

UW Policy for the Euthanasia of Finfish Species – 2013

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Fish commonly used in research: Small fish species (2-6 cm in length) typically utilized in laboratory research include zebrafish, medaka, fathead minnow, goldfish, swordtail, and platyfish among others. Large fish species (>6cm in length) typically used include salmon, trout, tilapia, catfish, hybrid-striped bass, bass, bluegill, sturgeon and others.

Background: The purpose of this document is to provide information regarding UW-approved methods of euthanasia for finfish at the University of Washington. The 2007 AVMA Guidelines on Euthanasia were revised in 2013, and some changes were made with regard to euthanasia of finfish. For example, eugenol is now considered an acceptable method of euthanasia by the AVMA, and has been added to the UW-approved list of approved methods. The methods listed below are not an exhaustive list of all methods covered in the AVMA document; rather, this policy contains the methods that are most commonly used at the UW. The current AVMA Guidelines for the Euthanasia of Animals: 2013 Edition lists some non-physical and physical methods that are not included in this document (e.g., quinaldine sulfate, 2-phenoxyethanol, isoflurane, sevoflurane, pentobarbital (1 step & 2 step), and other physical methods).

Policy: Chemical agents such as buffered tricaine methanesulfonate (MS-222) or buffered benzocaine hydrochloride are generally the method of choice for the euthanasia all fish species. The dosage required for euthanasia with either of these compounds is higher than the anesthetic dose, but varies greatly with species, life stage, size, and water temperature. In addition, a longer time of exposure to the agent (relative to anesthesia) is required to ensure that death occurs. Proposed euthanasia methods not listed below will be reviewed on a case-by-case basis, based on the AVMA Guidelines and the needs of the proposed work.

A. Methods that do not require an adjunct method to ensure death:

Note: For all methods utilizing an immersion bath, fish must be placed in an appropriate volume of water to provide free movement. Early life stages typically require higher concentrations than adults.

1. Tricaine methanesulfonate (MS-222): Dosage: 250-500mg/L, buffered to the pH of the system or source water with sodium bicarbonate (NaHCO₃). Fish should be removed from the euthanasia solution only after ten minutes have passed since their last observed opercular movements (respiration).
Note: Fish euthanized with MS-222 cannot be used for human or animal consumption.
2. Benzocaine hydrochloride: Dosage: \geq 250mg/L, buffered to the pH of the system or source water with sodium bicarbonate (NaHCO₃). Fish should be removed from the euthanasia solution only after ten minutes have passed since cessation of opercular movements (respiration).
Note: Fish euthanized with benzocaine cannot be used for human or animal consumption.
3. Eugenol: Concentrations required for euthanasia will vary depending on species and size of the finfish. Researchers should consult with Veterinary Services to determine the appropriate euthanasia dosage for a particular finfish. Standardized and known concentrations of the essential oil must be used for accurate dosing. Fish should be removed from the euthanasia solution only after ten minutes have passed since their last observed opercular movements (respiration).
Note: Fish euthanized with eugenol cannot be used for human or animal consumption, and must be disposed of according to FDA guidelines.

4. Exposure to Water Saturated with Carbon Dioxide (CO₂): Fish are placed in source or system water, which is then saturated with carbon dioxide delivered by the use of an air stone submerged into the water with a moderate flow rate. The carbon dioxide must be from a compressed gas cylinder. Certification is required for individuals to utilize this technique at the University of Washington and can be obtained through the Animal Use Training Program.
5. Rapid Chilling of Tropical Species: The AVMA Guidelines for the Euthanasia of Animals: 2013 Edition concludes that rapid chilling is acceptable for some tropical finfish such as zebrafish (*Danio rerio*) as long as the transfer from acclimatized temperatures to 2-4°C water occurs rapidly with as little transfer of the warmer water as possible. Fish should be immersed in an ice water bath maintained at a temperature of 2-4°C for a minimum of 20 minutes after cessation of opercular movement, followed by an adjunct physical method such as freezing. The water used for the ice water bath should be system or source water. Fish should not come in direct contact with the ice. Because of surface-to-volume considerations, use of this method is not appropriate for medium to large-bodied tropical finfish (>3.8cm).

B. Methods that must be followed by an adjunct method to ensure death:

1. Sharp Blow to the Head (Blunt Force Trauma): This method must be followed by exsanguination (via severing the large branchial vessels or large caudal tail vessels), pithing, or decapitation. Certification is required for individuals to utilize this technique at the University of Washington and can be obtained through the Animal Use Training Program.
2. Decapitation: Decapitation must be followed by pithing of the brain or other appropriate secondary physical method. Certification is required for individuals to utilize this technique at the University of Washington and can be obtained through the Animal Use Training Program.

References:

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