



Metabolomic and transcriptomic study from fish liver Centre for Environment, Fisheries and Aquaculture Science, Cefas Weymouth Laboratory / School of Biosciences, The University of Birmingham, UK

CONTEXT

The goal of this study was to investigate both transcriptomic and metabolomic changes in response to xenoestrogen exposure. Male stickleback fish were used as a model organism for this study as they are environmentally relevant and the stickleback genome draft sequence was available. Key to this study was the ability to carry out both omics techniques upon the same liver tissue homogenate ^{1,2}.

MATERIAL

- Precellys[®]24.
- Precellys[®] kit: 03961-1-003 (ceramic beads 1.4mm).
- Sample: 50 to 150 mg (wet mass) of liver tissue.
- Buffer for omics analyses : 8 ml/g (v/w) methanol and 2.5 ml (v/w) water.
- Qiagen RNEasy RNA kit, DNA-Free (Ambion).

PROTOCOL

- Precellys[®]24: 6400 rpm, 2x10sec, 30s break.
- Metabolomics analysis: Extraction with methanol, chloroform and water to separate the hydrophilic and hydrophobic metabolites / storage at -80°C → one-dimensional ¹H NMR spectroscopy.
- Transcriptomics analysis: Total RNA preparation by RNEasy, DNase treatment, cDNA synthesis and fluorescent labeling for microarraying.

- 1) I. Katsiadaki et al., Aquatic Toxicology doi:10.1016/j.aquatox.2009.07.005
- 2) H. Wu et al., Anal. Biochem. 37 (2008) 204–212



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RESULTS

¹H-NMR metabolomics gave highly reproducible data and multivariate analyses suggested a concentration-response relationship.

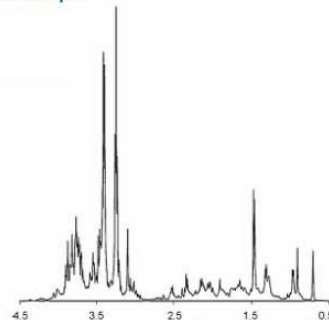


Figure 1: representative one-dimensional ¹H NMR metabolite spectrum of stickleback liver

Transcriptomic analysis showed induction of hepatic vitellogenins and choriogenins, together with a range of other xenoestrogen-responsive genes. Vitellogenin C was identified as a major responsive isoform, in contrast to other fish species.

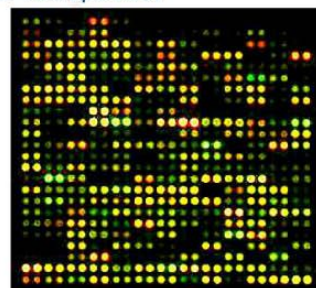


Figure 2: mRNA expression; one of 48 subarrays of the stickleback 15K cDNA array

CONCLUSION

Application of multiple omics techniques requires a homogenization technique that is fast, to avoid metabolite and RNA degradation, that allows no cross-contamination between samples, and that can produce a homogenate suitable for input into multiple different purification procedures.

Precellys[®]24 is easy to use, simple, efficient and much faster than former methods. We have therefore obtained high quality omics data from exactly the same material.

For more details, please contact
precellys@bertin.fr



Problem



Solution



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■東京

〒162-0805 東京都新宿区矢来町 113 番地 TEL(03)3235-0661(代) FAX(03)3235-0669

■大阪

〒532-0005 大阪市淀川区三国本町2丁目12番4号 TEL(06)6396-0501(代) FAX(06)6395-2588

■福岡

〒812-0054 福岡市東区馬出 1 丁目 2 番 23 号 TEL(092)631-1012(代) FAX(092)641-1285

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