

Working with Hygromycin in Zebrafish SOP
University of Washington

Standard Operating Procedures for Chemicals or Processes	
#1 Process (if applicable)	<p>Hygromycin is used as part of an animal research protocol to kill hair cells.</p> <p>Fish are transported to CD056 in dishes. The dishes are transported in a leakproof secondary container such as a Styrofoam or plastic box. Fish are moved using a wide-bore pipette into a 48-well plate, where they are then treated with hygromycin at low concentrations for 1-24hrs.</p> <p>After treatment, fish are euthanized with MS222, and fixed with paraformaldehyde.</p>
#2 Chemicals and Hazards	<p>Small volumes of Embryo Media, Hygromycin, MS222 (zebrafish anaesthesia), paraformaldehyde.</p>
#3 Personal Protective Equipment (PPE)	<p>The following PPE will be worn when working with Zebrafish:</p> <p>Nitrile or latex gloves will be worn when handling fish containers or hazardous chemicals.</p> <p>Safety glasses are kept in a labeled drawer next to the microscope and worn when handling liquids.</p> <p>Individuals will bring their own lab coat to CD056 and wear long pants or long skirt, and fully closed shoes.</p> <p>Gloves will be changed immediately if contaminated, torn, or punctured</p>
#4 Environmental/ Ventilation Controls	<p>Powder and liquid forms of drugs should be weighed, reconstituted, and diluted in a chemical fume hood.</p>
#5 Special Handling Procedures & Storage Requirements	<p>Avoid breathing dust. Avoid contact with eyes, skin and clothing. Signs and symptoms of exposure –Nausea, headache, and vomiting.</p> <p>Zebrafish will be transported in labeled and covered non-breakable, leakproof secondary containers.</p> <p>Chemicals or reagents needed for imaging will also be transported to the room in non-breakable secondary containers</p>
#6 Spill and Accident Procedures	<p>Minimize contact with skin or eyes and avoid inhalation of vapors (liquid) or dust (powder). Liquid –absorb on sand or vermiculite and place in closed containers for disposal. Powder –Sweep up and hold in bag for waste disposal. Ventilate area and wash spill site after material pick-up is complete</p>
#7 Waste Disposal	<p>Correct dilution of drug is mixed and appropriate amount is given to animal. Dilution is saved for further experiments. Little to no waste is generated</p>

#8 Special Precautions for Animal Use (if applicable)	
Particularly hazardous substance involved?	<input type="checkbox"/> YES: Blocks #9 to #11 are Mandatory
	<input checked="" type="checkbox"/> NO: Blocks #9 to #11 are Optional.
#9 Approval Required	
#10 Decontamination	
#11 Designated Area	
Name: _____ Title: _____	
Signature: _____ Date: _____	