

## **University of Idaho**

### **Animal Care and Use Committee**

#### **Standard Operating Procedure (SOP)**

<b>Number</b>	2001.4
<b>Version</b>	1
<b>Last Updated</b>	12/14/2000
<b>Title</b>	Systemic exposure to BrdU in fish
<b>Species</b>	Goldfish

#### **Purpose**

The adult goldfish retina contains proliferative cells that are capable of ongoing neurogenesis, and regenerative activities in response to injury. This makes the goldfish eye an ideal location for the study of retinal development and retinal regeneration, in a highly accessible situation. Intraocular injection of materials that are suspected or known to influence these processes, followed by experimental determination of effects, has added and continues to add to our knowledge of neurogenesis and neuronal differentiation. Other uses for intraocular injection include the exposure of the retina to bromodeoxyuridine (BrdU), which is taken up by proliferative cells and can be used as a marker for the timing of cell birth.

The intraocular injection procedure carries with it risk of exposure of the investigator to the injected chemicals (which may be cytotoxins), as well as considerations for the health of the animal recovering from the injection.

#### **Potential Impact on Animal Subjects**

Given appropriate anesthetic procedures and proper conditions for recovery, there should be no major health risks. However, if the materials injected are cytotoxins, these materials will cause damage to the goldfish retina, followed in most cases by a regenerative response. Unhealthy animals may be at risk of not recovering from the injection procedure.

#### **Description**

Goldfish are removed from their home aquaria and transported to the laboratory in shoebox-sized containers with system water, then transferred to the BrdU solution.

BrdU is prepared as a 5 mM solution in aquarium water, along with 0.03 g tetracycline/L, and fish are maintained for up to 9 days in 1 L of this solution, held in 2 L beakers (to avoid splashing). Only 2-3 fish are maintained in each beaker. The solution is aerated with portable pumps and aeration stones. The BrdU exposure is done in a fume hood, and the fume hood is clearly marked so that lab personnel are aware of potential hazard. Beakers are loosely covered with parafilm or aluminum foil, and held within a labeled plastic container lined with absorbent material. Fish are fed daily, and 1-2 hours after feeding, the solution is changed.

After the experiment, the BrdU-containing water is disposed of as liquid hazardous waste in accordance with University procedures. All disposable materials coming into contact with BrdU (e.g. weigh boats and other plastics) is disposed of as solid hazardous waste in accordance with University procedures. Fish carcasses are also disposed of as solid hazardous waste. Handling of powdered BrdU is done using appropriate protections (mask, goggles, and protective clothing).

The BrdU exposure is followed by a 1-2 day chase exposure to thymidine (i.e. 5-10mM thymidine in aquarium water, with tetracycline) to replace BrdU for incorporation into proliferating cells.

## References

Raymond PA, Reifler MJ, Rivlin PK. 1988. Regeneration of goldfish retina: rod precursors are a likely source of regenerated cells. *J Neurobiol.* 19:431-63.