

SOP Test 15 - Adult Optokinetic Response

1 Purpose

Assessing visual performance by triggering stereotyped eye movements in adult fish.

2 Scope

This method applies the optokinetic response to adult fish.

3 Safety requirement

4 Associates documents

5 Notes

The only modification to the assay for larvae is the special holding chamber that constrains the fish and flushes water over the gills of the fish. Therefore this assay is particularly useful to spike the water with pharmacological agents.

6 Quality control

7 Equipment

Peristaltic pump (e.g. Gardner SR25, 65 rpm, 24 V DC, with novoprene tube N 4.8x1.6 mm)

Aquarium heater (any will do)

Dissecting microscope (e.g. Olympus SZH-10, Zeiss Stemi)

Infrared-sensitive CCD-camera (e.g. Guppy F-038B NIR, *Allied Vision Technologies*) equipped with an infrared-pass filter (e.g. Olympus RG 715)

Infrared-emitting diodes (e.g. Kingbright BL0106-15-28; $\lambda_{\text{peak}}=940$ nm) shielded by diffusor.

Mirror

LCD projector (e.g. SonyVPL-CX1)

Wide-angle conversion lens (e.g. Raynox HD-4500PRO)

8 Supplies

35 mm cell culture dishes

Centrifuge tubes (50 ml; Cat.-No. 227 261; Greiner Bio-One GmbH, Austria)

Pipettes (plastic)

E3 medium

Methyl cellulose solution (3%, prewarmed to 28°C)

9 Procedure

1. Briefly anesthetized adult fish in 300 mg/l MS-222 dissolved in system water
2. Gently clamped the fish between two pieces of sponge, leaving the head with the eyes and gills free.
3. The pieces of sponge are stabilized by two halves of a plastic pipe and the restrained fish, together with the pieces of sponge and the plastic half pipes, is fitted into a custom-made glass chamber (W x H x L = 12 x 12 x 65 mm³; prepared from a centrifugation tube)
4. A constant flow of fish water is directed straight on its gills through two inlets attached to both sides of the glass chamber. We used a maximal flow-rate of 40 ml/min on each side, which was generated by a simple peristaltic pump.

5. The water effuses from the chamber at its rear end through a third tube and is directed back to the supply tank. The water in the supply tank is maintained at a constant temperature of 28°C using a water bath equipped with a standard aquarium heater.
6. The flow-through chamber is placed under the dissecting microscope, to which an infrared-sensitive CCD-camera, equipped with an infrared-pass filter is attached.
7. The fish in the flow-through chamber is illuminated from below with a cluster of 15 infrared-emitting diodes ($\lambda_{\text{peak}}=940$ nm, BL0106-15-28, *Kingbright*, Taiwan) shielded by a diffusor.
8. A white paper drum (d=9 cm) on a glass plate (d=9 cm) with three small openings at the bottom edge, two for the water-supply tubes and one for the effluent tube, is placed around the fish in the flow-through chamber.
9. Visual patterns are projected via a LCD projector through a wide-angle lens onto a mirror deflecting the image inside the drum. We use open source “Vision Egg” to generate the stimulation pattern, but any other program will do.
10. Record the eye movement with a customized recording software of the measurement computer, and evaluate.
Instead of taking real-time measurements, the stored movie files can be evaluated after the recording session is over (e.g., using Image J software, available at <http://rsb.info.nih.gov/ij/>).
We use a customized software based on LabView 7.1 and NI-IMAQ 3.7 (*National Instruments*, USA) . For details refer to publications listed in additional information.

10 Supporting Information

Mueller, K. P., **Neuhauss, S. C.** (2010). Quantitative measurements of the optokinetic response in adult fish. *J Neurosci Methods*. 186, 29-34