

# Main challenges for the use of biomarkers towards a sustainable management for the marine environment protection

Challenges related to the linking of impact prognostics (predictions) and diagnostic monitoring tools. Challenges in balancing environmental cost contra benefit.

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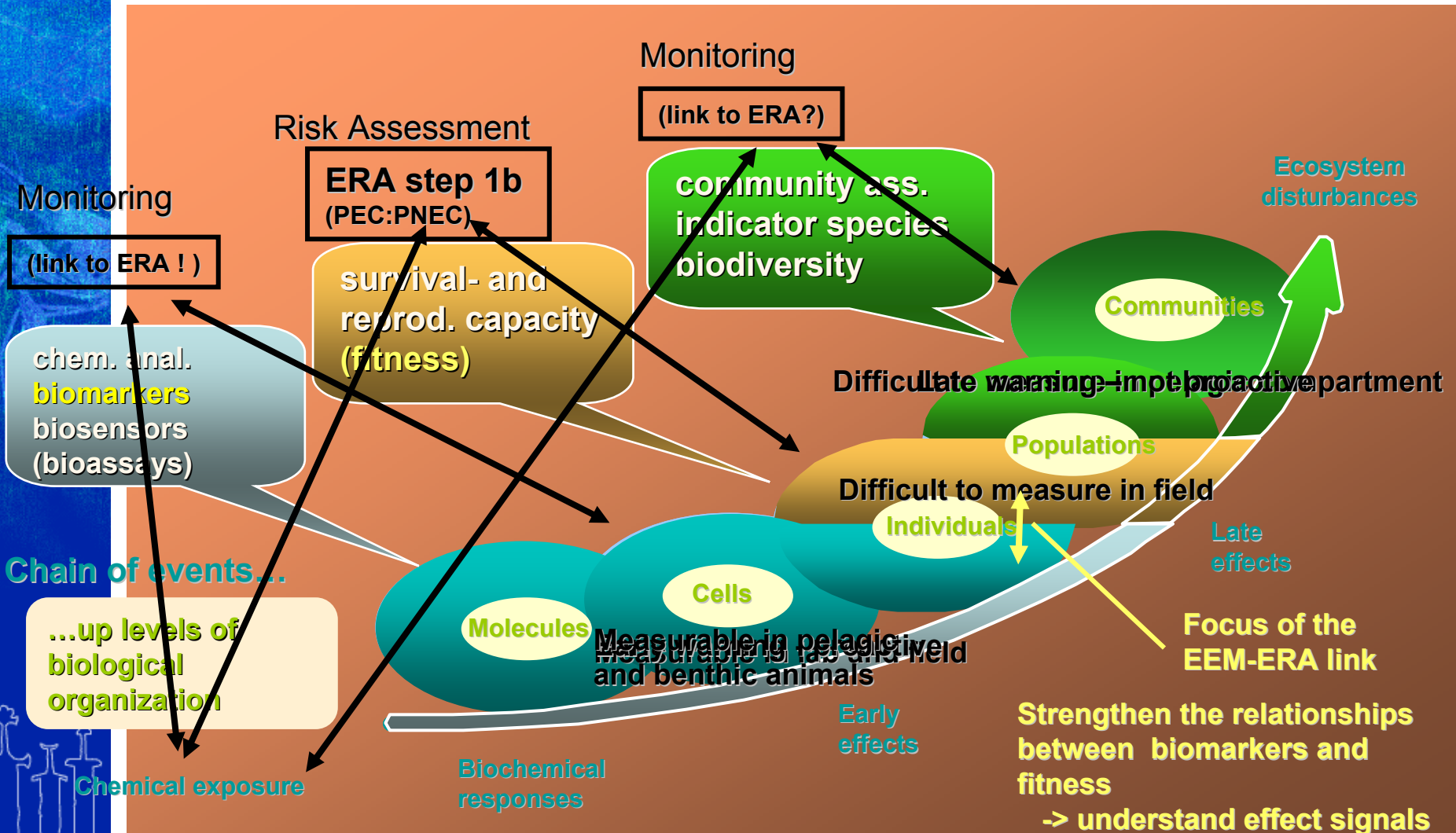
# Linking of impact prognostics (predictions) and diagnostic monitoring tools

- Integration of Biomarkers in ERA
  - Motivation
  - Approach
  - Solutions
  - Challenges



# Integration of Biomarkers in ERA

## Motivation and Approach



# Integration of Biomarkers in ERA

## EEM - ERA link

- Approach

- Build a 'Biomarker Bridge'

- Why?

- enables field monitoring of accepted biological effect levels determined by risk assessment

- How?

- strengthen the relationships between biomarkers and fitness (lab studies)

- Additional benefit!

- this will improve understanding of biomarker signals  
-> contributes to interpretation basis for biomarkers

- Alternative approach:

fully unite 'Test endpoint' to 'Assessment endpoint' (=Biomarkers)

- Realistic alternative?

- Ecological relevance of biomarker responses remains to be fully defined

- Definition of threshold levels and criteria to quantify ecological relevance must be established for biomarkers !



# Integration of Biomarkers in ERA

## Challenges

- The correlations between fitness and biomarkers are not necessarily based on causal relationships !
  - Only some of the links between biomarkers and fitness effects are well known and understood in a mechanistic way
  - Same dilemma as in human health research
- To overcome this problem
  - There is a need for sufficient amount of data to be able establish **statistical** relationships or '**weight of evidence**' to determine the relevance of biomarkers in relation to fitness and ecological effects
  - An advantage before human health research is that we can be **more experimental** !

# Integration of Biomarkers in ERA

- Solutions

- Building the 'Biomarker bridge'

- Two examples:

- Vertebrate

- » Fish (sheepshead minnow - *Cyprinodon variegatus*)

- Invertebrate

- » Crustacea (Northern shrimp - *Pandalus borealis*)

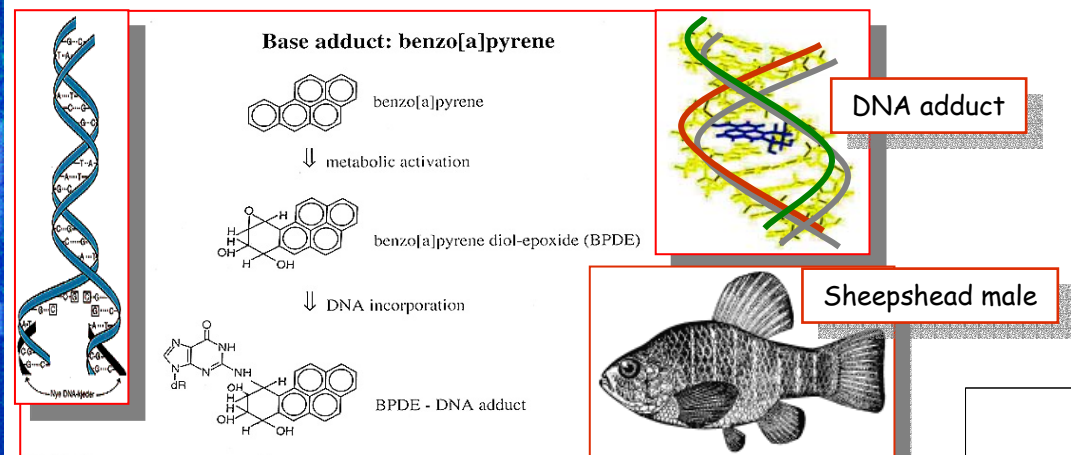




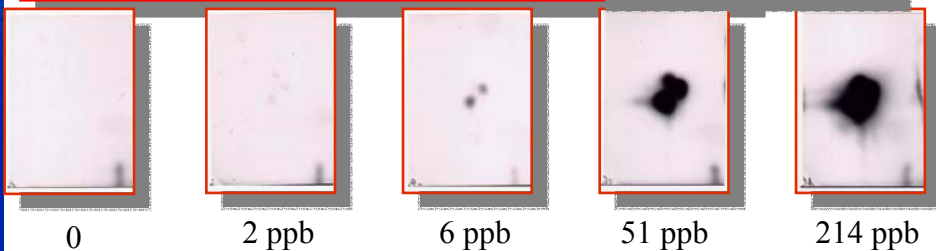
# Building of the 'Biomarker bridge'

## Correlations fitness and biomarkers (example):

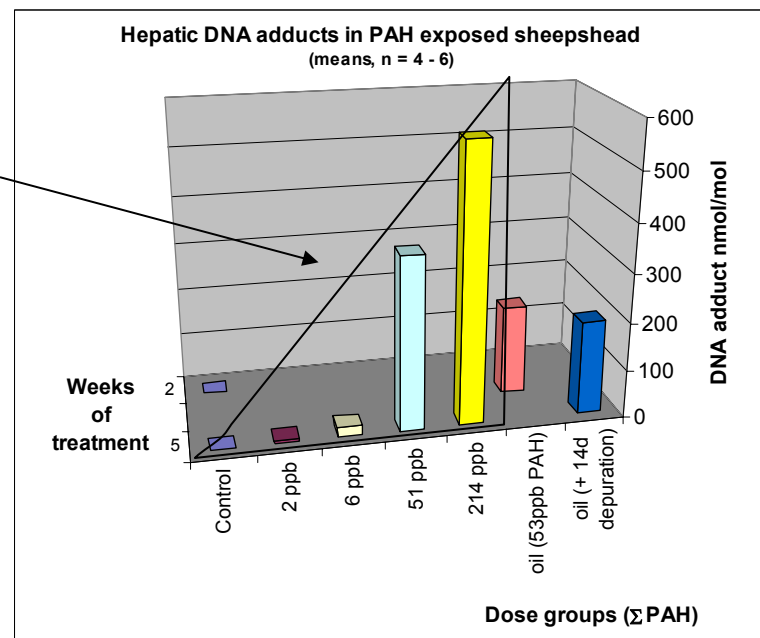
### Lab. Effect study exposure of fish



**Biomarker:**  
Benzo[a]pyrene  
DNA adduct



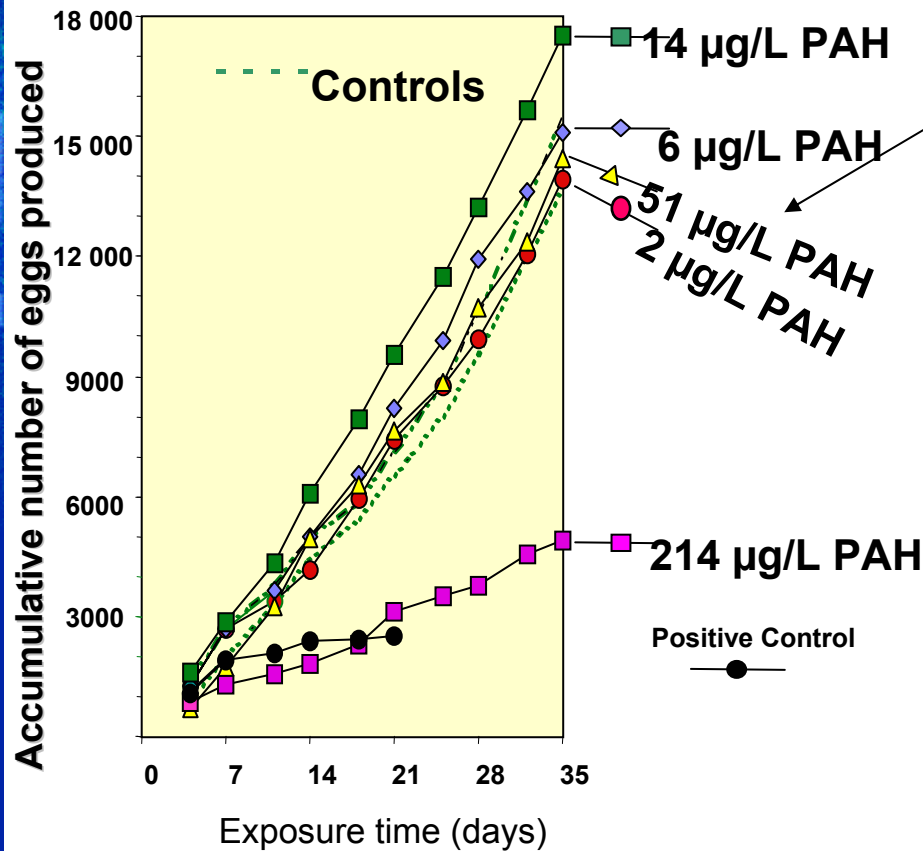
Increasing PAH dose (ppb in water) /  
Increasing biomarker effect (adducts in liver)



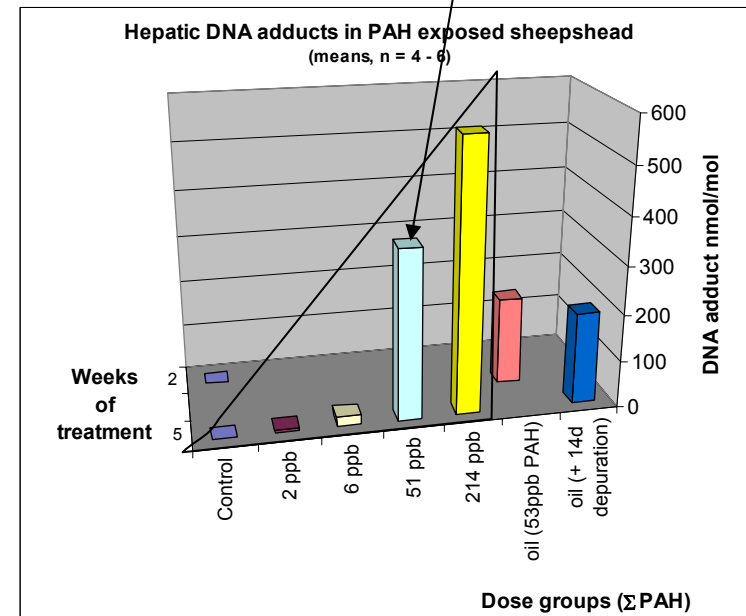
(data & ill. J. Beyer)

# Building of the 'Biomarker bridge'

Fitness: reproductive output (as fecundity)  
coherent with to biomarker values



- Egg production 'NOEC' (51 µg/L PAH)
- corresponds to
- a DNA adduct level of approximately 300 nmol/mol



(data & ill. R.K. Bechmann & J. Beyer)



# Building of the 'Biomarker bridge'

## PROOF "Validation" - experiments

- ERA effect validation & Biomarker responses

	Effect validation	Biomarker responses
Invertebrates	<b>Oil dispersions - sensitive life stage test</b>	<b>Oil dispersions – sensitive life stage test</b>
	(PROOF – RF-AM) <b>Drilling Discharges - field experiment</b>	<b>Drilling Discharges - lab experiment</b> <b>Drilling Discharges - field experiment</b>
Fish	(PROOF-IMR)	<b>Alkylated Phenols - lab. Experiment</b>
	<b>Oil disp. / sim.PW - lab. experiment</b>	<b>Oil disp. / sim.PW - lab. experiment</b>
	<b>Produced Water - field experiment</b>	<b>Produced Water - field experiment</b>

Practical connections to:

ERMS

BioSea JIP

PROOF

WCM - OLF



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# Building of the 'Biomarker bridge' PROOF "Validation" - experiments

- ERA effect validation & Biomarker responses

	Effect validation	Biomarker responses
Invertebrates	<b>Oil dispersions - sensitive life stage test</b>	<b>Oil dispersions – sensitive life stage test</b>
	(PROOF ...)	<b>Drilling Discharges - lab experiment</b>
Fish	<b>Crustacean &amp; mollusk (&amp; echinoderm): North Sea dispersed oil</b>	
	<b>survival offspring</b>	
	mollusks: exposed parents / exposed larvae; <b>crustaceans: exposed as embryos / as larvae</b>	

Practical connections to:

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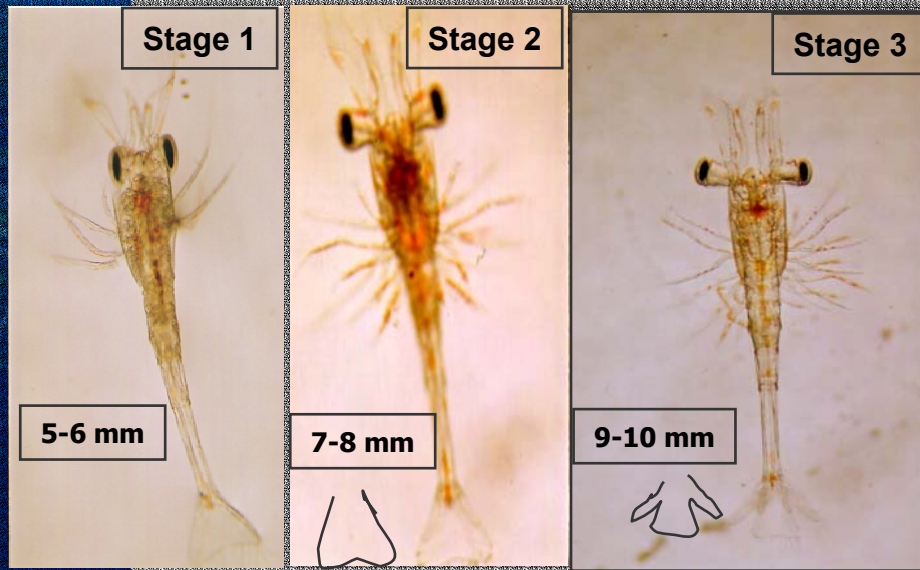


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# Building of the 'Biomarker bridge' Early life stage mortality - Northern shrimp

**Bodil K. Larsen**



- significant mortality when exposed as embryo & larvae
- dose dependent mortality when exposed as embryo
- significantly higher mortality in 0.09 ppm
- embryo exposure most severe

# Building of the 'Biomarker bridge' PROOF "Validation" - experiments

- ERA effect validation & Biomarker responses

	Effect validation	Biomarker responses
Invertebrates	Oil dispersions - sensitive life stage test	Oil dispersions - sensitive life stage test
	(PROOF - RF-AM)	Drilling Discharges - lab experiment
	Drilling Discharges - field	Drilling
Fish		<b>Crustacean &amp; Mollusks</b> <b>N.Sea oil exposure</b> <hr/> <b>Chemical burden &amp; Biomarkers</b> (1 month exposure) <hr/> <b>GST, Catalase, TOSC</b> Larvae: Comet (M), Alk.Unw. (C)

Practical connections to:

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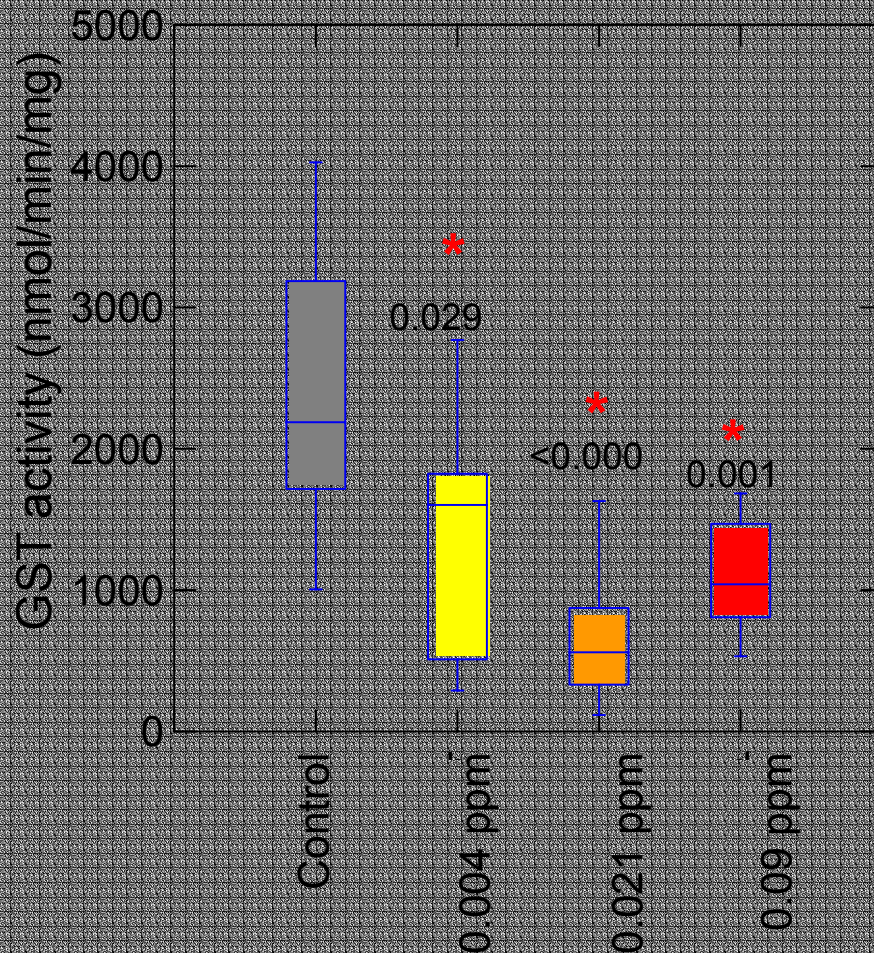
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# Building of the 'Biomarker bridge'

## Example of Biomarker in shrimp: Glutathione S-Transferase (GST)

**Bodil K. Larsen**



- other biomarkers measured for other projects in the same exposure
- joint evaluation of a larger biomarker set
- ->



# Building of the 'Biomarker bridge

## Example of conclusion based on joint evaluation

*Bodil K. Larsen*

### shrimp biomarkers :

- **GST:** Responds to oil exposure, although response pattern is probably not linear, rather U-shaped.
- **Lysosomal stability:** Works very well.
- **Alkaline unwinding:** Works well, but the sensitivity is not clear.
- **Catalase:** Large variability, bell-shaped response.
- **TOSC:** Large variability, small non-significant changes





# Integration of Biomarkers in ERA

## Use of the results...

- Compare (validate!) the PNEC values currently used in ERA by comparison with the found NOEC levels
  - based on organism health and fitness parameters
- Biomarker values associated with these NOEC levels can be established as threshold level signals
  - "Biomarker NOECs"
- Then these biomarkers can be used in Environmental Effect Monitoring (EEM) after field validation (QA, etc.→)
  - linked to Environmental Risk Assessment (ERA)!



# Other Challenges

## New frontiers of oil & gas industry...

- Research...

- adapt methods and tools to the new frontiers
  - deep-sea and Arctic
  - other geographical areas
- adapt methods
  - from North Sea & laboratory conditions
- develop tools (ERA, EEM)
  - based on the existing ones

another example:  
'tropicalisation of ERMS'

An example:  
Blood diagnostics



...with biomarkers in deep-sea sea stars  
(Girassol - Total Cold Water project)

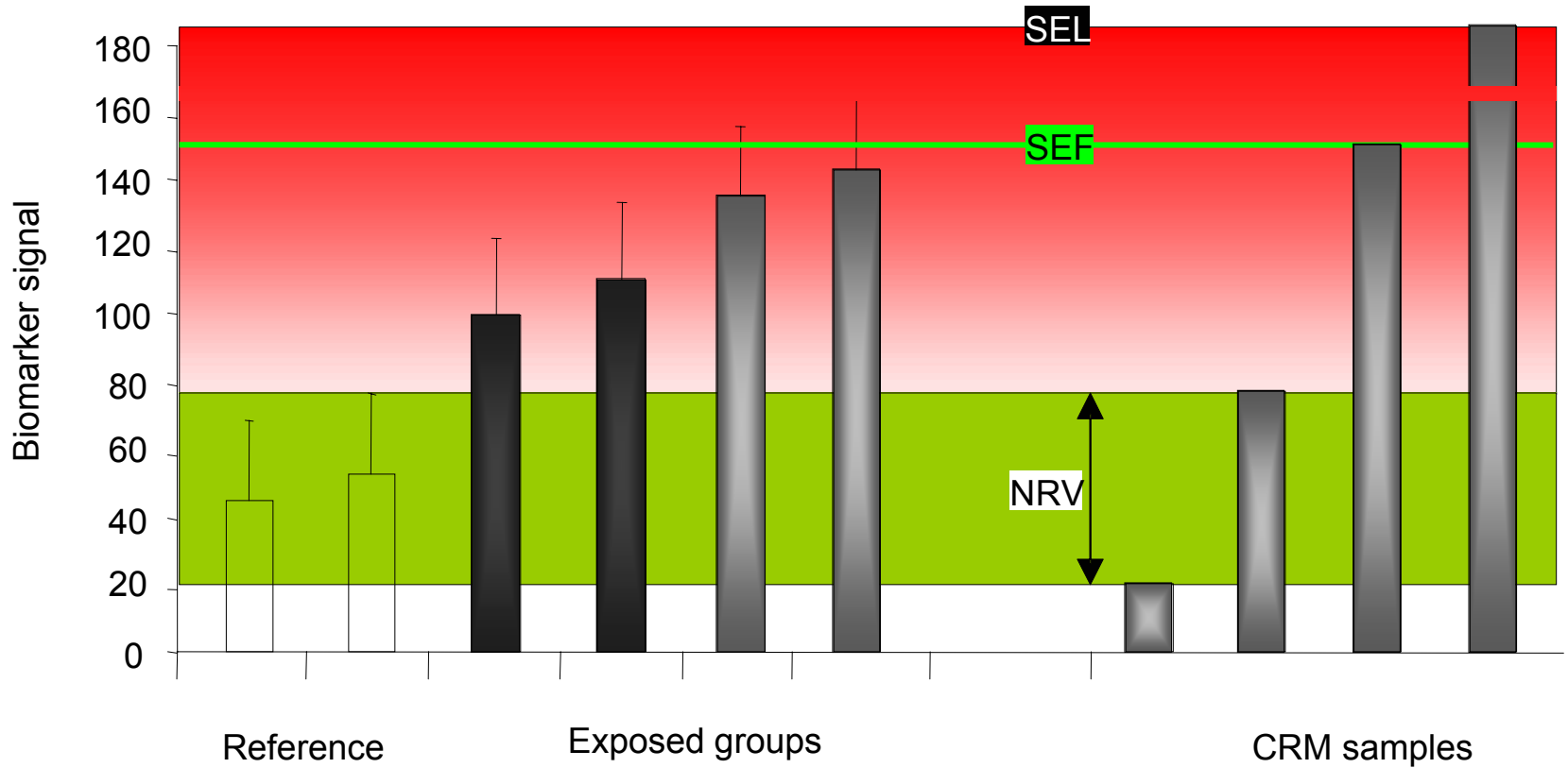


# Interpretation – different aspects

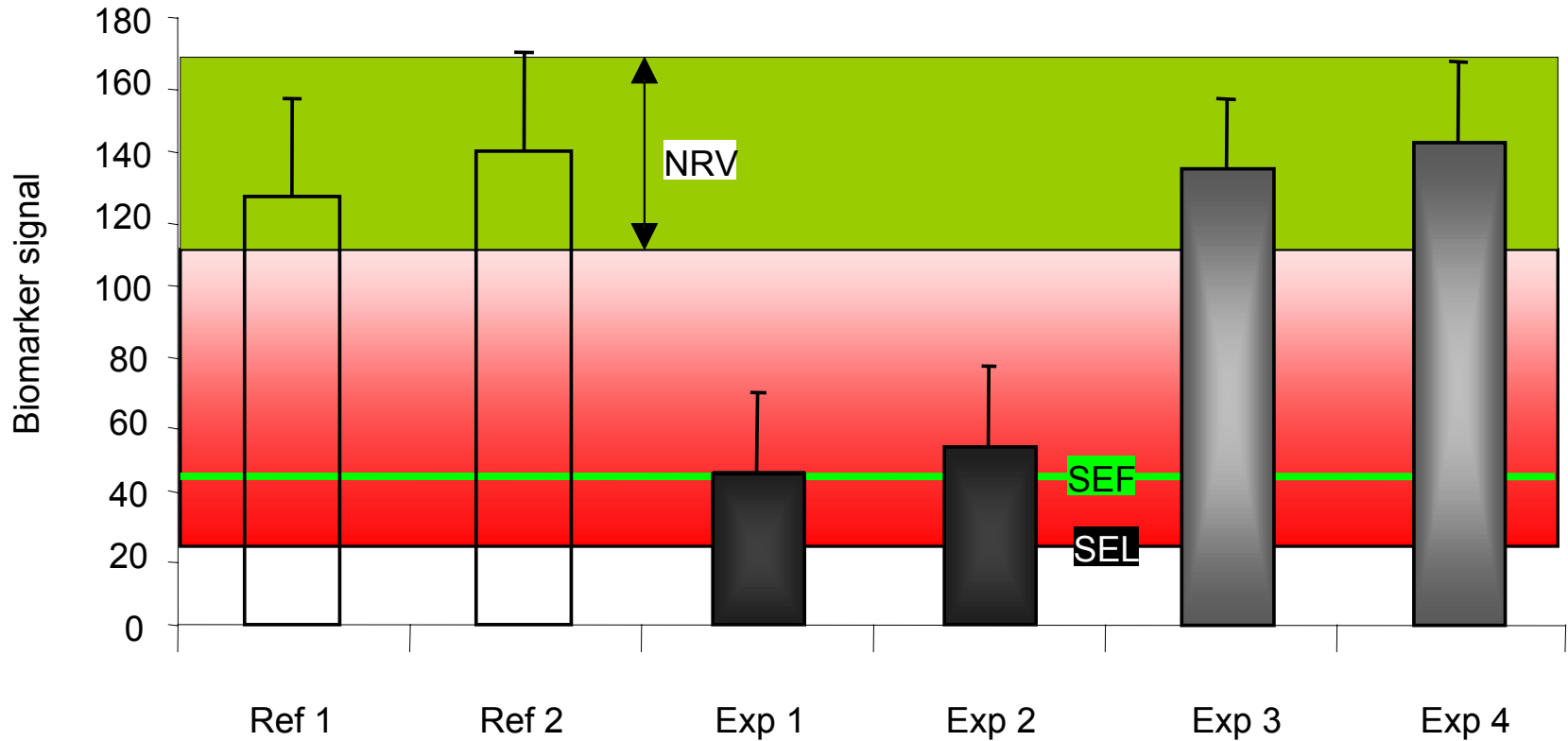
- Standardization
- Analytical variance
- Other QA aspects
- Spanning the scales
- Determination of threshold levels
- Combination of information



# Biomarker interpretation basis



# Biomarker interpretation basis



# Optimizing the biomarker approach

- **Stepwise EEM !**
  - **Environmentally relevant and cost efficient**
    - **1.step Screening**
      - Simple techniques
      - On-board analyses
    - **2.step Effect cause and severity**
      - » (dependant on results of 1.step)
      - Follow-up investigations (e.g. the North Sea DNA adduct case)
        - » Application of additional adequate biomarkers
        - » Chemical analysis
        - » Population or community investigations
  - **Combination with classical methods**





# Decision support – Balancing environmental cost / benefit

- Poorly developed
  - In Norway:
    - The zero harmful effects discharge sets a regulatory / politically based environmental goal
      - Which strongly influences the decisions
  - In Oil contingency planning :
    - Net Environmental Benefit Analysis (NEBA)
  - Environmental cost / benefit (= 'Risk benefit') needs to be developed further and integrated with ERA and EEM in the total Risk Management process ->





# Summary of Main challenges

- For the further use of biomarkers towards a sustainable management for the marine environment protection following challenges must be met
  - Biomarkers and ERA must be integrated ('Biomarker bridge')
  - Definition of threshold levels and criteria to quantify ecological relevance must be established for biomarkers
  - Adapt the methods to the new frontiers of the oil & gas industry
  - QA: the biomarker protocols must be standardized and the modes of interpretation biomarker results must be harmonized
  - Optimize the biomarker approach to a stepwise EEM
  - Balance the environmental cost / benefit in the Risk management process