STANDARD OPERATING PROCEDURES
DIVISION OF COMPARATIVE MEDICINE
UNIVERSITY OF SOUTH FLORIDA

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TITLE: Naked Mole Rat (*Heterocephalus glaber*) 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 Naked Mole Rats

I. PURPOSE

1. To ensure the highest quality of animal health and preclude the development of spontaneous diseases or disorders, which could compromise the integrity of studies and the interpretation of results.

2. To describe the care and health assessment for the USDA covered species, the naked mole rats (NMR).

3. To ensure personnel handling NMR’s 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. GENERAL CHARACTERISTICS

1. NMR’s are hairless rats with small hairs arranged across the body which make them unable to regulate temperature effectively.

2. They have poor vision and have very large incisors used for digging which may need to be trimmed occasionally.

3. NMR’s are eusocial, like to live in colonies, and must not be separated from their colony groups or combined with other colonies, as they will become extremely aggressive. They are extremely sensitive to changes in the environment such as housing, smell, vibration and noise.
   a. Each colony is composed of 1 queen female and 1-3 dedicated males for breeding.
   b. Their life span has been reported to be up to 32 years
   c. They are insensitive to Carbon Dioxide (CO₂) unlike other rodents.

IV. PROCEDURES

1. Receipt
   a. Individual cage cards are prepared for each animal and include the following information:
      1. Investigator
2. IACUC #
3. Species
4. Sex
5. DOB (or age on arrival if unavailable)
6. Body Weight
7. Arrival Date
8. Source
9. USDA assigned animal number (e.g. Fiscal Year- N- # of animal)
b. Animal Medical Records are required for all animals, and can be maintained as either individual medical records or as colony records: (see SOP #012 entitled, Animal Medical Records)
c. Shipping crates are inspected for signs of damage and shipment accuracy.
d. Technician will notify the Facility Manager regarding any damage to crate or contents.
e. Facility Manager will notify the Fiscal and Business Specialist of damaged or rejected shipments, or orders improperly filled (i.e., weight, age, number requested).
f. Clinical Veterinarian will be immediately notified of health concerns or deaths occurring in transit.
g. Shipping containers are lightly wiped with Oxivir Tb™ or Peroxigard and taken to the housing room. Upon opening, an environmental swab is taken and pooled feces from the shipping container collected for diagnostics.
h. NMR's are very docile and rarely bite when handled.
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 Logbook. Notation in the Progress Notes will include the vendor source and a statement reflecting general appearance on arrival.

2. Husbandry
   a. Environment
      1. NMR's will be housed in the Tenciplast Aria Ventilated Cabinet (BIO-C36). See SOP 1168.1 to set units to the following specifications:
         a. Temperature Set Point (87°F or 30.6°C): Temperature is maintained between 85-89°F (29.4-31.7°C). Temperature is not allowed to drop below 84°F (28.9°C) because this can cause body necrosis/death to the animals. Temperature can be maintained consistently in the environmental cage system.
         b. Humidity Set Point (50%): Humidity set point will need to be between 40-60% relative humidity which is essential to the well-being and reproduction of this species.
         c. Light cycle: 12:12 or as per protocol
      2. Noise/Vibrations: NMR's are extremely sensitive to vibrations and noises and become stressed by loud sounds or new vibrations.
      3. Open and close doors slowly and quietly so they do not slam shut.
      4. Bedding: Shredded Aspen shavings with approximately ¼ inch of bedding per cage compartment or soft pelleted paper bedding
         https://www.labsupplytx.com/products/bedding/wood/
         https://www.envigo.com/7084-pelleted-paper-contact-bedding
   b. Cage Changing
      1. Caging systems are generally composed of
         a. Cage/tunnel system
b. Separate nesting chamber

c. Separate urination/defecation (toilet) chamber

2. NMR’s are known to be docile when handling and can be handled like other rats, but bite proof gloves worn under nitrile gloves may be needed.
   a. They are highly scent driven and care must be taken when using cleaning supplies or handling with gloves.

3. The Guide recommends a cage sanitation interval of at least weekly, for most species but due to their natural habitat, this will result in animal health issues and overall decline in their wellbeing.

4. They are extremely sensitive to changes in housing, smell and temperature fluctuation which can result in injury leading up to death due to fighting or pup cannibalism.

5. Limiting the amount of cage changes is in the best interest of the NMR’s health and welfare in order to mirror their natural habitat

6. This species has adapted to higher levels of gases such as ammonia (NH₃) and carbon dioxide (CO₂) which are common reasons for the increased frequency of cage changes or increased ventilation for other more common laboratory rodents, (i.e., mouse and rat).

7. Excess manipulation of the caging system will cause noise and vibration which the NMR are very sensitive to. Any abrupt disturbances can lead to a panic response.

8. The caging system, except for the toilet chamber should not be cleaned unless necessary since they depend on odors in the absence of good eyesight. (See NMR Room Sheet CMDC 271)

   c. A complete cage change can lead to increased mortality.
      1. Dirty bedding and fecal transfer are required in order to prevent detrimental effects.
      2. The following schedule should be followed to avoid adverse effects and noted on the Room Status Sheet Daily.
         a. Remove and throw away old food left inside each compartment and change gloves in between each compartment
         b. Inspect and remove fecal pellets or mold from all components including their toilet chamber
         c. Carefully inspect the bedding for animals burrowed or decreased
         d. Ensure temperature and humidity requirements are within set limits and document on Room Status Sheet
         e. Check animal health and notify veterinary staff if any issues
         f. Count and record any new births

3. The following schedule should be followed on a Weekly basis (or more as needed) and recorded on the Room Status Sheet.
   a. Dump and wipe soiled cages using hot water with a disposable towel as needed and on a rotating basis with a minimum frequency of once per 7 days for each cage in the system.
   b. The Toilet Chamber should NEVER be dumped and wiped clean on the same day as other cages in the system.
   c. Remove a sample cup of dirty bedding (without fecal pellets) from the Toilet Chamber and place it in two of the other cages in the system to maintain the scent of the colony.

4. The following schedule should be followed on a Monthly basis (or more as needed) and recorded on the Room Status Sheet.
a. Prepare a clean temporary holding cage(s) prior to performing replacement of all cages (except Toilet Chamber) in system. The number of temporary holding cages is based on the colony size.
b. Place a small amount of bedding from the Toilet Chamber into a clean holding cage(s).
c. Replace each cage (except Toilet Chamber) in the colony caging system with a sanitized (cage-washer at 180°F or higher) cage or component (clear polycarbonate tube, T piece) at least Monthly. Toilet compartment change must be delayed by one week to help the NMR reestablish the colony scent.
d. Replace the Toilet Chamber with a sanitized (cage-washer at 180°F or higher) the following week from all other cages.
e. Take used bedding material from Toilet Chamber and distribute among other cages in the system.

3. Feeding
   a. The feeding schedule (Will be laminated and posted in the room and inside of the room book).
   b. Clean vegetables and fruit thoroughly with hot water and inspect carefully before chopping into (1/2”) small pieces. If there are any rotten or moldy parts, throw the entire piece away. Carefully check the sweet potatoes, as they are a major food source for the NMRs. Potatoes could contain mold in the pest hole, which is covered by dark skin or soil.
   c. The amount of food given depends upon the size of the colony. (Each animal eats roughly four (1/2”) inch pieces of food daily).
      1. Pairs receive 4-6 small pieces of sweet potatoes.
      2. Large colonies should receive 4-6 large handfuls of cut food (excluding sweet potatoes).
   d. Food should be placed in an identified food cage compartment and not in the nest or toilet area.
   e. For colonies with pups younger than 3 months of age, it may be important to provide cereal feedings twice a week. This helps provide nutritional support to the nursing mother and pups.
   f. DO NOT PROVIDE WATER! NMR’s obtain water requirements from vegetables.
   g. Please see the following chart for feeding schedule:

<table>
<thead>
<tr>
<th>Day of the Week</th>
<th>Food (Daily)</th>
<th>Additional Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monday</td>
<td>Gerber or Pro-Nutro Cereal (mix to thick paste) Approx. 1 tablespoon paste per 2 animals</td>
<td>Make sure there are sweet potatoes in addition to the cereal.</td>
</tr>
<tr>
<td>Tuesday</td>
<td>Sweet Potatoes &amp; Raisins</td>
<td>Put food in at least 2 cage compartments if there are more than 10 animals in the colony (Applies to daily feeding)</td>
</tr>
<tr>
<td>Wednesday</td>
<td>Sweet Potatoes &amp; Celery</td>
<td></td>
</tr>
<tr>
<td>Thursday</td>
<td>Sweet Potatoes &amp; grapes</td>
<td>Peas or frozen mixed vegetables can be substituted as needed</td>
</tr>
<tr>
<td>Friday</td>
<td>Sweet Potatoes &amp; carrots</td>
<td></td>
</tr>
<tr>
<td>Saturday</td>
<td>Sweet Potatoes &amp; Apples</td>
<td></td>
</tr>
<tr>
<td>Sunday</td>
<td>Sweet Potatoes &amp; Corn on the Cob</td>
<td></td>
</tr>
</tbody>
</table>
h. Additional food items that can be used include: Apples, Fresh beans, Turnip, Carrots and Cucumbers (useful to maintain hydration of sick/injured animals).
  i. Avoid chow or supplements with high calcium level.

4. 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:
      1. On weekdays health surveillance will occur twice per day, prior to 9:30 AM, and again about mid afternoon.
      2. On weekends and holidays the initial health surveillance should take place upon the arrival of the Animal Care Technician, and again just prior to completing their assigned shift.
   c. Observations and communications must be noted on the Room Status Sheet and the Facility Manager's Animal Health and Environmental Concern Form.
   d. Performance of twice daily observations and general health surveillance
      1. Each animal/cage is observed to evaluate:
         a. Food intake
         b. Urine and fecal output
         c. Assurance of fresh vegetables and fruit are present
         d. General appearance (presence of lesions/injuries, coat condition, disposition)
   e. Emergencies must be immediately reported to the veterinarian include but are not limited to:
      1. Active bleeding
      2. Dull, lethargic, or unresponsive
      3. Ataxia, seizure, other neurologic signs
      4. Open mouth breathing, pale or blue mucous membranes
      5. Increase or excessive vocalization
      6. Limb paresis or paralysis
      7. Any evidence of trauma
   f. Colony medical records must include, at a minimum, weekly entries made by the research staff in the Progress Notes, which at least summarize the following for the colony:
      1. An impression of overall condition
      2. Food intake and voiding’s
      3. Any clinical abnormalities or complications
      4. Any treatments administered in response to observed abnormalities
      5. Any experimental procedures

Whenever health status/observations for an individual animal differ from colony animal's health status/observations, an entry specific to that animal must be made in the colony records. Refer to SOP #006 entitled, Animal Health and Environmental Surveillance for specific procedures of reporting and recording health concerns.

5. Room Duties
   a. Upon completion of cage changing, feed and daily health surveillance, the following tasks are to be performed: (Room duties described below are minimal
requirements, additional duties and frequencies are at the Facility Manager’s discretion).

1. **Daily** - 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** immediately)
   b. Feed available.
   c. Husbandry duties (e.g., caging/equipment changes/procedures performed).
   d. Housekeeping duties (e.g., room duties/procedures performed)
   e. Significant health or environmental concerns.
   f. Time of observation and the initials of technician.

2. Assure no investigator’s supplies or trash is left in the room.

3. **Check and replenish supplies** as needed.

4. Wipe down counter/sink areas with Oxivir Tb™

5. Assure any new arrivals are recorded on **Per Diem Sheet**.

b. Floors are to be **swept and mopped** Monday, Wednesday, Friday or when soiled.

c. **Weekly room duties** include:
   1. Sanitize mop head.
   2. Sanitize enrichment items.
   3. Wipe down door and doorframe with Oxivir Tb™
   4. Physically count animals within the room
   5. Review paperwork

d. **Semi-monthly room duties** include:
   1. Sanitize all room cleaning equipment (e.g., mop, mop bucket, broom, dustpan, feed pan, etc.).
   2. Check/clean HVAC vents and replace air filters as needed.

e. **Monthly room duties** include:
   1. Confirm diurnal light timers are accurately controlling animal room lights (HOBO) and record.
   2. Wash walls with Oxivir Tb™ as needed.