

Comparing biomarker responses with risk estimates used for decision analysis

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Abstract

This paper deals with the coupling between biomarker responses and modelling environmental risks for environmental impacts in the field. The use of filter feeding scallops and mussels were used in an exposure experiment where discharges of drill cuttings and mud were taking place. The drilling wastes revealed biological impacts as physical disturbance affecting energy storing and reproduction (reduced gonad weight), algae filtration and metabolism. The changed conditions for the exposed transplanted animals increased the oxidative stress and revealed significant DNA damages in the assumed highest drilling waste sited mussels. The biomarker responses were compared to numerical simulation results where the discharges were mimicked with the numerical model. An environmental risk calculation was performed for the location of the largest exposures/responses. The biomarker responses were then compared with the estimates of the environmental risks for damage on the biota simulated with the numerical model. The results indicate that the biomarker responses appear to be sensitive to the concentrations calculated for the actual locations of the biota, even for low levels of environmental risks simulated for the same site. This result indicates that the biomarkers may serve as an "early warning" tool for revealing potential environmental damage.

Keywords: environment, discharge, offshore, drill cuttings and mud, risk analysis, biomarkers, numerical modelling.

1 General

The ERMS project (ERMS = *Environmental Risk Management System*) is aimed at developing models for prediction of impacts from regular releases to sea



caused by the offshore industry. The types of discharges considered are discharges during production (basically produced water releases) and discharges during drilling (basically discharges of drill cuttings and mud). The main purpose of the ERMS project is to develop risk based tools for predicting potential environmental impacts caused by the regular releases to sea generated by the offshore industry.

In order to validate risk estimates calculated with the ERMS model, a field trial was therefore conducted, deploying cages with sea scallops and blue mussels in the vicinity of an offshore drilling platform. The biomarker data are then to be compared with risk estimates obtained with the numerical model developed. For this purpose, a planned drilling operation on the Sleipner field in the North Sea was selected. The drilling program included discharges from 4 production wells (one template) on the Sleipner Vest Alfa Nord condensate field (the "SVAN" field for short, position about N 58°30' E 1°43'). The water depth is 110 m.

At the same time, RF-Akvamiljø has been granted by the Norwegian Research Council (NFR) to carry out a project termed "Validation" over the NFR PROOF Programme. This project is aimed at validation of methods for carrying risk analysis offshore. Because the ERMS project is aimed at developing numerical models for carrying out risk analysis for discharges to sea offshore, it was decided that RF-Akvamiljø should join the project by deploying cages with sea scallops and blue mussels close to the discharge site. Then the methods validated by RF-Akvamiljø could be tested on the real field case, by comparing risks deduced from the responses on the biota with the risks calculated by the numerical models developed as a part of the ERMS project.

Results presented in this paper are an extract from a part project report from these two projects (presently under development).

2 Site selection and ambient environmental data

The field data were collected at the SVAN field in the North Sea (Figure 1).

Ocean current measurements and ambient stratification were measured during the field trial. These data were used as input for the numerical simulation of the discharges. Figure 2 shows the location of the actual deployment of the instruments.

Vertical profiles of temperature and salinity were recorded for the determination of the ambient water stratification. The measurements showed a rather massive surface layer with temperatures at about 14–15°C down to 30–40 m depth. Below 40 m depth the temperature was generally within the interval 8–11°C. Salinity variations in the vertical were recorded to be small, within 34.6–34.9 ppt. Thus, it is the temperature variations that will cause a density change in the ambient water masses. These will be most pronounced close to 40 m depth.



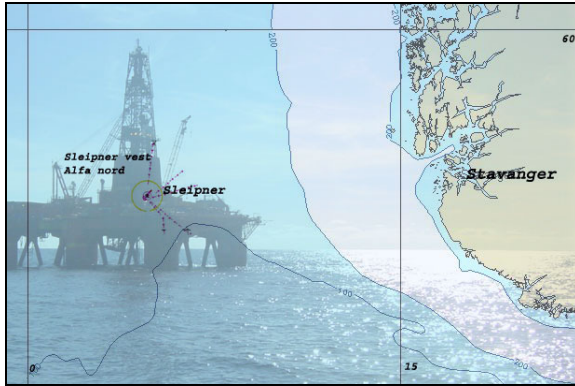


Figure 1: Location site of the drilling rig Transocean Searcher at Sleipner Vest Alfa Nord (SVAN).

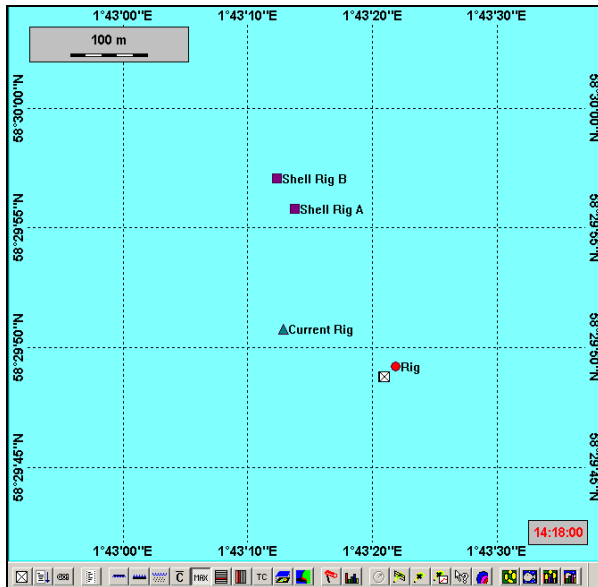


Figure 2: The location of the Shell cage stations A and B and the ocean current rig location. The discharge location is denoted with a square with a cross inside. The cages with the sea scallops and the mussels were deployed about 200 m from the discharge point.

3 Amounts of discharge

The discharge took place from the drilling rig while drilling the 17½" and 12¼" drilling sections. The debris from the drilling hole was separated at the shaker, separating out the cuttings (to be discharged) from the mud (to be re-used). The cuttings (with some mud attached to it) were led through a pipe with an outlet



opening located at about 5 m depth below the sea surface. In addition to the cuttings, the discharge contained barite bariumsulphate (BaSO_4) particles (also called barite) and chemicals. The barite is used as a weighting material during the drilling process. The barite particles were assumed to represent the largest source for the environmental stress on the (filtering) sea scallops and mussels in the cages deployed.

The cages were deployed before discharge start at 9 September 2003 and retrieved at 12 October 2003 (33 days) During this time period, about 350 tonnes of barite were discharged at intervals during the drilling process. The barite has a fine particle structure of irregular shape, with particle diameters of order 0.1–100 μm . The particles are assumed to impact on the filtering organisms due to the irregular shape of the particles.

About 1500 tons of particle cuttings material was discharged as well, but these particles have generally larger diameters and are expected to sink down on the sea floor rather than impact on the filtering organisms in the cages.

4 About the ERMS numerical model

As a part of the ERMS project, a numerical model was developed to simulate fate and behavior of the drilling discharges to the sea. The discharge is assumed to spread in the ambient water where the discharge depth (and location) acts as the source point. The discharge is assumed to form a “near field” underwater plume that spreads out in the recipient. The plume sinks down due to the content of the barite and cuttings particles which are heavier than the ambient water. The plume will stop sinking when the density of the plume equals the density of the ambient water (depth of “trapping” of the underwater plume). When the depth of trapping is reached, the discharge will separate into two parts. One part spreads out horizontally in the water column. This part will contain fine particles (with low sinking velocities) and dissolved chemicals. The other part will sink down on the sea floor. This part consists of coarser particles (with larger sinking velocities) and also chemicals/metals that are attached to the particles. It is the part of the discharge that spreads in the water column that forms the basis for the environmental impact on the cages deployed in the water column The part of the discharge that sinks down on the sea floor forms that basis for the impact on the bottom sediment (not considered here).

Further details of the ERMS model developed as a part of the ERMS project are given in a separate project report and will not be repeated here (SINTEF [1]).

As an example calculation, the concentration field for the particle concentrations looks like as shown in Figure 3. The concentration field contains a plume area with relatively large concentration (up to some ppm level), combined with a larger area where the concentrations are considerably smaller (typically order 5 – 50 ppb).

Due to the presence of the stratification in the water masses, the maximum concentrations shown are not present in the surface layers. Due to the presence of the cuttings and the mud in the discharge, the discharge has a much larger density than the ambient water. The discharge plume will therefore sink down



until the density of the plume equals the density of the ambient water. This happens in the depth close to 40 m below the sea surface. It is therefore expected that it will be the cage(s) deployed at about 40 m depth that will experience the largest particle stresses.

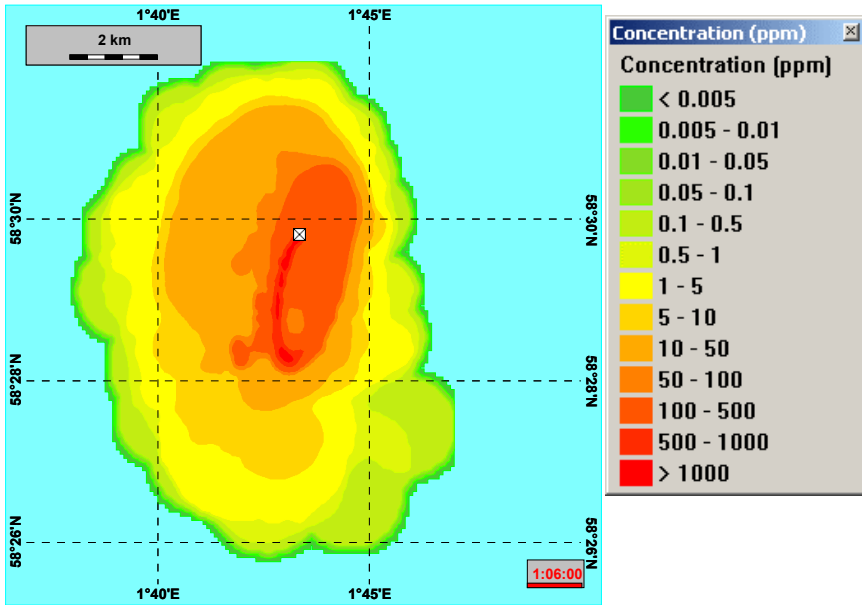


Figure 3: Concentration field for particle concentrations (sum of cuttings and barite) 30 hours after start of release from the drilling platform (9 September 2003).

Figure 4 shows a calculation of a maximum particle concentration level at the location where shell rig A was deployed. This rig contained one cage with filtering organisms at the 40 m level where the discharge is expected to have the largest impact. Due to tidal motion of the currents, the concentration are shown to be highly time variable at the cage location during the drilling period.

5 Risk estimates based on the ERMS numerical model

One of the purposes of the field trial was to compare environmental impact at the cage locations with biomarker responses determined for the sea scallops and the blue mussels. Particular attention was drawn to the presence of the barite, which may have a particle effect on filtering organisms (Cranford et al. [2]).

Since no continuous measurements of the barite concentrations were carried out at the different cage locations that were retrieved, the concentrations were deduced from the numerical simulations of the barite (or particle) concentrations. The numerical model developed was therefore run for the whole period where the shell rigs were deployed (about 33 days).

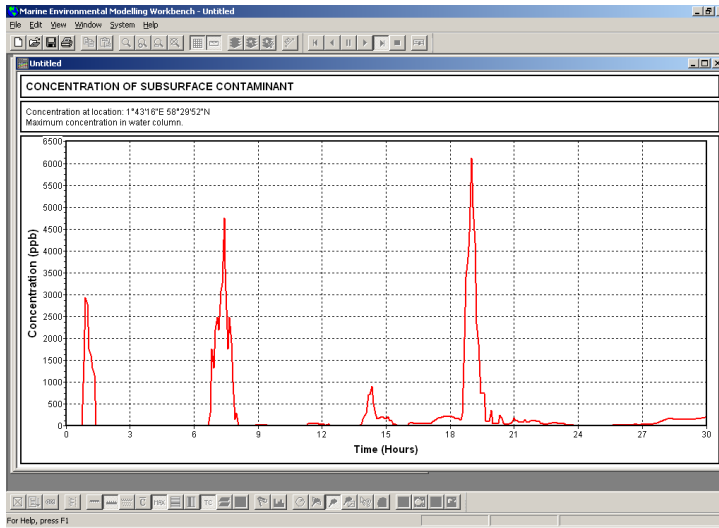


Figure 4: Particle concentration calculated for the shell cage at 40 m depth at location Shell cage A for the time period 9 – 10 September 2003 at the SVAN field.

The present risk method developed during the ERMS project is based on a PEC/PNEC approach, where the PEC is the "Predicted Environmental Concentration" and the PNEC is the "Predicted No Effect Concentration".

The principle used is in accordance with the recommendation from the EU "Technical Guidance Document" (TGD [3]). The predicted concentration level (the PEC, in this case PEC is produced by modeling the concentration levels) is to be compared with a fixed concentration level (PNEC) below which no or acceptable potential impact on the biota is encountered. The PNEC level is associated with a level of 5% probability for damage or impact on biota in the recipient.

The environmental impacts from the particles (BaSO_4) chemicals are treated in the same way as for the chemicals impacting on the biota in a recipient. The PEC/PNEC ratio for the chemical or compound in question is then to determine the probability of impact on the recipient in terms of probability of risk for damage. The method used is presently according to a method developed by Karman et al., [4] (and also published in Karman and Reerink [5]). When $\text{PEC/PNEC} = 1$, this corresponds to a level of probability of damage equal to 5%. When $\text{PEC/PNEC} < 1$, the probability of damage (risk) is lower than 5%. When $\text{PEC/PNEC} > 1$, the risk is correspondingly higher than 5%. Figure 5 shows an example of the relation between the PEC/PNEC ratio and the probability of damage (risk).

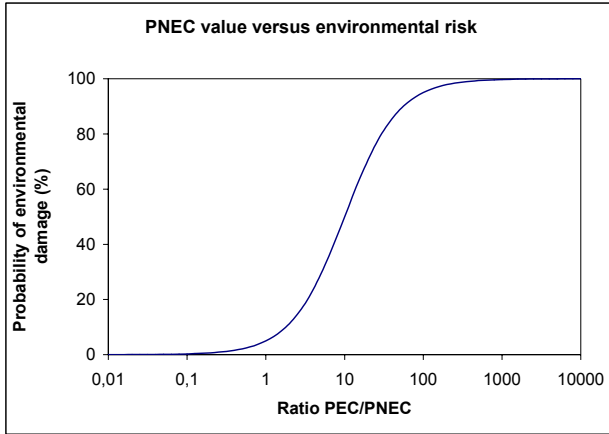


Figure 5: The relation between the PEC/PNEC level and the risk level (in %) for damage on biota. Note that at the level $PEC/PNEC = 1$, the probability of damage is 5%. Based on Karman et al. [4].

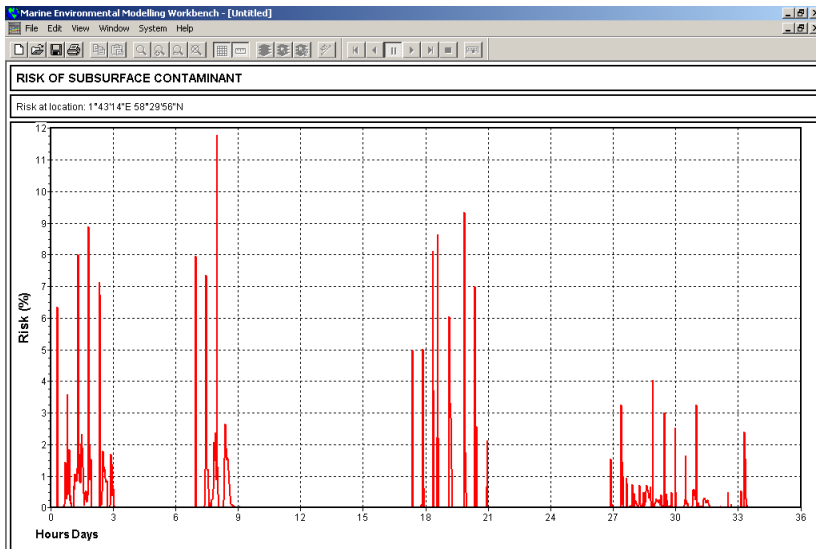


Figure 6: The time series risks due to particle concentrations (essentially barite) calculated for the cage at 40 m depth at shell rig location A. Vertical axis represents the probability of risk of damage caused by particle barite deduced from “Species Sensitive Distributions”. Horizontal axis represents the time of exposure (days) for the cage at 40 m depth for the 33–34 days period of deployment.

The risk calculation results for the 40 m depth location at the Shell rig A is shown in Figure 6. The risks are generally relatively small, of order within 10% probability of damage or impact on the biota. It should be stressed that the actual risk level is very sensitive to the PNEC level determined, mainly based on laboratory testing on impact caused by barite particles brought into suspension in the lab.

The long time discharge of drill cuttings and mud into the water column represent a possible impact towards pelagic and sediment linked organisms. Mussels are often used as indicator organisms that are particular vulnerable in the connection to pollutions in the sea since they have restricted or no opportunities to move away when the conditions become unfavourable. Both particles and dissolved chemicals will enter the shell, and dependent of the property, the material will be accumulated, metabolized or rejected. Scallops and mussels reject particles based on the ability to discriminate against size and inorganic material. The productions of faeces are efficient when the content of organic material is higher than 50%, reducing its ability significant when the organic part is less than 25%. The productions of faeces are also more efficient for larger particles (greater than 10 μm) compare to small particles (Hovgaard [7]; Hardy [8]).

A variety of different biomarker responses for the sea scallops and the mussels were tested out, where some gave responses on the impact, and some did not show a clear signal on impact. Just one example is given here, namely the comet assay on DNA damage. To our knowledge, no genotoxicity study on marine organisms exposed during drilling operations has been performed. However, evidence of long- term adverse effects, such as cancer, due to relatively high concentrations of heavy metals in marine animals has been shown in field and experimental studies (Bolognesi, Rabboni et al. [9]). Genotoxic effect may be involved in the mechanism of metal carcinogenicity. The comet assay has been used to detect DNA damage caused by metal exposure of fish (Risso-de Faverney et al. [10]) and mussels (Bolognesi et al. [9], Black et al. [11]).

The comet results measured at reference mussels (from 20 meters depth) and mussels exposed at 40 meters (Figure 7), indicating that exposed mussels caused statistically significant ($p < 0.05$) increase in % DNA in comet tail compared to the reference group when the comparison is based on all cells measured. In both reference and exposed group the DNA in comet tail exceeds the 10% natural boundary by respectively 49% and 61%. The increase of DNA damages in mussel's haemocytes caged at 40 m correlates well with the turbidity measurements showing that the particle exposure was most elevated at this depth.

Significant DNA damages have been found at less barite concentration (0,5 mg/l, for 4 weeks) (Bechmann and Taban [12]) and only DNA damages from greater depths differences than 20 m have been reported. The results from the present study show that DNA damages could be found in haemocytes (blood cells) by exposing *Mytilus edulis* (mussels) to barite particles.



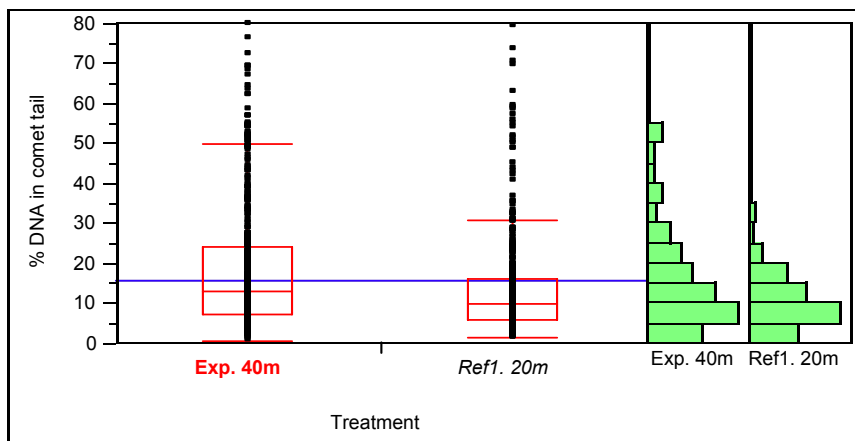


Figure 7: Comparison based on all cells measured from mussels exposed at 40 m depth with all cells from reference mussels (20 m). Grand mean is given as a horizontal line as well as boxes with median values.

The results from the biomarker responses showed that drill cuttings and mud cause biological impact as demonstrated with the applied methods. The practical approach of using organisms deployed in cages seems useful for screening of drill cutting and mud discharges. Statistical significant increase of DNA in the comet tail was found for exposed 40 m mussels, but not for the 20 m exposed group, compare to reference group at 20 m. The conditions in the exposed 40 m zone during the one-month stay, involves mussels in exposure conditions that produced genetically damages in the DNA.

Laboratory experiments performed under different barite exposure regime (Bechmann and Taban [12]) strengthen the result that barite have a negative affect on the ability for filterfeeders as mussels and scallops to sustain a normal feeding regime. The laboratory exposure revealed a reduction for scallops at exposed to 0,5 mg/l barite and 20 mg/l barite for mussels (Bechmann and Taban 2006). During the SVAN field exposure the barite concentrations at 40 m reach pulses up to 5 – 10 mg/l barite with an average exposure level varying from 0,09-0,7 mg/l barite. Since most of the exposure is below 2 mg/l and earlier lab experiments revealed only a effect for mussels at 20 mg/l a short term exposure pulse could be the main responsible for the reduced filtration rate. If so the effect from these few exposure hours are essential and sustain a reduced filtration rate even after 14 days. The disposal of barite at 20 m is much lower than 40 m and has no effect on the filtration rate for mussels. An average exposure under 0,01 mg/l with barite pulses up to 0,16 mg/l gave a significant reduction in the filtration rate for scallops. This could mean that scallops are even sensitive to barite concentrations as low as 0,01 mg/l or that the discharges in pulses affects and sustain a reduced filtration rate after a two-hour exposure time.

6 Relation between risks modeled and the biomarker responses

The challenge is to be able to link the biomarkers to risk assessment in such a way that it builds a bridge between prognoses made in risk assessment and subsequent diagnosis in field monitoring. In probabilistic risk assessment the risk is calculated by combining predicted exposure concentrations with a Species Sensitivity Distributions (SSDs) which hold information about probability of adverse effects. One example is shown in Figures 5 and 6. In the so-called "Validation" project under the PROOF programme a validation link between biomarker signals and risk for produced water discharges has been established (Smit et al. *in prep.*). It seems possible to establish similar validation links related to drilling discharges. This can be judged by the present biomarker based exposures in cages near drilling sites and results from laboratory studies of biomarker signals in response to simulated drilling discharges (Bechmann et al. *in prep.*).

To give an introduction to how this can be accomplished for drilling discharges, the main features of the validation link and the principles followed is presented in the following, see Figure 8.

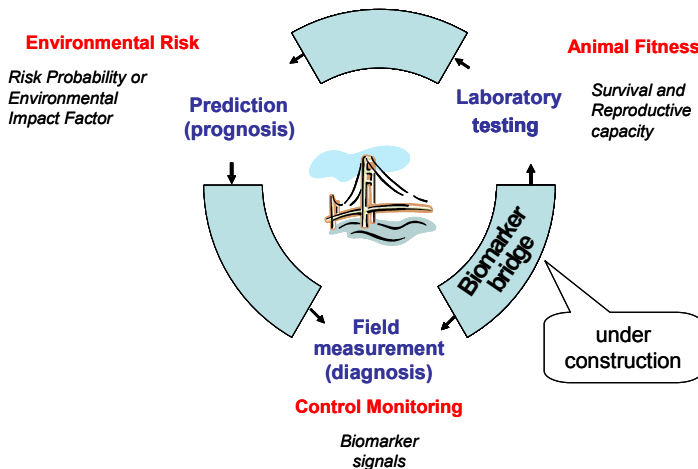


Figure 8: Illustration of a unified concept of Environmental Risk Assessment with Control Monitoring. The Control Monitoring is based on comparison between Field measured Biomarker signals and Predicted Impact in the Risk Assessments. The Predicted Risk Impact is based on Animal Fitness information obtained in Laboratory Tests. The concept is tied together by relationships between Biomarker signals and Survival and Reproductive capacity. This is often referred to as the "Biomarker Bridge".

The approach found most suitable in building the “biomarker bridge” was to establish Biomarker Sensitivity Distributions (BSDs) analogous to the Species Sensitivity Distributions (SSDs) that are used in the present risk calculation procedures. The BSDs may be grouped into different categories according to types of biological effects (genotoxicity, oxidative stress, endocrine disruption etc.). The curves between the Risk curves and the different BSDs represent the actual bridge between the risk and biomarker signals (see Figure 9).

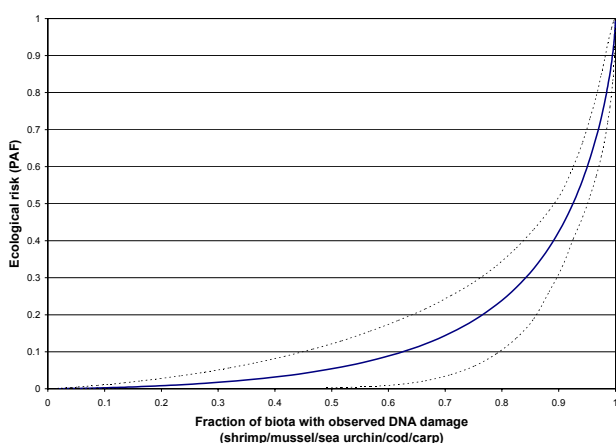


Figure 9: Line with 95% confidence limits showing the relationship between a Risk curve (SSD, vertical axis) and a Biomarker Sensitivity Distribution curve (BSD, horizontal axis), representing the bridge (validation link) between the environmental risk and biomarker signals.

The above represents the main features of the concept, which will be developed in further detail in the last phase of the “Validation” project. This project is expected to be completed by September 2006. This project is basically treated to discharges of produced water, but should in principle also be applied to drilling discharges. Preliminary results from the laboratory effect studies of drilling discharges (Bechmann et al. *in prep.*) and the biological measurements carried out in the present study indicate that there is correspondence between laboratory and field data. This is a prerequisite to be able to develop a unified concept of risk assessment and monitoring for drilling discharges.

7 Concluding remarks

The risk analysis presented above has been used extensively by the oil companies as a basis for reducing their impact on the environment due to discharges to the sea. This method is based on numerical model simulation results, producing the PECs. The PNEC values used in the simulations are based on laboratory trials to determine the threshold values for impact on certain



species. Also, risk functions (that is, SSDs) are based on results from laboratory trials. Thus, the impact reducing measures are therefore solely based on numerical simulation results combined with laboratory trials. No measurements in the field or on the actual site are carried out to be included as a part of the material available for decision making on the measures planned. The biomarker response experiments are however carried out in the field. Here the actual responses on biota are determined by placing relevant species in the vicinity of the discharge point. Thus, by combining the biomarker response results (through the BSD's) with the SSDs, a direct link is established between the decision making tool (the risk analysis) and the biomarker response signals.

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