Sub-lethal effects of the water soluble fraction of tunnel wash water on juvenile brown trout (*Salmo trutta*)

Mathilde H. Skarsjø¹
Ingvild Marie Dybwad¹, Tor Fredrik Holth¹, Sissel Ranneklev², Merete Grung¹,², Sondre Meland³,⁴, Ketil Hylland¹

¹University of Oslo - Department for Biosciences, ²Norwegian Institute for Water Research (NIVA), ³Norwegian Public Roads Administration, ⁴Norwegian University of Life Sciences, Dep. Of Environmental Sciences
Overview

• Background
• Aim
• Study design
• Results
• Conclusions
Tunnel wash water (TWW)

- Tunnels accumulate road related contaminants between washing events
- TWW contain a high concentration of road related contaminants (e.g. metals and PAHs)
- Sedimentation ponds receiving tunnel wash water removes particle bound pollution
  - The removal of dissolved contaminants may be questioned
Aim

Investigate sub-lethal effects on brown trout exposed to the water soluble fraction of tunnel wash water trough a controlled exposure study.
Study design – exposure study

- Semi-static exposure experiment with four treatments
  - Control x5
  - Positive control x5
    - 150 µg/L Pb and 1µg/L Benzo[a]pyrene
  - Granfoss tunnel wash water x5
  - Nordby tunnel wash water x5

- The tunnel washes were conducted without the use of soap
- Water was filtered before used in the experiment
CYP1A

- Major enzyme conducting phase I metabolism ("detoxification-process") of coplanar chemicals

\[
\text{benzo[a]pyrene} \xrightarrow{\text{O}_2, 2\alpha, 2\text{H}^+} \text{(+benzo[a]pyrene-7,8-epoxide}} \xrightarrow{\text{H}_2\text{O}} \text{(-benzo[a]pyrene-7,8-dihydriodiol}}
\]

Induction of CYP1A

Formation of reactive intermediate metabolites

Increased excretion of coplanar chemicals
PAH-metabolites

1-OH-prene (ng/g)
Day 0 Day 5 Day 25
Control Positive control Granfoss Nordby

3-OH-benze[al]pyrene (ng/g)
Day 0 Day 5 Day 25
Control Positive control Granfoss Nordby

1-OH-phenanthrene (ng/g)
Day 0 Day 5 Day 25
Control Positive control Granfoss Nordby

PAH-metabolites

n = 3-5
OH-phenanthrenes/anthracenes

\[ PAH-\text{metabolite} \]

Day 0 Day 5 Day 25
Control Positive control Granfoss Nordby

Day 0 Day 5 Day 25
Control Positive control Granfoss Nordby

\[ OH-\text{phenanthrenes/anthracenes} \]

\[ n = 3-5 \]
CYP1A

Activity in gills

Activity in liver

Concentration in liver

n = 3-5
ALA-D activity

- Activity of the enzyme δ-aminolevulinic acid dehydratase
- Catalyse the second step in prophyrin and heme biosynthetic pathway

![Graph showing ALA-D activity over days 0 and 25, with different groups indicated by different colors.](image)

*n= 3-5*
Other biomarkers - gene expression

Gill

Liver
Gene expression

Gill

Liver

CYP1A (phase I metabolism) and ALAS (heme synthesis)

Gluthathion S-transferase (phase II metabolism)

Vitellogenin (endocrine effects)

ABC transporter (transport out of cells)

Peroxisome proliferater activated receptor (lipid metabolism)

Methallotionin and Gammagluthamyl cysteine synthethase(ligase) (antioxidant defences)

Heat shock proteins (stress)
Gene expression

Gill

- CYP1A (phase I metabolism) and ALAS (heme synthesis)
- Gluthathion S-transferase (phase II metabolism)
- Vitellogenin (endocrine effects)
- ABC transporter (transport out of cells)

Liver

- Peroxisome proliferater activated receptor (lipid metabolism)
- Methallotionin and Gammagluthamyl cysteine synthethase (antioxidant defences)
- Heat shock proteins (stress)
Conclusions

• TWW exposed trout:
  – Have accumulated PAH-metabolites in bile
  – Have increased activity, concentration and gene expression of CYP1A
  – Does not show signs of lead poisoning
  – Have increased gene expression for several of the genes investigated in both gills and liver
Thank you for the attention!

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