“Using Carbon Dioxide as a Method of Euthanasia”

Euthanasia of experimental animals must be performed in a humane manner. The method of euthanasia depends on the species, the requirements of the research and the ability and training of the person performing the procedure. Methods which induce rapid unconsciousness with minimal pain or distress prior to death, such as anesthetic overdose or carbon dioxide inhalation in small rodent species are preferred.

This policy addresses the use of the commonly employed method of Carbon Dioxide (CO₂) gas exposure to euthanize rodents. Metered, controlled-flow, compressed CO₂ gas is the only approved source of CO₂ for euthanasia. CO₂ from other sources such as dry ice are not acceptable.

The person performing euthanasia must be trained in the specific method used. Training is provided in the introductory handling class for all new animal research staff. All trainees must display proficiency and knowledge of the procedure in order to satisfactorily complete the course.

Mice should be euthanatized in their home cage fitted with a solid top for CO₂ delivery whenever possible, in order to minimize stress. The rate of CO₂ flow to the euthanasia compartment/cage must be set properly to displace approximately 20% (10-30%) of the volume of air per minute. An appropriate flow meter must be in place to regulate the displacement rate. Successful euthanasia and death of the animal must be confirmed. This is particularly important after CO₂ euthanasia, since some animals which have ceased breathing may subsequently recover consciousness. To confirm that euthanasia is complete, the euthanatized animal must be observed for lack of respiratory or cardiac activity for one full minute, and the lack of response to a deep pain stimulus.
A terminal, definitive method of euthanasia such as cervical dislocation, decapitation, or bilateral thoracotomy is recommended.

Neonatal ("pinkie") rodents are especially resistant to CO2 asphyxiation. Neonates must be exposed to CO2 for at least 15 minutes. For neonates, a secondary physical method of euthanasia is required to ensure death (e.g. cervical dislocation, decapitation, bilateral pneumothorax).

EUTHANASIA SERVICES PROVIDED BY THE INSTITUTE FOR ANIMAL STUDIES

Euthanasia of unwanted experimental animals may be requested as a service to be performed by the Institute for Animal Studies. Euthanasia may be requested by placing a signed "Animal to be Discarded" tag (purple card) on the animal's cage in the animal room. Alternatively, mice, rats, or other small rodents may also be left in the designated (euthanasia) room in each animal facility.

Animals tagged with a purple card for euthanasia or animals placed in the IAS "euthanasia room" must be housed according to the same standards of care for all animals - with adequate food, water, and clean bedding - in cages that are NOT OVERCROWDED.

Nursing pups must be left with a lactating female; if a lactating female is not available, the pups must be killed immediately by the investigator's staff.

Animals that appear to be suffering, stressed, or distressed may not be purple-tagged in the animal room or left unattended in the IAS euthanasia rooms – these animals must be humanely euthanized by the researcher's staff as soon as possible.

ALTERNATIVE-ACCEPTABLE METHODS OF EUTHANASIA

The Einstein Institutional Animal Care and Use Committee (IACUC) and the Institute for Animal Studies (IAS) have approved a list of additional euthanasia methods based on current American Veterinary Medical Association (1) (AVMA Guidelines for Euthanasia) and National Institutes of Health (2) (NIH Guidelines) recommendations. Information for individual species is provided in the table: Recommended Doses for Anesthesia, Analgesia & Euthanasia (3) provided as a Resource on the IAS web page.

Specific guidelines for the physical method of cervical dislocation for euthanasia is addressed in IACUC Policy 9903 (4).
(1) AVMA Guidelines for the Euthanasia of Animals: 2013

(2) NIH Animal Research Advisory Committee Guidelines

(3) Recommended Doses for Anesthesia, Analgesia and Euthanasia

(4) Policy 9903 “Cervical Dislocation as a Method of Euthanasia”