transects were seen for histopathological changes in the blue mussel hepatopancreas (basophilic cell volume) and there was also reduced lysosomal stability at stations in the inner German Bight and close to the oil rig. In the German Bight, acetyl cholinesterase (AChE) was inhibited in both blue mussels and cod. Further, benzo(a)pyrene hydroxylase (BaPH) was induced in blue mussels kept in the inner German Bight. There were no significant changes in scope for growth of mussels kept at the different sites (in either area). The metal-responsive protein metallothionein did also not differ between sites. Tissues from blue mussel were subjected to 2D electrophoresis. Protein patterns differed between sites close to pollution sources and the reference sites. It is still too early to conclude on the utility of this method, but the relevant proteins should be identified.

There were obvious gradients in PAH metabolites in both areas. In the rig transect alkylated PAH metabolites comprised a substantial fraction not seen in bile from cod kept in the German Bight. In the German Bight, but not in the Statfjord transect, there was a gradient in EROD activity. Glutathione *S*-transferase (GST) activity increased at the sites closest to the oil rig, but there was no difference between sites in the German Bight (Figure 2).

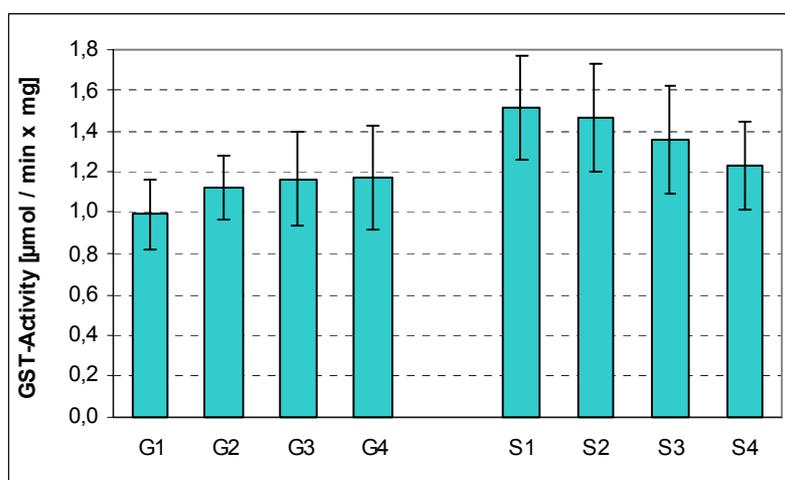


Figure 2. Glutathione *S*-transferase activity in cod liver from the German Bight (left) and the Statfjord transect (right). Results are means with standard error. Data from Dirk Danischewski (BFA-Fi).

There were low concentrations of DNA adducts in cod from both areas and no significant differences in vitellogenin levels (although there appeared to be a gradient). In the German Bight there were differences between sites in cellular energy allocation (CEA).

Biological effects – bioassays

The work on bioassays was hampered by contamination of extracts with triolein from the SPMDs, which had to be cleaned before analyses. Presumably due to the small amount of samples no response was seen following intraperitoneal injection of extracts into juvenile

salmon. Most of the applied methods only saw responses in the positive control, the produced water extract. That included all whole-organism tests, i.e. the ELS zebrafish test, mussel embryo, *Acartia*, *Tisbe* and *Skeletonema*. In addition, responses were only seen in the produced water extract for comet assay (cell line) and microtox/mutatox. Some methods did however indicate effects in other extracts (closer to pollution sources), e.g. fish primary hepatocyte culture (EROD, vitellogenin) and DR-Calux. A preliminary TIE (toxicity identification and evaluation) identified estrogenicity due to C3-C9 phenols in the produced water extract. Although conditions were optimal for the collection of surface microlayer (SSML), effects were only seen in a few samples. Phototoxicity (exposure with UV) was evident in a single sample.

Recommendations

The selection of methods obviously depends on the objective, whether it is local/regional monitoring, general effects or identification of specific substances and, finally, whether ecosystem relevance is important. Monitoring and research programmes should include chemistry, biology and modelling components (they are not mutually exclusive). Furthermore, *in situ* extraction combined with bioassays, caging and field sampling are complementary approaches. *In situ* extracts are useful to identify mechanisms of toxicity and specific substances, caging provides a direct link to local exposure, whereas field sampling is ecologically relevant.

How should useful methods be identified? There are some requirements that should be met: the method need to have an identifiable threshold or dose-response level(s) for effect; the method must have sufficient (statistical) power to detect predefined difference/change in relation to exposure; the method should be applicable to different species; a selected method must be executable in practice.

It is essential that a quality assurance system exists for methods that are selected for monitoring. At the present, the programme BEQUALM is being developed to include relevant methods.

Which species or systems should be used? There is no "universal" species found in all relevant geographical regions. It must probably be accepted that different species are used in different areas, but work should be initiated to "calibrate" the species in relation to each other.

Promising research directions

Although a wide range of methods was applied within BECPELAG, there are methods and systems that need to be further assessed. The microbial loop (bacteria, microzooplankton) constitute an important component of marine ecosystems and there is little knowledge of effects from contaminants. To decrease detection limits and facilitate work with small organisms, mRNA should be developed for e.g. zooplankton and fish embryos. In relation to caging, work should be initiated to cage other organisms. Briefly assessed within BECPELAG, proteomics offer a large potential to identify sensitive components in various species.

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