

Conventional Per Diem

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Summary Reagents and Materials Protocol

Summary:

The purpose of this policy is to outline the minimum standards of care for rodents based on the Animal Welfare Act, the Public Health Service Policy, and the ILAR Guide for the Care and Use of Laboratory Animals. These procedures are to be followed by trained staff responsible for providing husbandry care for the mouse colonies at the Mouse Biology Program.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
ILAR Guide		
PPE (laboratory coat, shoes/booties, exam gloves)		
Mouse cage setup (sterile cage and lid, food and		
water holder, food, water, enrichment)		
Mouse cage cards		
Litter/wean cage cards		
Pens (sharpie and expo)		
Feed barrel with sealable lid		
General waste barrel with sealable lid		
Dustpan and hand brush set		
Disinfectant		
Temperature monitoring apparatus		
Animal room monitoring sheet		
Compressed carbon dioxide (CO ₂) tank with		
functioning flow meter and attached hose		
Euthanasia cage and lid		
Decapitation Scissors		
Plastic bag		
Wypalls		

Protocol:

1.0 Daily Procedure:

Note: daily tasks must be performed every day 365 days a year 7 days a week including weekends and holidays and should be completed with appropriate PPE in place (refer to MBPVIV-20-102.2nd Personal Protective Equipment SOP)

1.1 Provide nutritionally adequate ad libitum feed, unless directed otherwise and approved in the applicable Animal Care and Use Protocol. Food must be uncontaminated, fresh and topped off as needed.

- 1.2 MBP primarily uses two types of rodent chow Envigo 2918 Irradiated (standard diet, referred to as "2918") and Envigo 2919 Irradiated (high fat diet, referred to as "2919"). Special diets may be required for special needs animals, client studies, etc. and will always be clearly indicated with a "SPECIAL DIET" card on the front of the cage.
- 1.3 Rodent chow must be used within 6 months of the milling date (*Guide*, pg. 39) unless manufacturer guidelines document a shorter or longer expiration date.
 - 1.3.1 Large amounts of feed are to be stored in the gray or yellow barrels in the room that are labeled with "FEED" on the lid, ensuring that the appropriate type of feed is stocked (refer to the barrel labels). If the label has fallen off of the lid or has become damaged, a new label must be printed. Barrels and lids must routinely be checked for damage, such as crack or holes, and replaced if necessary.
 - 1.3.2 The barrel fill date, mill date, expiration date, feed liner change date, and feed can disinfected date must be written in ink on the feed barrel label (attached to the barrel handle).
 - 1.3.3 Special diets may contain additives which reduce the shelf life or require special storage (refrigeration, etc.). Always refer to the diet label for proper storage and expiration dates.
 - 1.3.4 Disinfection of Feed Storage Bins and Accessories are followed as per MBP SOPs.
- 1.4 Provide sufficient potable water (*AWA section 3.55*).
- 1.5 Observe each animal and check for health concerns.
 - 1.5.1 Record sick or dead animals in accordance with the Vivarium Procedure for reporting sick or dead animals, using the Veterinary Health Check Request in Mosaic. Follow up procedures for addressing *Sick, Injured or Dead Mice* are followed as per MBP SOPs.
- 1.6 Change heavily soiled or flooded cages when discovered.
- 1.7 Keep room clean and organized.
- 1.8 Record minimum and maximum temperatures, (ideal room temperature range for rodents= 68-79° F, *Guide*). Reporting of out of range temperatures follow MBP SOPs.
- 1.9 Check the cage card daily to ensure all pertinent information for that colony is current; update as needed.
- 1.10Record completion of daily room tasks, initial and date on the Animal Room Monitoring.

2.0 <u>Weekly Procedure:</u>

Weekly Tasks

- 5.1 Remove each cage from the rack weekly, opening the cage, and check on the status of the mice. In addition to this, also:
 - 5.1.1 Top off food.
 - 5.1.2 Provide clean water HydroPacs or bottles
- 5.2 Other weekly non-animal tasks that are to be completed within the calendar week and marked off on the Mouse Room Monitoring Sheet.

3.0 Biweekly Procedure:

Biweekly Tasks

- 3.1 The biweekly tasks include: Changing or checking the cages and wiping/disinfecting the racks.
- 3.2 Biweekly tasks must be marked on the Mouse Room Monitoring Sheet and the racks that were changed or checked for the week must be indicated.
- 3.3 Details of Cage Changes and Cage Checks can are outlined in MBP SOPs and can be provided upon request.

4.0 Monthly Procedure:

Monthly Tasks

Note: At least once per calendar month

4.1 Change mop head

5.0 As Needed Procedure:

- 5.1 Change feed barrel liner
- 5.2 Change lids and Hydropac cups

6.0 Quarterly Procedure:

Quarterly Tasks

Note: At least once every 180 days

6.1 Disinfect the animal room (includes walls, floors and ceilings).

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- 6.2 Disinfect feed barrels and lids, replace feed liner.
- 6.3 Submit sentinels.

7.0 <u>General Procedures:</u>

7.1 Facilities

- 7.1.1 Physical Plant and Facilities Maintenance policy details are outlines in MBP SOPs and available upon request.
- 7.1.2 Temperature alarms must be installed in animal rooms housing USDA covered species.
- 7.1.3 Exceptions to this policy must be on file with the IACUC.
- 7.1.4 Minimum of 10-15 fresh room air exchanges per hour (Guide, pg. 32).
- 7.1.5 "Floors should be moisture-resistant, nonabsorbent, impact-resistant, and relatively smooth..."(*Guide*, pg. 74).

7.2 Caging

- 7.2.1 Recommended floor space per rodent (see chart below *Guide*, pg. 27).
- 7.2.2 All animals at MBP housed in Optimice cages are to be housed no more than 5 adult mice per cage in non-divided cages. Duplex cages can house 2 adult mice per side.
- 7.2.3 Cages must be in good condition and escape-proof.
- 7.2.4 Caging supplies with cracks may cause injury to the animals and must be discarded.
 - 7.2.4.1 All items must be marked with Sharpie "BROKEN", discarded, and replaced immediately.

Mouse Cage Size Requirements:

Weight of Animal	Floor Area/Animal (inches squared)	Height (inches)
<10 grams	6	5
Up to 15 grams	8	5
Up to 25 grams	12	5
>25 grams	>15	5

7.3 Identification

- 7.3.1 Rodents can be individually identified in addition to having an appropriately labeled cage card (Refer to Identification of Animals policy).
- 7.3.2 Appropriate identification for mice includes ear tag, ear notch, tail/toe tattoo, microchip, toe clipping.

7.4 Environmental Enrichment

- 7.4.1 Examples of rodent enrichments that are available and promote species-typical behaviors include pair or group housing, shelters (Shepard's shacks), nesting materials, and crinkle bedding material.
- 7.4.2 Devices used for environmental enrichment must be easily sanitized or disposed of when cages are changed.
- 7.4.3 Devices must be checked regularly for wear and discarded if they pose a safety hazard.

7.5 Euthanasia

7.5.2

- 7.5.1 Carbon dioxide
 - A dedicated CO₂ chamber is the most commonly used method of euthanasia for small rodents.
 7.5.2.1 The chamber must allow visualization of the mice during euthanasia. Place the mice into a clean chamber that is <u>not</u> pre-charged with CO₂.
 - 7.5.2.2 Do <u>not</u> overcrowd the chamber; all mice in the chamber must be able to make normal postural adjustments.
 - 7.5.2.3 The flow of CO₂ from the tank into the sealed chamber should be set at a flow rate to minimize distress. At MBP, this value is determined by using a displacement rate of 65%.
 7.5.2.3.1 This follows the AVMA guidelines of 30-70% per minute displacement of room air with CO₂.
 - 7.5.2.4 Leave the mice in the chamber for 4-5 minutes until respiration has ceased. Let cage sit for one additional minute to be sure respiration has ceased before removing the lid.
 - 7.5.2.5 A physical method (cervical dislocation, bilateral thoracotomy or exsanguination) may be used in conjunction with CO₂ to ensure death.

- 7.5.2.6 Remove the deceased mice and place into a plastic, resealable bag.
- 7.5.2.7 Using a permanent marker, record the room number, date and your initials on the bag if appropriate.
- 7.5.2.8 Sanitize the euthanasia cage with appropriate disinfectant (MB-10, Clorox wipes, etc.) to remove all urine, feces and fur.
- 7.5.2.9 After the cage has dried, place a new Wypall on the bottom, replace the lid, and position the cage back on the rack.
- 7.5.2.10 In M3 the cage used for euthanasia is to be placed in the dirty corridor and sent out as a dirty cage to cage wash. A new cage must be used each time euthanasia is being done and must not be retained in the animal room.
- 7.5.3 CO₂ generated from dry ice is <u>not</u> an acceptable method of euthanasia. <u>Never</u> use dry ice as a source of CO₂.
- 7.5.4 Euthanasia of Mouse Neonates
 - 7.5.4.1 Neonatal rodents are resistant to the effects of CO₂; therefore, alternative methods are required.
 - 7.5.4.2 <u>Neonates 0-7 days of age</u>
 - 7.5.4.2.1 Hypothermia may be used as a method of euthanasia of altricial neonates, provided they are **not** placed directly on the frozen surface (i.e., place in a paper towel and set that inside a resealable, clear bag).
 - 7.5.4.2.2 Decapitation using sharp scissors is acceptable for altricial neonates.
 - 7.5.4.3 <u>Neonates 8-14 days of age</u>: Injection of chemical anesthetics, decapitation, and cervical dislocation are acceptable methods of euthanasia.
 - 7.5.4.3.1 Injection of an overdose of chemical anesthetics should be used whenever possible.
 - 7.5.4.3.2 Cervical dislocation is an acceptable method of euthanasia with appropriate training and demonstrated proficiency.
 - 7.5.4.3.3 Decapitation is an acceptable method of euthanasia with documented training. Depending on the developmental stage of animals, this technique may require the use of a guillotine.