Adult zebrafish retinal cell culture

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Contact person

Annelies Van Dyck

Organisation

Name of the organisation Katholieke Universiteit Leuven (KUL)

Department Biology

Country Belgium

Geographical Area Flemish Region

SCOPE OF THE METHOD

The Method relates to	Animal health
The Method is situated in	Basic Research
Type of method	In vitro - Ex vivo
Species from which cells/tissues/organs are derived	Zebrafish (Danio rerio)
Type of cells/tissues/organs	Retina

DESCRIPTION

Method keywords

retina

cell culture

neurite outgrowth

microfluidics

Neurons

zebrafish
transgenic lines
compartment-specific treatment
network formation

Scientific area keywords

axonal regeneration
bioenergetics
intraneuronal remodeling
dendritic remodeling
bio-imaging

Method description

Since adult zebrafish retinal ganglion cells (RGCs) can fully regenerate upon axonal injury, these neurons form the ideal subject to study what is driving the recovery process. The use of an adult zebrafish retinal cell culture in a microfluidic set-up enables to create a neuronal network, mimicking the normal neuronal environment. Additionally, it allows to visualize/interfere with specific intraneuronal compartments, providing a clear advantage compared to *in vivo* models. Overall, this state-of-the-art setup facilitates the study of processes associated with spontaneous regeneration at a single-RGC level, and high-throughput *in vitro* screening of potential pro-regenerative/neuroprotective therapeutic targets on a selected set of neurons. The protocol includes: retinal dissection and dissociation, cell culturing, immunostainings, and (confocal) microscopy.

Lab equipment

Microfluidic neuronal culturing devices ;
Inverted (confocal) microscope ;
Horizontal laminar flow ;
Sterile biological safety cabinet ;
Specific cell incubator ;
Dissection microscope.

Method status

Still in development

PROS, CONS & FUTURE POTENTIAL

Advantages

This retinal cell culture provides an animal saving strategy, where in addition, a neuronal network is created between two populations of neurons, mimicking the *in vivo* neuronal environment. Lastly, this microfluidic device enables easy and high-throughput screening and facilitated directed compound administration.

Challenges

As we are the first to try this new technique of culturing adult zebrafish retinal neurons (in a microfluidic setup) a lot of optimization and validation steps are required along the process.

Modifications

Once this protocol is optimized, we will build our own designed microfluidic setup, to fit all the aspects of our research question.

Future & Other applications

This set-up allows the incorporation of a neuronal population of choice in the neuronal network, thereby providing primary information about the effect of selected compounds on neuronal survival/regeneration in an *in vivo* simulated environment.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References

V. Grozdanov, A. Muller, V. Sengottuvel, M. Leibinger, D. Fischer A method for preparing primary retinal cell cultures for evaluating the neuroprotective and neuritogenic effect of factors on axotomized mature CNS neurons Curr. Protoc. Neurosci., Chapter 3 (2010) Unit 3.22

Other remarks

Coordinated by
Method not published

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