LINKING MOLECULAR EVENT AND APICAL EFFECT TO ASSESS OIL TOXICITY: ADVERSE OUTCOME PATHWAYS

GRACE consortium meeting Tallinn

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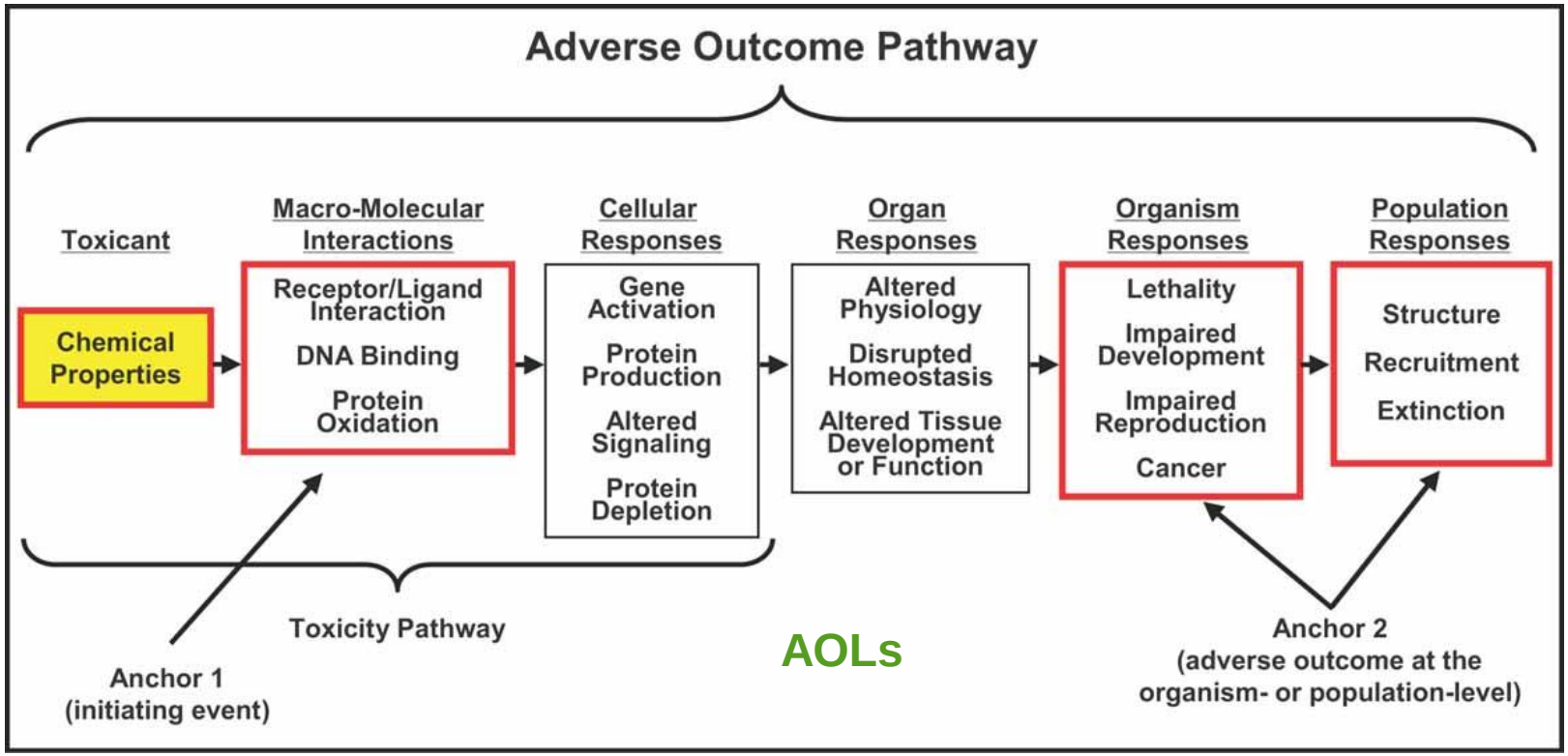
23.5.2019

AOP = ADVERSE OUTCOME PATHWAY

- Ankley et al. 2010: “Adverse outcome pathways: a conceptual framework to support ecotoxicology research and risk assessment” Environmental toxicology and chemistry 29(3), 730-741.

- “A framework within which data and knowledge collected at many levels of biological organization can be synthesized in a way that is useful to risk assessors”
- “AOPs represent a set of plausible connections that leads all the way from the molecular initiating event to an adverse effect considered relevant in risk assessment. In the case of ecological risk assessment, this generally means well-quantified endpoints of demographic significance that can be used to predict or infer potential population impacts”

ADVERSE OUTCOME PATHWAY CONCEPT



Environmental Toxicology and Chemistry, Volume: 29, Issue: 3, Pages: 730-741, First published: 09 November 2009, DOI: (10.1002/etc.34)

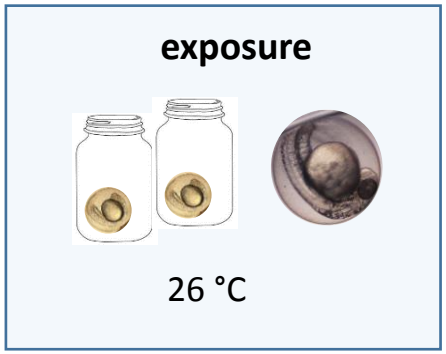
Biological complexity increasing →

STUDYING ADVERSE OUTCOME LINKS IN WP3

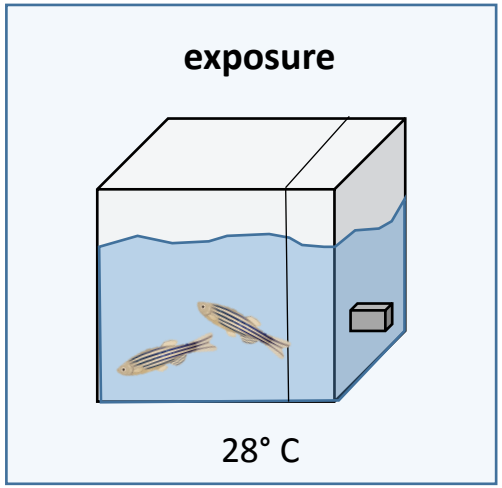
- RWTH Aachen, UPV/EHU, SYKE, NTNU
- Naphthenic North Atlantic crude oil + Finasol 51 / 52 (some tests also with other oil types
- Chemical composition of oil in water and organisms also measured
- Environmental variables considered

ZEBRAFISH EXPOSURE

- Exposure of both embryos and adults
- Naphthenic North-Atlantic crude oil water accommodated fraction (WAF) and dispersant Finasol 52
- Exposure time for embryos up to 120 hpf
- Exposure time for adults 3 and 21 days



embryos



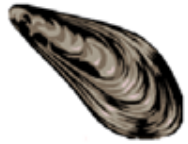
adults

ZEBRAFISH ADVERSE OUTCOME LINKS

Biological complexity increasing →

Population effects	Swimming behaviour	• Embryos: significant reduction of swimming activity during dark phases due to oil and dispersed oil
Organism effects	Morphological changes	• Embryos: Reduced diameter of retinal cell layer (photoreceptor cells)
Organ effects	Enzyme activity	• Embryos: biotransformation, neurotoxicity • Adult: biotransformation, neurotoxicity, oxidative stress
Cellular effects	Transcriptome analysis	• Embryos: strong upregulation of genes involved in xenobiotic metabolism
Molecular interactions		• down-regulation of genes encoding functional and structual components of zebrafish eyes

NORTH ATLANTIC MUSSEL EXPOSURE

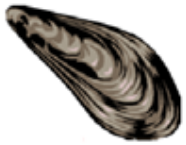


- Mussels from Trondheim acclimatized to 3 temperatures corresponding to 3 different latitudes in the Northern Atlantic: arctic, subarctic and temperate regions

- Exposure to naphthenic North Atlantic crude oil 5% and 25% WAF and 5% WAF and dispersant

- Exposure time 11d, 26d and after 7d recovery period

NORTH ATLANTIC MUSSELS AOLS



Population effects

Organism effects

Organ effects

Cellular effects

Molecular interactions

Condition parameters

- Condition index
- Reproductive status

Histopathology

- Lysosomal condition
- Tissue lesions

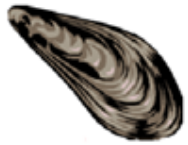
Biochemistry

- Oxidative stress, endocrine disruption, genotoxicity

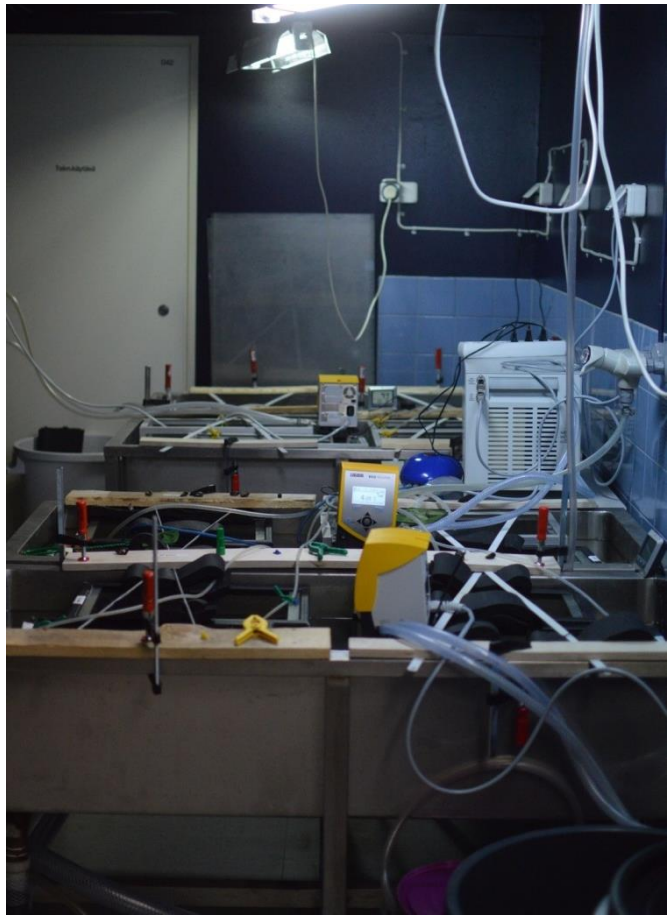
Gene expression

- Analysis ongoing

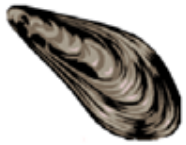
BALTIC SEA MUSSEL EXPOSURE



- Mussels from Hanko (Southern Finland) were acclimatized to two salinities corresponding to northern and southern regions in the Baltic Sea
- Experiments done in two temperatures corresponding to winter and summer conditions
- North Atlantic crude oil WAF 5% and dispersant Finasol 51
- Exposure time 1d, 7d, 21d (winter), 7d recovery (summer)



BALTIC SEA MUSSEL AOLS



Population effects

Organism effects

Organ effects

Cellular effects

Molecular interactions

Condition parameters

- Condition index
- Reproductive status

Histopathology

- Lysosomal condition
- Tissue lesions

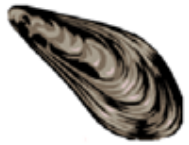
Biochemistry

- Oxidative stress, neurotoxicity, genotoxicity

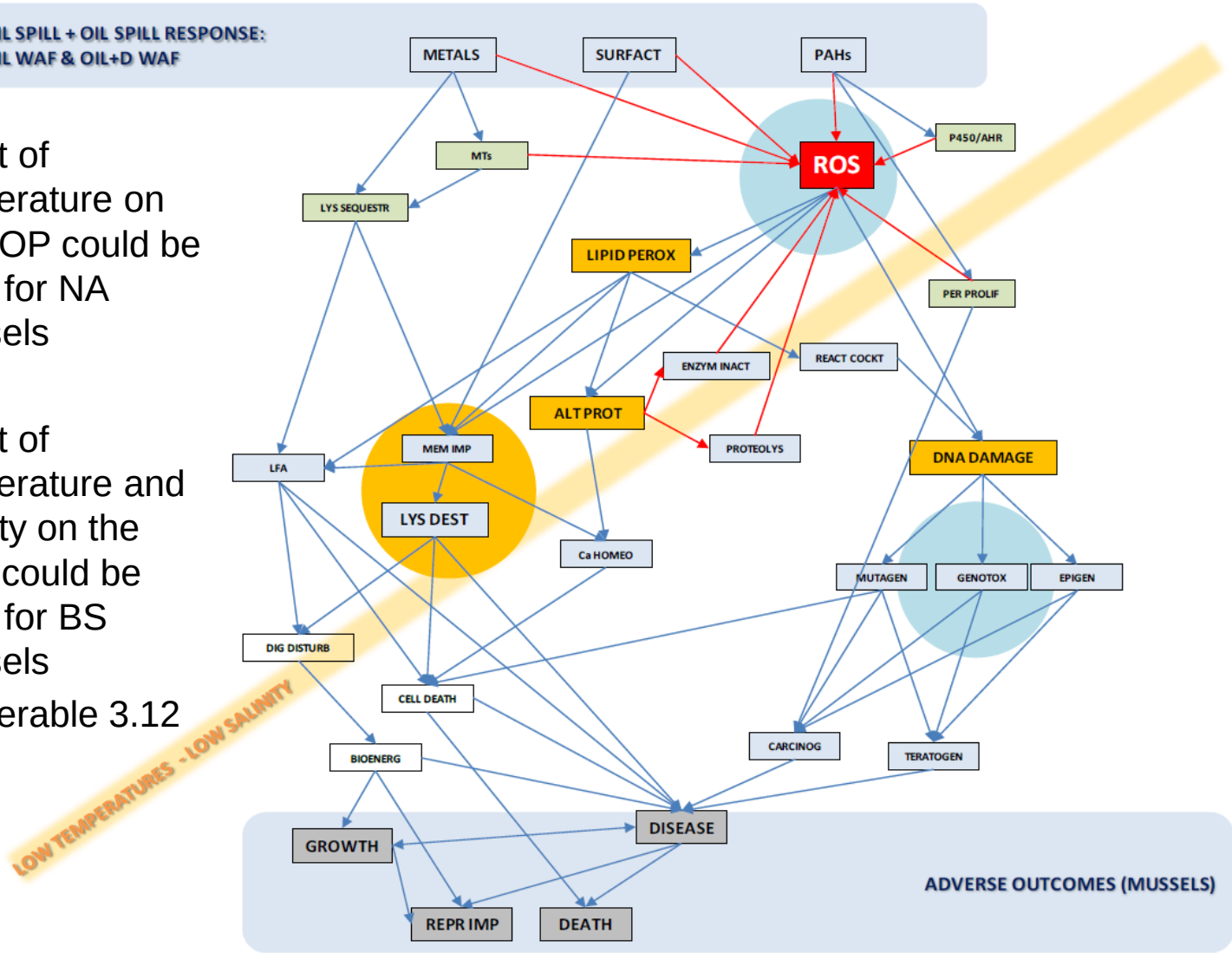
Gene expression

- SOD, CAT, p53, PDRP, MVP and 18S planned

MUSSEL ADVERSE OUTCOME LINKS



- Effect of temperature on the AOP could be seen for NA mussels
- Effect of temperature and salinity on the AOP could be seen for BS mussels
- Deliverable 3.12



COPEPOD EXPOSURE

- Experiments with North Atlantic *Calanus finmarchicus* and Baltic *Limnocalanus macrurus*

- Exposure to WAF and WAF + dispersant of naphthenic North Atlantic crude oil (*C. finmarchicus* also other oil types)

COPEPOD ADVERSE OUTCOME LINKS

Population effects

Organism effects

Organ effects

Cellular effects

Molecular interactions

mortality

Oxidative stress enzymes

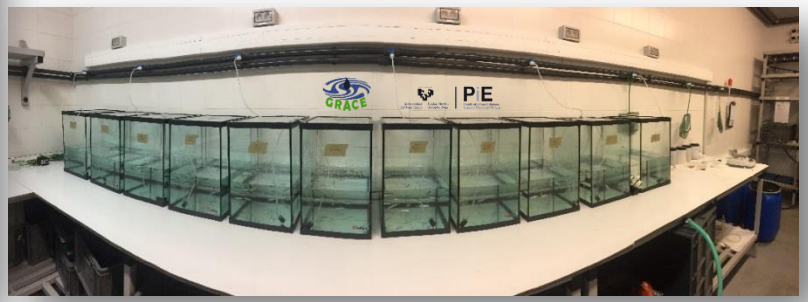
Metabolome profile

Gene expression

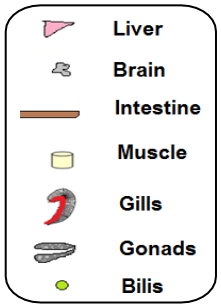
- Acute toxicity LC50: dispersed oil more toxic, differences between oil types
- GST and MDA most promising biomarkers for oil exposure
- Metabolome profile analysis suggests energy balance impairment after oil exposure, malonate and proline identified as good biomarkers for oil exposure
- Biomarker related genes: CYP1A2, CYP330A1, GST, GSH, SOD, CAT, HSP70, HSP90

STICKLEBACK EXPOSURE

Exposure conditions: 15ºC 33 psu
Sampling at day 0, 3, 14.



- C, 5%, 25% of NNA WAF
- C, 5%, 25% of NNA+D WAF



STICKLEBACK ADVERSE OUTCOME LINKS

Population effects

Organism effects

Organ effects

Cellular effects

Molecular interactions

Biometry

- Weight and length
- No differences in GSI or HSI

Histology

- Ongoing

Biochemistry

- Ongoing

Molecular biology

- Preliminary results / ongoing

Hsp 90 = Heat Shock Protein 90 Da
Hsp 70 = Heat Shock Protein 70 Da
Elonf1 = Elongation factor 1

- Valuable information of oil and dispersed oil toxicity mechanisms useful in ecological risk assessment of oil spills and oil spill mitigation
- Different species, different biological levels
- Deliverables D3.11, D3.12, D3.14