

IACUC Guidelines for Fish (Excluding Zebrafish) Euthanasia

Last Review: 4/2022

Revision History: 9/2020

Acceptable:

- Intentional overdose by Immersion in anesthetic solutions
 - Fish should be left in the anesthetic solution for a minimum of 30 minutes after cessation of opercular movement
 - Agents: Benzocaine, lidocaine, and MS-222, isoflurane, quinaldine sulfate, 2-phenoxyethanol, ethanol
 - MS222 must be buffered when administered and a concentration of 250 to 500 mg/L
 - Hypoxic tolerant species need a 2 -step method (cichlids and goldfish: step 1 immersion step 2 decapitation, pithing or freezing)
- Rapid chilling for small-bodied tropical and subtropical stenothermic fish

Acceptable with conditions: (needs IACUC approval)

- Immersion in CO₂ saturated water or clove oil and derivatives (eugenol/ isoeugenol)
- Maceration (research)
- Decapitation followed by pithing
- Cervical Transection
- Manually applied blunt force trauma followed by pithing or exsanguination
- Penetrating captive bolt (large fish going into food supply)

Early life stages

- May require higher concentrations of immersion anesthetics or a longer duration of exposure

- Rapid chilling followed by immersion in a dilute sodium hypochlorite or calcium hypochlorite solutions

Adjunctive Methods

- Decapitation
- Pithing
- Exsanguination
- Freezing
- Other physical or chemical methods after the fish have been rendered unconscious prior with an acceptable method

Unacceptable Methods

- Slow chilling or freezing of unanesthetized animals
- Anoxia
- Dessication
- Exposure to caustic chemicals
- Prolonged traumatic injury

Ensuring death

- Loss of movement, loss of reactivity to any stimulus, initial flaccidity
- More useful:
 - Cessation of rhythmic opercular activity for a minimum of 30 minutes
 - Loss of eye-roll- (vestibulo-ocular reflex, the movement of the eye when the fish is rocked from side to side)

MS222 Use

MS222 in solution should be frozen or refrigerated in between use and should be disposed of appropriately if it discolors or is not effective. The expiration date on the label of the powder form should be followed.