

STANDARD OPERATING PROCEDURES

DIVISION OF COMPARATIVE MEDICINE

UNIVERSITY OF SOUTH FLORIDA

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TITLE:	Rodent Husbandry
SCOPE:	Animal Care Personnel
RESPONSIBILITY:	Facility Manager, Technical Staff, Veterinary Staff
PURPOSE:	To Outline the Proper Procedures for Receiving, Physical Examination, Evaluating Health Status, Handling, Care, and Husbandry Practices Related to Rodents

I. PURPOSE

1. To ensure the highest quality of animal health, and to preclude the development of spontaneous diseases or disorders which could compromise the integrity of studies, and the interpretation of results.
2. To ensure personnel handling rodents perform their duties in a manner that complies with all current governing laws, regulations, and guidelines.

II. RESPONSIBILITY

1. The Veterinarians oversee all aspects of animal health and are assisted by all program staff.
2. Facility Managers ensure implementation of all procedures.

III. PROCEDURES

1. Receipt

- a. Individual cage cards are prepared for each cage and include the following information:
 1. Principal Investigator
 2. IACUC #
 3. Species +/- strain
 4. Sex
 5. DOB (age on arrival if unavailable) or body weight
 6. Arrival date
 7. Source
 8. Prior procedures or conditioning performed
- b. Shipping containers are inspected for signs of damage and shipment accuracy.
- c. Technician will notify the Facility Manager regarding any damage to shipping containers or contents.
- d. Containers that are damaged, wet, or contaminated are rejected, and animals are euthanatized.
- e. Facility Manager will notify the Animal Shipping/Ordering Coordinator of damaged or rejected shipments or orders improperly filled (i.e., weight, age, number requested).
- f. A veterinarian is consulted whenever rodents in undamaged containers appear ill or members have died in transit.

- g. Shipping containers are lightly sprayed with Oxivir Tb and allowed to set for 5 minutes prior to being taken to the housing room.
- h. Animals are placed in cages according to *Guide* (i.e., *Guide for the Care and Use of Laboratory Animals*) recommendations.

Mice	Weight (g)	Static	Green Line	Blue Line
	up to 25	5	5	5
	> 25	4	5	5
	female with litter +/- male	1	1	1
Rats	Weight (g)	Static	Green Line	Blue Line
	up to 400	3	3	5
	up to 500	2	2	3
	> 500	2	2	3
	female with litter +/- male	1	1	1

- i. Animals are observed for signs of illness or any abnormality.
- j. Number of new animals and new cages are recorded on the ***Per Diem Sheet*** and ***Progress Notes*** located in the ***Room Log Book***. Notations in the ***Progress Notes*** will include the vendor source and a statement reflecting general appearance on arrival.
- k. Room technicians ensure there are no animals remaining in shipping containers prior to removal from the housing room. Animal care staff ensure a second time that all shipping containers have been opened and are empty prior to removal from the facility for disposal.

2. Handling and Restraint

- a. **Rats:** Gently handled, rats become tame and will rarely bite unless startled or hurt. Rats may be picked up by the base of the tail for brief periods, such as observation or cage changes. For a more secure restraint, grasp the rat by placing one hand firmly over the animal's back and rib cage. Gently but firmly press the forelegs toward the head and restrain the head with thumb and index finger immediately behind the mandibles; the other hand should provide support or grasp the tail base. An alternative hold, useful for dosing or examination, is referred to as the 'v' hold. Here, the technician positions the animals head between the index and middle finger and wraps the ring finger and thumb gently around/under the elbows. In this hold, the rat's head may be secured and the body supported in a manner that makes most dosing routes easily carried out.
- b. **Mice:** Mice can also be picked up by grasping the base of the tail between the thumb and forefinger. Restraint for manipulations is performed by grasping the scruff of the neck using the thumb and forefinger and securing the tail against the palm with the fourth and fifth fingers. Grasping as much loose skin as possible will prevent a possible bite.
- c. **Hamsters:** Hamsters are likely to bite, if unaccustomed to handling, startled, or awakened suddenly. A small container can be used for transferring between cages. Restraint for manipulations involves placing one hand directly over the animal and pressing it flat as one curls the fingers and thumb in a manner that allows all the loose skin of the animals' back to be tightly scruffed.

- d. **Gerbils:** Grasp by the base of the tail to pick up animals for observation or cage changes. The tail tip is very fragile and picking the animal up by the tip can result in injury to the tail. Another method is to cup both hands under the gerbil and gently lift it from the cage. Restraint of a gerbil for examination or treatment is similar to the restraint used for a mouse.

3. Environmental Enrichment and Social Housing

- a. Appropriate social interactions among members of the same species and enrichment of the animal's environment are essential to normal development and well-being.
- b. **All rodent housing receives environmental enrichment**, unless an exemption has been approved in writing by the IACUC. See **SOP #403 Rodent Enrichment**. Examples of environmental enrichment include nesting material for mice and wooded chew sticks for some rodent species.
- c. Any environmental changes made must be communicated to and approved by the researcher prior to implementation.
- d. **Single housing of rodents should be the exception** and must be justified based on experimental requirements or veterinary-related concerns about animal well-being.

4. Feeding and Watering

- a. Refer to **SOP #022**, entitled **Feed and Bedding**, for a list of approved diets. **NOTE: procedures below are not for animals on special diets or water.** Special diets must be described in an approved IACUC protocol (e.g., type, amount, form).
- b. Feeding
 1. Mice, rats, and gerbils will be **fed ad libitum** via wire bar lid unless otherwise specified by protocol.
 2. Hamsters are fed **ad libitum** via a J-feeder or by placing feed directly on bedding.
 3. Feed supply is checked daily and additional feed added as needed.
 4. Feed is completely replaced with new feed at the time of wire bar lid/J-feeder change every two weeks. Changing of the wire bar lid/J-feeder is recorded on the **Room Status Sheet**.
- c. Watering
 1. **Water will be given ad libitum** via water bottles unless otherwise specified by protocol.
 2. Water bottles will be checked daily and replaced as needed to ensure adequate supply is available.
 3. Water bottles, sipper tubes, and stoppers are sent to the cagewash for weekly sanitation. Changing of the bottles is recorded on the **Room Status Sheet**.

5. Health Surveillance

- a. Each housing room is assigned to an Animal Care Technician. Daily observations of the Animal Care Technician are recorded on the **Room Status Sheet** within the **Room Log Book**.
- b. Surveillance frequency:

Health surveillance will occur twice per day on weekdays at the ALZ, COM, IDRB, PSY, and SRB facilities. The initial health surveillance will occur prior to 9:30 AM and again about mid-afternoon. Weekdays, after the second pm health and environmental check, a subsequent "Final Food/Water" check is

performed and recorded on CMDC #244 prior to staff leaving. Staff conducting the "Final Food/Water" check and completion of CMDC #244 are the technician that is assigned to that animal housing room for that day.

- c. Health surveillance on weekends and holidays will occur once per day
- d. Observations and communications must be noted on the **Room Status Sheet** and the Facility Manager's **Animal Health and Environmental Concern Form**.
- e. Performance of daily observations and general health surveillance:
 - 1. Each animal/cage is observed to evaluate:
 - a. Food and water intake
 - b. Fecal output
 - c. Assurance of water supply and patency
 - d. General appearance (e.g., presence of lesions/injuries, coat condition, disposition)
- f. Refer to **SOP #006**, entitled **Animal Health and Environmental Surveillance**, for specific procedures of reporting and recording health concerns.

NOTE: If at any time there is a medical emergency (e.g., moribund, dystocia, bleeding, irretractable seizures, lethargy, dehydration, etc.), the clinical veterinarian is to be notified immediately.

6. Caging and Cage Changing

- a. A complete cage unit for mice, rats, and gerbils includes a polycarbonate cage bottom, a micro-isolator filter top, a wire bar lid, clean bedding material, water bottle and sipper tube, cage card and holder, and approved environmental enrichment device(s). Rats are housed in polycarbonate caging that is at least 7 inches high.
- b. Hamsters are housed in polycarbonate shoebox caging that is at least 6 inches high. Since hamsters are burrowing animals, extra bedding should be provided. Due to the shape of its muzzle, hamsters may have trouble reaching cage-top feeders and should be fed by way of J-feeders and/or feed may be placed directly on the bedding.
- c. **Prior to changing cages assemble all supplies and don appropriate PPE.** Push racks instead of pulling them. Hold open closures or use door stops to stabilize the door before guiding the racks through the framed opening. Disposable gown, gloves and Tyvek sleeves are required prior to handling rodents/rodent caging. Tyvek sleeves and gloves must be disinfected by saturate spraying with Oxivir Tb prior to opening rodent caging.
- d. **Disinfect work surfaces** of changing/work station with Oxivir Tb in accordance with **SOP #1129** entitled **NuAire Small Animal Cage Changing Station**.
- e. **Cages must only be opened/changed inside of a HEPA filtered laminar flow work station, when available** (e.g., cage-changing station, laminar flow clean work bench, or bio-safety cabinet; see **SOPs 1126, 1127, and 1129**).
 - 1. Cage changing will be performed once a week for all static microisolator shoebox caging when housing rodents at standard densities.
 - 2. Individually ventilated caging (IVC) is routinely changed every two weeks for most rodents, every two weeks for immune deficient mice, and as needed for rodents with special needs (e.g., diabetic that exhibit increased urine output).
 - 3. Due to scant urine production, IVC caging is routinely changed every three weeks for gerbils.
 - 4. Any cage excessively soiled will be changed as often as necessary to maintain an acceptable level of sanitation.

5. Cage changing will be noted on the **Room Status Sheet**.

- f. **Cages are changed one cage at a time** and husbandry practices planned so that the microisolator filter top is removed from the cage for the least possible amount of time and frequency.
- g. IVC caging is changed as a unit. IVC caging that is excessively soiled (e.g., rat caging, diabetic animals) may be changed weekly with bottoms and bottles changes one week and complete unit changed the second week.
- h. **Static micro-isolator filter tops, wire bar lids, environmental enrichment devices, and shelf/rack units will be changed on a rotational basis**, a minimum of every other week.
- i. Transferring rodents from soiled to clean microisolators using forceps decontaminated by soaking in Oxivir Tb is preferred. If handling neonates, pups should be cupped using a scoop previously saturate sprayed with Oxivir Tb and placed in the new cage.
- j. If transferring rodents with gloved hands that are dipped/sprayed and sleeves sprayed with Oxivir Tb, ensure a full one minute contact time before handling animals or opening cages. Gloves should be kept wet with Oxivir Tb and changed frequently and between rack-sides.
- k. **Soiled IVC caging is not to be disassembled within the cage changing station or housing room.** It is removed from the room fully intact and taken to the cage wash area for breakdown and sanitation
- l. Soiled static caging is always kept covered until bedding is properly discarded. This can be accomplished during cage changes by stacking static cages and covering the top cage with a microisolator filter top.
- m. **Disinfect work surfaces** of changing/work station with Oxivir Tb after cage changing procedures have been completed.
- n. **Caging micro-environmental conditions should be verified as adequate during daily observations** (e.g., condition of bedding, cage surfaces, observation of animal behavior, appearance). Increased cage change frequency, not anticipated as part of the protocol, exceeding minimums described in item 6d above, should be noted on the **Room Status Sheet** form **CMDC #041** and the **Animal Health and Environmental Concerns Form CMDC #077**.
- o. Cages will be moved to the new rack so that they retain the exact same cage position as they held on the original rack. When the rack is put back into place, the relative positions of all cages must not have changed.
- p. IVC equipment/accessories are changed/sanitized at the following intervals and memorialized on the **IVC AHU Maintenance Hang Tag** and **Room Status Sheet**
 - 1. Air Handler Unit (AHU) air exhaust pre-filter - every 4 weeks or as needed.
 - a. With AHU running, the exhaust pre-filter is spritzed with Oxivir Tb and carefully bagged-out.
 - b. AHU exhaust area is wiped with Oxivir Tb and a new filter installed.
 - c. Dirty exhaust filter bag is spritzed with Oxivir Tb prior to removing from the room for disposal.
 - 2. AHU rack hoses - every 6 months
 - a. Hoses are removed from the rack and immediately capped with glove being careful to not create aerosols.
 - b. Hoses are spritzed with Oxivir Tb and removed to dirty cage wash for cleaning and decontamination.
 - 3. IVC racks - every 6 months
 - 4. AHU air intake pre-filter - 6 months
 - 5. AHU- new AHUs are considered certified by the manufacturer, AHU hang tags are labeled with the date of receipt, and are serviced as needed

6. AHU HEPA filter – 3 years or as needed (based on reduced air-flow, or assessment during service)
7. AHU decontamination - as needed with Oxivir Tb or VHP
8. The user passcode for changing default settings is 10.

7. Room Duties

- a. **Daily** room duties: Upon completion of cage changing, feed and watering, and daily health surveillance, the following tasks are performed. Empty rooms do not require that “daily duties” be performed or recorded but room must be prepared prior to animals being present. Room duties described below are minimal requirements; additional duties and frequencies are at the Facility Manager’s discretion.
 1. Complete **Room Status Sheet** by recording:
 - a. Minimum and maximum room temperature and humidity (measurements out of the acceptable range are described under the **Health and Environmental Concern** column of the **Room Status sheet** and reported to the Facility Manager via the **Health and Environmental Concern Form**)
 - b. Feed and water are available
 - c. AHUs and racks are functional (for IVC caging), the following should be checked and assured daily:
 1. Presence of power supply

Emergency Note: In the event of a power failure causing the air handling units for individually ventilated caging (IVC) to cease function, murine inventories housed in IVC may be left static for up to 36 hours. Rat inventories cannot remain static in IVC and must have the filter retainer popped off and filter removed within 3 hrs.

2. No alarms present (i.e., red exclamation point alarm, yellow or red on performance indicator)
3. Correct airflow
4. Positive mode (most areas) air changes per hour 70-75, exhaust HEPA filtered air -20-33%
5. Negative mode (biocontainment, quarantine) supply air – 20%, air changes per hour 70-75
6. Cages are fully docked into vent rack.
7. Plenums are connected
8. Hoses are connected
9. Openings are capped
10. No cage condensation
- d. Caging and equipment changes performed
- e. Housekeeping duties performed
- f. Significant health or environmental concerns
- g. Humane traps are checked once daily for live mice. Since traps are checked daily do not place food/water in them as they attract insects. If mice are discovered, they are to be humanely euthanized via CO₂, the veterinarian alerted, and the trap is sent to cage wash for sanitation before reuse. Humane trap openings are located on either end of the box, and should be positioned against a wall near a corner with the trap entrance openings against the wall (as rodents move along the wall).
- h. Anesthesia units have been checked (when present):

1. Check isoflurane levels during AM & PM checks to ensure vaporizer is full
 2. Ensure scavenging canisters (e.g., F/Air) are weighed at the end of the day.
 3. Confirm additional O2 tanks are available in the room.
 - i. Time of observation and the initials of technician
 2. Sweep and Mop floor with Decon-Spore 200 Plus
 3. Assure no investigator's supplies or trash are left in the room.
 4. Check and replenish supplies (e.g., soap, paper towels) as needed.
 5. Wipe down counter/sink areas with Oxivir Tb.
 6. Assure any new arrivals are recorded on appropriate *Per Diem Sheets*.
 7. Record any additions or subtractions made by room technician or research staff as observed (e.g., weanlings, breeding pairs set up/removed, euthanasia).
- b. **Weekly** room duties include:
1. Clean and disinfect changing station and clean/replace filters in accordance with **SOP #1129** entitled ***NuAire Small Animal Cage Changing Station***.
 2. Clean and disinfect biosafety cabinets, if present, in accordance with **SOP #1127** entitled ***Biological Safety Cabinets***.
 3. Sanitize mop head
 4. Wipe down door and door-frame with Oxivir Tb
 5. Physically count animals within the room
- c. **Semi-monthly** room duties include:
1. Sanitize all room cleaning equipment (mop, broom, mop bucket, dust pan).
- d. **Monthly** room duties include:
1. Clean HVAC vents, and replace air filters as needed.
 2. Confirm diurnal light timers are accurately controlling animal room lights (HOB0), and record.
- e. **Quarterly** room duties include:
1. Ceilings, ducts, pipes, & fixtures are cleaned and sanitized with Oxivir Tb.
- f. **Semi-annually** room duties include:
1. Change-out and vaporized hydrogen peroxide (VHP) decontaminate all IVC racks and AHUs.
- g. **Annually** room duties include:
1. Change-out and VHP decontaminate all IVC racks, AHUs, changing stations, biosafety cabinets, shelving units, anesthesia quad units, and any other portable equipment.
 2. Wash walls, ceiling, ducts, pipes, and fixtures with Oxivir Tb.

Approved:

Date: