Curtin University Standard Operating Procedure

EUTHANASIA OF FISH

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**Aims / Objectives:** To provide guidelines on the euthanasia of fish for any researchers using fish and requiring euthanasia of the fish as part of any research projects at Curtin Aquatic Research Laboratories (CARL).

**Definitions:**

**AQUI-S:** A sedative and anaesthetic agent, easily dispersed in water with a wide safety margin.

**Benzocaine:** A form of local anaesthetic commonly used in fish as a sedative, anaesthetic agent, and euthanasia agent.

**Euthanasia:** From the Greek meaning "good death" and refers to the practice of intentionally ending a life in order to relieve pain and suffering. It is done to minimise pain and try and make it distress free. It is thought to be humane to firstly induce a loss of consciousness prior to the loss of motor function.

**Ike jime:** A method of paralysing fish. It involves the insertion of a spike quickly and directly into the hind brain, thereby causing immediate brain death. When spiked correctly, the fish fins flare and the fish relaxes, immediately ceasing all motion.

**MS 222 – or Tricaine methanesulfonate:** A white powder used for anaesthesia, sedation, or euthanasia of fishes. It is a muscle relaxant that operates by preventing action potentials.
Procedures
Euthanasia of fish requires specific knowledge of the particular species requiring euthanasia. Depending on their size and species, different methods may be recommended.

Prior to any project occurring in CARL, the researcher must complete a risk assessment for the whole project, and if any chemicals are to be used, an individual chemical risk assessment sheet must be completed. This must then be assessed and approved for use by the Health and Safety Manager for Curtin University, prior to AEC approval being granted.

The Safety Data Sheets for individual chemicals can be found through ChemAlert on the Curtin University Website at http://healthandsafety.curtin.edu.au/hazardous_substances/chemicals.cfm

Approved methods for euthanasia include:

1. Chemical Methods - these are the preferred methods.
   a. AQUI-s is a strong sedative/anaesthetic that can also be used for euthanasia. Dose rates for finfish are either 100mg/L for 40 minutes, or 175mg/L for 20 minutes. For larger fish, AQUI-S is usually followed by Ike jime.
   b. Immersion of fish in Benzocaine. This should be at a dose rate of greater than 100mg/L, and the fish must be left in the water for a minimum of 10 minutes after the cessation of opercular movements. It can be followed by physical Ike Jime to ensure death after this time. Benzocaine can be difficult to dissolve, but this can be assisted by using Ethanol.
   c. Immersion of fish in MS 222, which is very water soluble and used in a bath at greater than 100mg/Litre.
   d. Use of an injection of Sodium pentobarbitone intraperitoneally. This is not the preferred method, as requires some handling and removal from the water, but may be used if other methods are unacceptable.

2. Physical Methods
   a. Ike Jime – this method is used when chemical methods are not appropriate for that particular species or research project. An example of this is if the flesh needs to be chemical free for analysis post death.
   b. Ice Slurry – This is not recommended unless the other forms of chemical or physical euthanasia are not suited to the research protocol and the AEC approves it. See the separate SOP on Ice Slurries.
References:

NHMRC 2008 Guidelines to Promote the Wellbeing of Animals Used for Scientific Purposes