THE UNIVERSITY OF ADELAIDE AUSTRALIA

Information on Euthanasia of Fish

As vertebrate animals fish are covered by the Code, and although they may not have the same pathways for pain perception as are recognised in mammals, there is evidence that they do feel pain and therefore humane methods should be employed in the killing of fish.

3.3.18 *When it is necessary to kill an animal, humane procedures must be used. These procedures must avoid pain or distress, be reliable and produce rapid loss of consciousness until death occurs. The procedures should also be compatible with the scientific or educational aims.*
Australian Code of Practice for the Care and Use of Animals for Scientific Purposes. 7th Edition

There are more than 20 000 species of fish which display great diversity in physical characteristics and environmental requirements so the following should be regarded only as a general guide for the reference of the committee. Specific consideration should be made of the type of fish involved in a given project and further information or expert opinion sought where necessary.

Chemical Methods

Humane killing may be accomplished by dissolving anaesthetic agents in the tank water. To avoid distress due to handling or to changes in the physical and chemical parameters of the environment this should be undertaken in the fishes' normal water wherever possible. Lowering the water level, although not to levels that would distress the fish, is advised to facilitate rapid action of the agents. Fasting of the fish prior to euthanasia may enhance absorption by the gut and minimise regurgitation, which may affect the action of the drug on the gills. The tanks should allow for effective observation of the fish. Fish may be killed by physical means once anaesthetised or the agents may be used at high concentrations to produce death directly.

Tricaine methane sulphonate (TMS, MS 222)
This benzocaine type drug acts by depression of the central nervous system and has been reported by an international working party[1] to be the most effective way to kill most fish. It is soluble in both fresh and salt water and should be combined with a buffering agent to neutralise the water to pH 7.5. In large fish it is possible to produce euthanasia by removing the fish from the water and flushing a concentrated solution of MS-222 across the gills using a syringe. This compound is reasonably expensive.

Benzocaine (ethyl aminobenzoate)
This has a similar action to MS 222. It is insoluble in water and must be dissolved in acetone or ethanol before being added to the water. **Benzocaine hydrochloride**, again similar on action to MS-222, is directly soluble in water. Because it has a breakdown time in water of approximately four hours benzocaine preparations may be useful where control of

environmental contamination is important. The Australian Pesticides and Veterinary Medicines Authority (APVMA) has issued a minor use permit for Benzocaine that includes environmental disposal conditions approved by Environment Australia.

Clove oil (eugenol/isoeugenol/methyleugenol)
Clove oil or its constituents are used in food flavouring, dental and fragrance applications although it is not licensed for use in food producing animals. It produces anaesthesia and, at higher concentrations, death in fish. The most active constituent of clove oil, **isoeugenol**, is available commercially as a fish anaesthetic (**AQUI-S™**) and is the only anaesthetic agent licensed in Australia for use in the harvesting of fish for human consumption. In commercial aquaculture anaesthetic doses are given and followed by physical methods of killing, but it is possible to produce euthanasia directly through use of higher concentrations.

Halothane
The anaesthetic halothane may be bubbled through the tank water to produce anaesthesia in the fish. Death must be ensured by physical destruction of the brain once the fish are anaesthetised.

Pentobarbitone
Injectable barbiturates such as pentobarbitone can be used to humanely kill fish, but because this approach necessitates the stress of handling and removal from the water other methods will be preferable in most circumstances. Practical considerations regarding size and number of fish will also limit the use of this technique. The intra-peritoneal route is generally recommended.

Physical Methods

Concussion
Killing with a blow to the back of the head can be an effective and humane method in the hands of an experienced operator. Death should subsequently be confirmed by destruction of the brain.

Cervical dislocation
Small or medium size fish may be killed by inserting a rod or thumb into the mouth and displacing it dorsally whilst stabilising the fish's body with the other hand. The degree of handling required is likely to produce significant stress. This method is not effective or humane in larger fish.

Maceration
Small fish of less than 2cm in length can be humanely killed by mechanical maceration.

Methods for confirming death in anaesthetised or insensible fish

Decapitation
Because there is some doubt about how rapidly a decapitated fish head loses consciousness it is recommended that decapitation only be carried out under anaesthesia or after stunning. This is simple to perform in small fish but more difficult in larger ones.

Pithing
Physical destruction of the brain by inserting a spike or rod and physically disrupting the brain tissue can be used to confirm death in unconscious fish.
In the hands of experienced operators, for example in commercial tuna aquaculture or in large game fishing, killing of the fish by spiking/pithing is likely to produce a rapid and humane death.

Exsanguination
Severing of major vessels can be performed to prevent recovery in fish which have been rendered insensible. Due to the relatively high resistance to hypoxia shown by fish and the potential technical difficulty of locating appropriate vessels exsanguination alone does not provide a rapid humane means of killing a conscious fish.

Inhumane methods for the euthanasia of fish [1]

(Specific experimental justification would be necessary for any of these methods to be considered)

Removal from water
Distress is cause because of the prolonged time to unconsciousness. Cooling prolongs this period markedly.

Electrocution
This may be ineffective in some fish. Alternating current produces tetanic muscle contractions and not anaesthesia. Electrical current obviously also poses a risk to the operator.
Where non-lethal electrostunning of fish is suggested (e.g. for survey purposes) careful consideration of the method and likely effect is necessary.

Hypothermia
Cooling fish with ice or in a freezer may produce immobility but prolongs the time to insensibility and does not reduce the ability to feel pain.

Hyperthermia
Fish placed in hot water will close their opercula tightly and thereby maintain a depot of oxygen which will prolong the time to insensibility whilst very hot or boiling water will produce extreme pain.

Carbon dioxide
Carbon dioxide is very slow acting in fish and produces a period of reflex nociceptive hyperactivity (response to pH and carbon dioxide concentration), followed by immobility prior to insensibility.

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1. Recommendations for euthanasia of experimental animals: Part 2
Working Party Report
Laboratory Animals (1997) 31, 1-32