GUIDELINES FOR RODENT EUTHANASIA IN FIXED FACILITIES
(Mice and Rats)

Purpose: To provide guidance regarding humane and effective euthanasia of mice and rats. Investigators should consult with the Attending Veterinarian (AV) for additional information, questions, or unique circumstances that may require deviation from these guidelines.

Background: According to the Guide for the Care and Use of Laboratory Animals and PHS Policy on the Humane Care and Use of Laboratory Animals, methods of euthanasia must follow the most current AVMA (American Veterinary Medical Association) Guidelines on Euthanasia (http://www.avma.org/resources/euthanasia.pdf). In addition, methods must be specified and approved in Animal Care and Use protocols. Methods are chosen to minimize pain and distress to the mice and rats with consideration of the scientific needs of the research.

Guidelines: The method of euthanasia must be an acceptable method as outlined in the 2007 AVMA Guidelines on Euthanasia. Scientific justification is required if conditional methods, including cervical dislocation and decapitation, are used on mice and rats over 14 days of age.

Euthanasia should be performed in procedure rooms or laboratory space away from other mice and rats, when possible, and from general lab foot traffic. Death must be ensured prior to processing the animal for disposal.

Chemical methods of euthanasia (CO2 chamber, inhalant anesthetics) must be followed with a physical method (cervical dislocation, bilateral thoracotomy, or exsanguination) to ensure death.

A. Use of Carbon Dioxide (CO2) for Euthanasia
A CO2 chamber is the most commonly used method of euthanasia for small rodents. The chamber must allow visualization of the mice and rats during euthanasia. Proper technique must be followed to ensure a humane death, as CO2 may have noxious properties. The proper steps to be followed are listed below.

1. Place the mice or rats into a clean chamber that is not pre-charged with CO2.
2. Do not overcrowd the chamber; all mice or rats in the chamber must be able to make normal postural adjustments.
3. Gradually increase the flow of CO2 into the sealed chamber to minimize distress. For example, an appropriate flow rate is approximately 10-20% of the chamber volume per minute. Carbon dioxide flow rate is dependent on cage or chamber size, e.g., a 10 liter box should have a flow rate of approximately 1-2 liters CO2/minute.

The University of Texas at El Paso (UTEP)
Attachment. The University of Texas at El Paso Incidental Death/Euthanasia Record
4. Leave the mice or rats in the chamber for 4-5 minutes until respiration has ceased.
5. Remove the mice or rats and confirm the absence of respiration. A physical method (cervical dislocation, bilateral thoracotomy, exsanguination) is to be used in conjunction with CO₂ to ensure death.
6. Clean the chamber with disinfectant to remove all urine, feces and fur. Leave the chamber lid off to air out the residual amount of CO₂ in the chamber.

**NOTE:** Use of CO₂ generated from dry ice is not an acceptable method of euthanasia.

**B. Euthanasia of Mouse and Rat Fetuses and Neonates**

Neonatal rodents are resistant to the effects of CO₂, therefore, alternative methods are required. (Mice and rats older than 14 days should be euthanized following the guidelines for adult rodents in the 2007 AVMA Guidelines on Euthanasia).

1. **Fetuses up to 14 days of gestation:** Euthanasia of the dam or removal of the fetus results in rapid fetal death since fetuses cannot survive outside of the uterus.
2. **Fetuses from 15 days of gestation to birth:** Decapitation with surgical scissors, hypothermia, or cervical dislocation are acceptable physical methods. An injection of a chemical anesthetic overdose is an acceptable chemical method. The Attending Veterinarian should be consulted for the appropriate dosage information.
3. **Neonates up to 14 days of age:** Injection of chemical anesthetics, decapitation, and cervical dislocation are acceptable methods of euthanasia. Hypothermia may be used as a method of euthanasia of altricial neonates provided they are less than 5 days old and they are not placed directly on the frozen surface (i.e., place them in a latex bag or cloth).

**C. Animal Disposal and Documentation**

1. After death has been ensured, place the mouse or rat carcasses in a disposable waterproof biohazard bag (provided by Veterinary Services).
2. Complete the Veterinary Services (VS) Incidental Death/Euthanasia Record. Forms are located inside the viviaria near the entrance. This form must be filled out immediately following the procedure. A sample form is attached.
3. Seal the waterproof biohazard bag. Record the protocol number, date, and number of animals euthanized directly on the bag with a permanent maker, and place the bag in one of the freezers specifically designated for carcasses located within the vivaria.
4. Veterinary Services and Environmental Health and Safety will ensure incineration or alternate appropriate disposal is completed. Mice and rats that are infectious must be disposed according to the procedures stated in the Principal Investigator’s approved Animal Use Protocol.
5. Place the completed Incidental Death/Euthanasia Record in one of the documentation bins; bins are located within each vivaria.
The University of Texas at El Paso Incidental Death/Euthanasia Record

This form must be completed by all researchers and staff to record and track all animal deaths regardless of whether an animal is found dead or is euthanized.

All attempts will be made by the staff to contact the Principal Investigator, or their designee, prior to euthanasia of any animals, particularly those animals currently on study.

Protocol No.: A-2009  
PI: Dr. J. Doe  
Date: 11 Dec 08

Species: *Mus musculus*  
Strain: Balb/C  
Sex: Male  
DOB (Age): 5 weeks

Facility: BRB  
Room: 1.000  
Number Euthanized: 3

Found Dead: y / n (circle one)  
Euthanized: y / n (circle one)  
Disposition of the carcass(es): Freezer

Reason for Euthanasia: Humane endpoint/Found injured/Complications/Found deceased/etc.

Person Performing Euthanasia: Dr. J. Doe  
Total Number Euthanized: 3 (1 treated)

Primary Method of Euthanasia: carbon dioxide  
Secondary Method of Euthanasia: cervical dislocation

History: Four animals on study had surgical procedures and developed complications.

If on study, describe procedures and duration: mini-pump implantation

If had surgery: Type and when did it occur? mini-pump implantation/9 Dec 08

Was animal ill prior to death? y / n (circle one)  
Eating and drinking normally prior to death? y / n (circle one)

At time of death, was the cage checked to ensure adequate feed and functioning water bottle? y / n (circle one)

Further comments on history if any (continue on page 2 if needed): Treatment was prescribed by AV (see page 2); 1 of 4 animals responded well to treatment; 3 were euthanized.

Notifications:

Prinicipal Investigator: y / n  
Date:  
Time:

Phone Instructions:

Secondary Contact: y / n  
Date:  
Time:

Phone Instructions:

Tertiary Contact: y / n  
Date:  
Time:

Phone Instructions:

Veterinarian: y / n  
Date:  
Time:

Phone Instructions:

Necropsy done by: y / n  
Date:  
Time:

Samples taken:

Disposition of samples:

Attachments (necropsy reports, pathology reports):

Person(s) completing this form: Dr. J. Doe

Signatures: / S /
The University of Texas at El Paso Incidental Death/Euthanasia Record (continued)

Additional amplifying information:

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<td>e.g., description of treatment prior to euthanasia.</td>
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