



## CO2 EUTHANASIA OF RODENTS

### Overview/Purpose

Euthanasia techniques should result in rapid loss of consciousness followed by cardiac or respiratory arrest and ultimately loss of brain function. Methods used must be listed in the IACUC protocol. The selected method of euthanasia must be consistent with the recommendations of the [2013 AVMA Guidelines on Euthanasia](#) unless scientifically justified. Improper euthanasia is considered noncompliance with PHS policy and the [Guide for the Care and Use of Laboratory Animals](#) and is reportable to the Office of Laboratory Animal Welfare (OLAW) (<http://grants.nih.gov/grants/guide/notice-files/NOT-OD-02-062.html>).

### Definitions

1. **Rodents** – mammals of the order Rodentia which includes mice, rats, hamsters, and guinea pigs

### Requirements

1. **Personnel must be appropriately trained to perform euthanasia.** Training is available free of charge by University Laboratory Animal Resources (<http://ular.osu.edu/training/>).
2. **CO2 euthanasia:** The only IACUC approved source of CO2 is compressed gas in cylinders because the inflow to the chamber can be regulated precisely. Dry ice as a source of CO2 is unacceptable. **The flow rate for CO2 should be set to 10-30% displacement of the chamber volume/min.** Carbon dioxide flow must be maintained for at least 1 minute after respiratory arrest. **Chambers prefilled with CO2 are unacceptable.** The chamber should be inverted and cleaned between groups.
3. **Animals must be observed during euthanasia** to ensure that death is humane. If chambers are used to contain animals during administration of inhalants, the chambers must be clear or have a viewing port.
4. **Confirmation of death is required** prior to disposal. The Ohio State University IACUC **requires a secondary physical method** of euthanasia following euthanasia from an inhaled agent and confirmed lack of consciousness. Acceptable secondary methods include creation of a pneumothorax, removal of a vital organ, decapitation, exsanguination, or cervical dislocation. Cervical dislocation must not be performed in rats >200 gm.
5. **To minimize stress:** animals held in a cage awaiting euthanasia must be compatible (e.g. multiple foreign males must not be placed together for euthanasia); animals must be of the same species; and the total number of animals in a single cage should not exceed 8 in a small rodent cage or 15 in a large rodent cage for mice, or 2 (>250 gm) or 4 (<250 gm) rats or guinea pigs in a large rodent cage. To minimize stress, euthanasia in the home cage is preferred by placing the cage in the chamber.
6. Disposal bags should be labeled with the date, initials, and name of the Principal Investigator (PI).

## Applicable Regulations

1. Animal Welfare Act (AWA, Public Law 89-544, 7 U.S.C.)
2. Animal Welfare Act Regulations (AWAR, 9 CFR, Chapter 1, Subchapter A)
3. Health Research Extension Act of 1985 and Public Health Service (PHS) Policy on Humane Care and Use of Laboratory Animals
4. National Research Council *Guide for the Care and Use of Laboratory Animals*, Eighth Edition. National Academy of Sciences, 2011
5. AVMA Guidelines for the Euthanasia of Animals

## Additional Information/Guidance

1. None

## History of Revisions

**034-00** - new policy approved 06/01/09

**034-01** – information was removed and reflected in another policy approved 05/17/13

**034-02** – revisions reflect a new format for the policy approved 12/20/13

**034-03** – revisions reflect a new format for the policy including a definition of rodent and changing the title to remove g.pigs, approved 11/18/2016