Viability assay with fish gill cell line to assess acute toxicity

Commonly used acronym: *RTgill-W1 cell line assay*

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Organisation

Name of the organisation Vlaamse Instelling voor Technologisch Onderzoek (VITO)
Department Health
Country Belgium
Geographical Area Flemish Region

Partners and collaborations

Swiss Federal Institute of Aquatic Science and Technology (EAWAG)

SCOPE OF THE METHOD

The Method relates to	Environment
The Method is situated in	Translational - Applied Research
Type of method	In vitro - Ex vivo
Species from which cells/tissues/organs are derived	Rainbow trout, Oncorhynchus mykiss
Type of cells/tissues/organs	Gill tissue

DESCRIPTION

Method keywords

cell viability test
fish gill cell line
cell metabolic activity
lysosomal membrane integrity
cell membrane integrity

Scientific area keywords

fish acute toxicity
chemical exposure

Method description

The rainbow trout gill cell line assay quantifies cell viability using fluorescent measurements for metabolic activity (Alamar Blue, AB), cell membrane integrity (5-CarboxyFluorescein DiAcetate AcetoxyMethyl ester, CFDA-AM) and lysosomal

membrane integrity (Neutral Red, NR). Chemicals are added to confluent RTgill-W1 cell monolayers in 24-well plates with L-15/ex medium (a simplified version of L-15 cell culture medium without serum). Cells are incubated for 24 hours in the incubator (19°C, normal atmosphere, in the dark). At the end of the exposure, cell viability measurements are performed with 3 fluorescent indicator dyes on the same set of exposed cells.

Lab equipment

Laminar flow ;
Incubator (room temperature, no CO2) ;
Microplate reader for fluorescence detection.

Method status

History of use
Internally validated
Published in peer reviewed journal

PROS, CONS & FUTURE POTENTIAL

Advantages

Cell line model with limited requirements ;
Robust assay: repeatability and reproducibility is shown through inter- and intralaboratory studies ;
Alternative model to predict fish acute toxicity.

Challenges

Exposure of chemicals (bioavailability).

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References

Fischer, M ; Belanger, SE ; Berckmans, P ; Bernhard, MJ ; Blaha, L ; Schmid, DEC ; Dyer, SD ; Haupt, T ; Hermens, JLM ; Hultman, MT ; Laue, H ; Lillicrap, A ; Minarikova, M ; Natsch, A ; Novak, J ; Sinnige, TL ; Tollefsen, KE ; von Niederhausern, V ; Witters, H ; Zupanic, A ; & K. Schirmer (2019). Repeatability and reproducibility of the RTgill-W1 cell line assay for predicting fish acute toxicity. Toxicological Sciences, 169 (2), 353-3640.

Tanneberger, K., Knoebel, M., Busser, F. J. M., Sinnige, T. L.,Hermens, J. L. M., and Schirmer, K. (2013). Predicting fish acute toxicity using a fish gill cell line-based toxicity assay. Environ. Sci. Technol. 47, 1110–1119.

ISO 21115:2019. Water quality — Determination of acute toxicity of water samples and chemicals to a fish gill cell line (RTgill-W1).

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