

ASRC  
UNIT B  
SMALL ANIMAL  
RESEARCH FACILITY

**STANDARD**  
**OPERATING**  
**PROCEDURES**

UNIVERSITY OF MISSOURI-COLUMBIA  
ANIMAL SCIENCE RESEARCH CENTER  
(Revised & adopted as of September 9, 2010)

STANDARD OPERATING PROCEDURES (SOP) – UNIT B

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### I. General Information

- A. Treatment and handling of animals will be conducted in a humane manner, thus it is the responsibility of ALL individuals working in the animal facility and conducting research in the ASRC animal facility to be familiar with the contents of this Standard Operating Procedure (SOP) manual (<http://animalsciences.missouri.edu/resources/>). Any actions that occur that are in violation of these procedures will subject the individual as well as the Principal Investigator's entire animal care program to a Unit B SOP violation. **Thus, it is important to read and be familiar with the document in Appendix I pertaining to SOP violations.**
- B. The ASRC small animal research facility was designed to accommodate small laboratory animals for use by Principal Investigators in the College of Agriculture, Food and Natural Resources. Principal Investigators from other Colleges may be assigned space on a cooperative basis for studies of nutrition, genetics, physiology, endocrinology, toxicology and related disciplines as space allows. Space in the facility will be used to house animals under the following animal husbandry conditions: conventional, germ-free, or those involved in Biosafety Level 1 or 2 research.
- C. Operation of the ASRC Small Animal Research Facility is the responsibility of all Principal Investigators and employees. All individuals using the facility must maintain cleanliness, humane treatment of animals, complete records, and proper respect for the rights of all personnel. Principal Investigators are required to provide the animals with proper housing, handling, feed and water while in the facility. The health monitoring will be done daily, including weekends and holidays.
- D. This SOP serves as a guideline for the housing and care of laboratory animals in the Small Animal Research Facility and serves the following functions:
1. Assurance that all personnel follow the same procedures.
  2. Training document for new personnel and reinforcement of procedures for established personnel.
  3. Reference source of information about standard procedures.
- E. It is the responsibility of the Facility Manager and the Unit Supervisor to oversee the daily operations of the animal facility and assure compliance with the standard operating procedures (SOPs) outlined in this document.
- F. Individual room assignments will be made on a yearly or short-term basis. In the Spring, users of the facility may apply for space to meet their laboratory animal housing needs for the following fiscal year. This application is now available on-line: [http://animalsciences.missouri.edu/resources/b\\_space\\_request.php](http://animalsciences.missouri.edu/resources/b_space_request.php)  
These applications are reviewed and assignments will be made for continuous (annual) use effective July 1-June 30 of the next year. Short-term use may be requested as needed. Principal Investigators must provide a copy (i.e., attach as a pdf to room request submission) a current Animal Use Protocol on file in the ACQA office.
- G. **Charges & Billing:**
1. Principal Investigators will receive an invoice via email monthly from the ASRC Building Administrator, Cyndi Jennings, to cover the costs of space/room rental in this facility. Charges for additional services (e.g., use of the surgery vaporizer, or some SOP violation)

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will also be placed on monthly statements. Bills should be paid in a timely manner. Questions about the charges on your invoice should be directed to Cyndi.

2. Conversion to usage of plastic cages, rather than wire hanging cages, for mice and rats necessitated implementation of a fee structure to pay for replacement cages. The fund, initiated in July 1998, helps pay for cage replacement costs and other related costs. Principal Investigators are charged on a per diem basis for all shoebox cages being used in their room. Principal Investigators and their staff are required to keep an accurate record of their daily cage inventory on the appropriate sheet posted on the outside of the animal room door.
- H. **Joint-use space** (e.g., diet-mixing room, N158; Sample prep room, N165) are available for all Principal Investigators with approved animal room space on a first-come, first-serve basis. It is expected that Principal Investigators and their staff will return all joint-use space to its original clean and neat condition as soon as they have completed their use. Joint-use rooms may be reserved up to one month prior to the time it is needed. Reservations may be made by signing up on the calendar posted on the door to the room that is needed. If space is needed on a regular basis or at a high frequency the PI is encouraged to discuss such requests with the Facility Manager. The Facility Manager will mediate any conflicts over joint-use space.
- I. **Freezers & refrigerator space** is available for all Principal Investigators with approved animal room space. This space is limited, so each Principal Investigator is assigned specific space within the facility's walk-in coolers and freezers.
1. All items placed in a cooler or freezer must be marked with: (1) contents; (2) date placed in storage; (3) the Principle Investigator's last name.
  2. It is preferred that cardboard boxes NOT be used to store items in either coolers or freezers in Unit B for prolonged periods of time since they tend to become moldy over time.
- J. **Biosecurity Issues:**
1. Personnel must follow the guidelines of not entering any animal room that has a higher biosecurity rating (i.e., B, C, and D; being highest to lowest biosecurity rating, respectively) after being in a room with a lower rating. For example, if you have mice in a "B" and "C" room, personnel should complete their work in the "B" room BEFORE entering the "C" room. Personnel must shower and undergo a complete change of clothes before returning to a room of higher biosecurity.
  2. The Facility Manager must be informed about plans to bring in equipment from other animal facilities. Such equipment MUST be disinfected prior to or immediately upon arrival. Please discuss options for disinfection with the Facility Manager and report how equipment has been disinfected.
  3. In order to reduce the risk of spreading infectious disease, it is the facility policy that animals may not be returned to the facility after leaving the building. Exceptions to this policy may be permitted only if approved by the Unit Supervisor.
  4. Individuals that work with laboratory animals elsewhere on campus should prepare a plan for how they will traffic between Unit B and these other facilities. The plan MUST be approved by the Unit Supervisor, in consultation with the Facility Manager and OAR Veterinarian. Rules regarding trafficking between animal rooms of different ratings apply even if the rooms are in different facilities. Additional restrictions on personnel trafficking and animal movement may be imposed by the Unit Supervisor when circumstances warrant.

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- K. **Security:** Access to the ASRC Small Animal Research Unit will be limited to authorized personnel only. Authorized personnel (i.e., those with Unit B security cards in their name) must accompany all visitors at all times while they are in the Unit. Visitors must report to the Unit Manager's office and sign in on the Visitor Log posted outside this office. Note that PIs do not need to seek permission to bring visitors into the Unit. We only seek to be kept informed about when unauthorized personnel are in this facility and who is responsible for them during their visit. Instructors who will need access to Unit B for instructional activities should provide the Facility Manager with the name of the course, dates students will be using the facility, and the names of TAs or other individuals who will be overseeing the students' activities during their time in the Unit. We do not want this to be an impediment to the use of this facility for teaching purposes, since this is one of the missions of the Unit.
- L. **Housing:** Room population density upper limits are as follows: 1 single-sided rodent rack for a cubicle or environmental chamber, 2 rodent racks for a small room, 4 rodent racks for a medium room and 5 rodent racks for a large room. The density may need to be reduced further if the number of animals per cage is increased or if the room air handling system is inadequate. In general, rats/mice may NOT be housed on the top shelf of the racks.
1. All animals are to be comfortably caged. Cage size should allow for the normal postural adjustments of the species involved.
  2. The Guide for the Care and Use of Laboratory Animals sets limits on the maximum number of animals that can be housed in any primary enclosure. This number varies with the size of the cage and the weight of the individual animals, and is not to be exceeded. See the SOP for specific animals for additional details or consult the "Guide".
  3. Only one species of animal is to be housed in an animal room. Animals should be segregated by sex and caged accordingly, unless experimental procedure, as described on an approved ACUC protocol, dictates otherwise.
- M. **Water:** All animals must have access to clean, potable water at all times unless approved experimental procedures dictate otherwise.
1. Water bottles are replaced at least once per week, or more often if necessary to assure access to clean potable water. Water bottles must be sanitized by running them through the cage washer. Sipper tubes and stoppers are sanitized in the cage washer or may be soaked in disinfectant solution before being reused.
  2. Principal Investigators using the automatic water system must check this system daily to assure that all animals are receiving water.
- N. **Feed:** All animals must have access to food at all times unless experimental procedures dictate otherwise and have been received ACUC approval. The food must be clean and free from contaminants.
1. Remove feed that is moldy or otherwise spoiled.
  2. Storage of diets in animal rooms must be in approved portable containers with lids.
    - a. Feed containers must be sanitized in cage washer once a month.
    - b. The date of setting up a new feed container must be documented on the feed log.
    - c. The feed milling date, located on the bottom of the feedbag, must be recorded on the lid of the feed barrel when the new feed is placed in the barrel.
    - d. Mouse, rat, hamster and rabbit feed should be used within 90 days of opening.
    - e. Guinea pig food should NOT be used if older than 90 days without vitamin C supplementation.
  3. Feed should be ordered in a manner to prevent excessive storage times.

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4. Unopened feedbags with a milling date of over 6 months must be discarded.

### II. ASRC Responsibilities to other Organizations

#### A. Institutional Animal Care and Use Committee (IACUC)

Each institution falling under the PHS or the Animal Welfare Act regulations is required to have an Institutional Animal Care and Use Committee of a prescribed composition, and to assign the IACUC specific responsibilities. The University of Missouri-Columbia Animal Care and Use Committee meets the prescribed membership requirements and is charged by the Institutional Official, the Vice Provost for Research, with a specified set of responsibilities, which include:

1. Semi-annual review of animal care and use programs;
2. Semi-annual inspection of animal care and use facilities;
3. Preparation of reports of the animal care and use program and facility evaluations;
4. Making recommendations to the Institutional Official regarding any aspect of the MU animal program, facilities or personnel training;
5. Reviewing and approving, requiring, modifications in (to secure approval), or withholding approval of proposed and ongoing protocols involving the use of animals in research;
6. Recommending suspension of activities involving animals that are not in compliance with existing standards, guidelines, and regulations;
7. Reviewing concerns involving the care and use of animals at MU.

#### B. United States Department of Agriculture (USDA)

The USDA conducts periodic inspections of animal facilities for compliance with the Animal Welfare Act rules and Regulations (Title 9 CFR). These inspections are unannounced and are conducted by a USDA veterinarian. All areas of the animal facility and all animal records must be made available for inspection upon request during normal business hours. As a rule, any deficiency pointed out by the inspector are to be corrected immediately either by corrective action or by removal of animals to an acceptable area. At the end of the inspection, the individual who accompanies the inspector will sign the inspection report and retain a copy. The accompanying individual must be sure that each cited deficiency is clearly understood so corrections can be made. Copies of inspection reports are filed in the Facility Manager's office.

#### C. The Association for Assessment and Accreditation of Laboratory Animal Care

(AAALAC International) conducts site visits of accredited facilities approximately every 3 years. The ASRC Small Animal Facility is a contract facility for any researcher holding a position at any accredited facility. This includes, the School of Medicine, Dalton Research Facility, the College of Arts and Sciences, the College of Veterinary Medicine, and the Christopher S. Bond Life Sciences Center. Any time one of these areas is inspected the ASRC animal facilities will be inspected also. These site visits are scheduled by AAALAC 1-2 months in advance and are conducted to assure that all aspects of the animal care and use program are consistent with standards in the Guide to the Care and Use of Laboratory Animals. In addition to a detailed inspection of the physical plant, site visitors review the programs of animal husbandry, veterinary care, personnel health and safety, and MUACUC activities.

#### D. The Public Health Service

The PHS requires that institutions, including MU, receiving PHS funding assure the humane care and use of animals regardless of funding source. This compliance takes the form of an Animal Welfare Assurance statement filed with, and approved by, the Office for Protection from

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Research Risks. MU's assurance includes a statement of policy to comply with appropriate laws and guidelines; a listing of facilities; descriptions of programs for animal care and use, occupational health, and training of personnel who work with animals; and a description of the IACUC, its functions and composition.

### III. Personnel Training & Responsibilities

- A. It is the responsibility of the Principle Investigator to provide adequate personnel to properly maintain his/her animals during both the work week and weekends, including holidays. Proper maintenance of animals includes: feeding, watering, cage changing, cage washing, and room sanitation.
- B. The ASRC has an **on-line training program** that is mandatory for all individuals that are new to working in this animal facility. Upon assignment to the Small Animal Research Facility (i.e., Unit B), new students and employees should report to the Research Animal Facility Manager to make arrangements for completing this training.
1. Individuals working with animals or equipment within the Small Animal Research Facility are required to complete the training prior to receiving security access to facility and before beginning work ~~unsupervised~~ in the facility.
  2. Gloves and lab coats **MUST** be worn in the animal rooms and cage wash area at all times. Eye protection should be worn while pre-washing cages.
  3. Animal care personnel are to work in their assigned area and avoid indiscriminate visits to animal rooms outside their responsibility. Individuals should try, when possible, not to return to the clean storage area or animal rooms after being in the dirty cage wash area.
  4. Eating and drinking are not permitted in animal rooms, service rooms, or hallways. Food and drinks intended for human consumption **MUST** be stored and consumed in the break room (N156) or in the Manager's office (N154).
  5. All persons in contact with animals must wash their hands thoroughly upon entering and leaving animal rooms. Individuals must be careful to practice good hygiene when working in a succession of rooms. This is for the protection of the employees as well as the animals.
  6. It is highly recommended that individuals use the bedding dump station located in the dirty cage room to minimize their exposure to allergens caused by bedding and animal hair and dander.
- C. **Bites & Injuries:** Any employee bitten by an animal or injured on the job **MUST** report immediately to their supervisor. The individual injured is encouraged to seek medical attention even for minor injuries. All injuries require the completion of accident reports within 24 hrs of the injury. Instructions and forms are available at <http://ehs.missouri.edu/work/accident-reporting.html>
- D. Persons conducting activities in the ASRC Small Animal Facility are **required** to enroll in the MU Occupational Health Program.  
The following health safety procedures are required for all employees with animal contact duties:
1. Supervisors are required to initiate the annual OHSP update form for all their employees.

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2. All employees with animal contact duties are required to complete a medical history questionnaire annually.
3. Tetanus immunization every 10 years and as recommended by physician.
4. Each employee is required to undergo training in zoonotic disease and allergy issues for the species they are working with.
5. Each employee is required to undergo training in PPE, handling and husbandry procedures for species they are working with.

(NOTE: It is the responsibility of the Principle Investigator to enroll their staff in this program. Information is available on the Environmental Health and Safety website...[http://bppm.missouri.edu/chapter7/7\\_020.html](http://bppm.missouri.edu/chapter7/7_020.html))

### IV. Procurement

#### A. Animal Procurement

The Facility Manager places all orders for animals to be housed in the Unit. Forms to request animals are available in the Unit office (N154) and on the ASRC Group Share website. Principal Investigators may NOT order animals unless:

1. They have an approved and up-to-date Animal Care and Use Protocol on file with the Facility Manager;
2. Principal Investigator has an approved space request in the Unit.
3. In addition, the Attending Veterinarian and Facility Manager must be made aware of the health status of animals before they are housed in the facility; (NOTE: Animals with pinworms, fur mites, Mouse Hepatitis Virus among other contagious diseases will NOT be allowed into the facility.)
4. Animals are from an approved source or have undergone quarantine with the OAR; (See VIII.)
5. Animals will be procured only when adequate cages and space are available.

#### B. Supply Procurement

1. Feed and bedding will be ordered and purchased by individual Principal Investigators and stored in the joint-use feed and bedding room (N188).
2. Joint-use supplies such as cleaning supplies, paper towels, trash bags, and air filters will be ordered and purchased by the Unit and stored in the feed and bedding room (N188).
3. The Facility Manager is available to assist with special orders.

#### C. Equipment Procurement

1. The initial inventory of caging and equipment in the Small Animal Facility was accumulated through purchases with start-up money and gifts from allied industries and faculty. The Unit, Principal Investigators, the Research Incentive Fund and the Food for the 21st Century Program have paid for subsequent purchases of new equipment. This included equipment needed for specific projects and replacement of equipment needing replacement due to wear and tear from long-term use.

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2. The Unit's policy is that any caging used in the Unit, regardless of its source, becomes property of the Unit and is made available for shared use, unless special arrangements have been made in advance. An example of an exception might be equipment borrowed from another facility on campus.

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The following (see list below) are sources of laboratory animals routinely used by the ASRC. Other sources are used at the Principal Investigator's discretion, provided that the Attending Veterinarian, Unit Supervisor, and Facility Manager can verify the health status of the animals. Arrangements to accommodate animals, which pose a risk to the health of other animals in the Unit, will be made with the Office of Animal Resources in conjunction with the Unit Supervisor and Facility Manager. The Principle Investigator will be responsible for all costs incurred while the animals are housed outside of the ASRC (e.g., Quarantine housing).

### Lists of Approved Animal Sources/Venders

#### Rabbits

Myrtle's Rabbitry Inc.  
4678 Bethesda Rd.  
Thompson Station, TN 37179  
(615) 790-2349  
(800) 424-9511  
FAX: (615) 794-9263

#### Rats

Sasco, Inc.  
Charles River Breeding Laboratories, Inc.  
251 Ballardale St.  
Wilmington, MA 01887  
(508) 658-6000  
(800) 228-4919  
FAX: (800) 255-8964

Harlan Sprague Dawley, Inc.  
P.O. Box 29176  
Indianapolis, IN 46229  
(317) 894-7521  
FAX: (317) 894-1840

#### Mice

Harlan Sprague Dawley, Inc.  
P.O. Box 29176  
Indianapolis, IN 46229  
(317) 894-7521  
FAX: (317) 894-1840

Jackson Laboratories, Inc.  
Bar Harbor ME 04609  
(207) 288-3371; (207) 288-5845  
(800) 422-MICE (For orders only)  
FAX: (207) 288-3398

Taconic Farms, Inc.  
273 Hover Avenue  
Germantown, NY 125 26  
(518) 537-6208 or (888) TACONIC  
FAX: (518) 537-7287

#### Guinea Pigs

Sasco, Inc  
Charles River Breeding Laboratories, Inc.  
251 Ballardale St.  
Wilmington, MA 01887  
(508) 658-6000  
(800) 228-4919  
FAX:(800) 255-8964

Harlan Sprague Dawley, Inc.  
(317) 894-7521  
FAX:(317) 894-1840

#### Hamsters

Sasco, Inc  
Charles River Breeding Laboratories, Inc.  
251 Ballardale St.  
Wilmington, MA 01887  
(508) 658-6000  
(800) 228-4919  
FAX:(800) 255-8964

Harlan Sprague Dawley, Inc.  
(317) 894-7521  
FAX:(317) 894-1840

#### Gerbils

Tumblebrook Farm Inc.  
P.O. Box 719  
West Brookfield, MA 01585  
(508) 867-2390  
FAX:(508) 867-2561

#### Chickens and Turkeys

Cuddy Farms  
Aurora, MO  
(417) 678-5021

Tyson  
Sedalia, MO  
(816) 827-8501

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### V. Animal Health Monitoring

- A. **Daily Health Checks:** The condition and welfare of all animals **MUST** be monitored daily **AND** noted on the Animal Health Observation sheet posted on the outside of the animal room door. Checking the conditions of bedding and food, availability of water, and health of the animals are to be considered essential duties of animal care personnel.
1. Animals **MUST** be observed daily, before 1:00 p.m., with the Animal Health Observation sheet filled out and signed.
    - a. Any animal showing any abnormality (e.g., grossly sick, injured, lame, circling, head tilt, loss of appetite, etc.) **MUST** be noted on the Animal Health Observation sheet; Report abnormalities even though they may seem to be of minimal significance, and regardless of whether experimentally-induced or spontaneous;
    - b. Next, the Principal Investigator or the Facility Manager **MUST** be notified;
    - c. While anyone can initiate a call to the Attending Vet, the Principal Investigator is ultimately responsible for making this determination for all animals assigned to his/her research program and ACUC protocol(s);
    - d. **Contacting the Attending Vet:** During normal business hours (8 am-5 pm, M-F) use pager number 441-4141. During evenings, weekends, and holidays use pager number 441-4198.
    - e. **Student workers that will be conducting daily health checks MUST receive specific training from the PI or other experienced full-time employee BEFORE they may sign the daily health sheet. For graduate students, evidence of this training will be demonstrated by co-signing the health sheets for a minimum of 5 working days, while undergraduates must co-sign for a minimum of 10 days.**
  2. **Dead Animals:** Any animal found dead **MUST** be recorded on the Animal Health Observation sheet regardless of whether death was experimentally-induced or spontaneous (i.e., unexplained).
    - a. Upon discovery of an unexpected dead animal, a determination should be made regarding whether the animal might be suitable for necropsy. Such a determination should be made by the Principal Investigator, Facility Manager, or Attending Vet. Necropsy of animals cannot be conducted on frozen animals.
    - b. Dead animals are to be placed in a clear plastic bag (available in rooms 126 and N165). Bagged animals are required to have a tag affixed to the bag with appropriate information filled out.
    - c. If the dead animal is to be submitted for necropsy, then follow the instructions in section VI, C: **RADIL submissions.**
    - d. If the dead animal will not be submitted for necropsy, then it should be taken to the freezer in room 126 or walk-in freezer, N192. The Facility Manager will see to it that the dead animal is disposed of properly.
- B. **BLUE CARDS:** Upon direction of the Attending Vet, an Animal Clinical Record (Blue Card) should be initiated for **EACH** sick/injured animal.
1. Cards are available outside the Facility Manager's office (N154).
  2. The Blue Card **MUST** be posted on the outside of the room door along with the Animal Health Observation sheet. The Facility Manager will remove and file all Blue Cards upon resolution of the case, regardless of outcome.
  3. The following information should be provided on a **BLUE CARD** for each animal:
    - a. Species or Strain

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- b. Animal/cage number of the ill or injured animal
  - c. ID# of animal observed to be ill or injured
  - d. Sex of the animal
  - e. Age of the animal - if the age is unknown, state whether the animal is a suckling, juvenile, or adult.
  - f. Protocol number to which the animal is assigned
  - g. Room #
  - h. Principal Investigator's name and phone #
  - i. Date and observed abnormality
4. **IMPORTANTLY:** Cage(s) with animals listed on the Blue Card(s) **MUST** be easily located/identified by the Attending Vet and Facility Manager.

C. **Escaped Animals:** Animals found outside of a cage are, if possible, to be captured and then placed in a clean cage separate from other animals. The Principal Investigator and Facility Manager should be notified immediately. Under most circumstances, such animals should be humanely euthanized and not returned to the colony. Exceptions to this policy can be made under some circumstances, but only after consultation with the Facility Manager and Unit Supervisor.

### VI. Animal Health Surveillance

A. **Overview-**The ASRC utilizes an animal health surveillance program to provide documentation of the health status of animals used by investigators.

For rabbits, poultry, and farm animal species, surveillance is limited to submission of animals, tissues, or swabs in cases involving clinical signs reported by the Principal Investigator. In rodents, the need for documenting and controlling health status are more critical. It is important to document presence or absence of disease to assure reliable experimental results. For these species, additional measures are utilized including careful selection of vendors, routine necropsy of animals that die unexpectedly, and the use of a sentinel animal program to systematically provide health information for each rodent animal room.

B. **Personnel:** The Facility Manager will work together with the Office of Animal Resources to assure compliance with the animal health program. The Manager will work with the OAR Animal Health Technician who is responsible for coordinating the use of sentinel animals, submission of animals for necropsy, and the compilation of necropsy reports.

#### C. **RADIL submissions:**

1. All submissions, including: animals (alive or dead), tissues, samples, or swabs **MUST** be accompanied by a RADIL submission form. All submissions that are not part of a specified research project **MUST** be reported to the Facility Manager and Attending Veterinarian.
2. Whenever possible, submissions should be made in the morning, as early as possible; submissions to RADIL should always be coordinated with the RADIL office, and/or the pathologist on duty.
3. For routine health surveillance activities (Sentinel Program), animals or specimens should be submitted early in the morning.
4. RADIL forms should be filled out as completely as possible.
  - a. Provide the following information; room number, source, age, and brief history.
  - b. Healthy sentinel animals should be marked as such on the form.

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- c. Indicate type of profile requested (Clinical, Basic, or Comprehensive profile).
5. The Principal Investigator is to cover the costs of samples submitted and analyzed by RADIL.

### D. **Rodent Sentinel Program** (See Section XVIII for OAR SOP on Sentinel Program)

1. The Office of Animal Resources (OAR) is responsible for obtaining the animals used in the sentinel program. The rodents will be supplied from a vendor and they will be previously screened for health problems. The animals will be between 1 and 2 months old. The requirement is to have two animals between 1-2 months old for every 70 cages in a room. The animals will be placed at a time determined by the Animal Health Technician. The tests will be done on a quarterly basis.
2. The sentinels will be housed in a shoebox cage(s) on the bottom shelf of one or more of the racks. Rooms that house rodents in metal hanging cages should house the sentinel animals in a shoebox cage.

The following procedure will be used for sampling dirty bedding. Periodically all animal care programs will be observed to assure the proper procedure is being followed.

#### **Sentinel Cage Bedding Change Procedure:**

1. At each bedding change, select a dirty cage from a rack shelf to be sampled. This cage will become the new sentinel cage.
2. Into this dirty cage, add a single scoop of dirty bedding from each cage on the shelf(ves) scheduled for sampling that week. A scoop is the amount of bedding that fits in a one (1) oz. disposable plastic cup. **\*\*\*A different cup should be used for each sentinel cage to prevent cross-contamination.**
3. Example: a rack with one sentinel cage on each side of the rack. Use one cup to sample cages on side A and a different cup to sample cages on side B.
4. After all scheduled cages have been sampled; adjust the volume of bedding by adding additional dirty bedding, removing excess dirty bedding, or by adding no more than two (2) scoops of clean bedding.
5. Transfer the sentinel mice from their old cage to the new one.
6. Track which shelves have been sampled, using the sentinel cage card. Record any deviations from the scheduled cage sampling on the cage card.

### E. **Rodent Vendors & Animal Transfers**

All rodents that are brought into the facility must originate from a vendor or source approved by the Attending Veterinarian. Approval to move animals into the facility will be based on review of health status by the Attending Veterinarian, as well as approval by the Unit Supervisor. Rodents coming into the facility from an off-campus source other than an approved commercial vendor routinely will be quarantined at the Office of Animal Resources (OAR) Medical Center facility before they can be housed at the ASRC Unit B facility. Before animals from other facilities on campus can be moved into this facility, the Principal Investigator **MUST** obtain approval from the Attending Veterinarian (via an MU OAR transfer form) and from the Unit Supervisor.

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### VII. General Animal Care Procedures

- A. **General:** The purpose of this SOP is to describe general procedures applicable to the routine husbandry of animals housed at the ASRC Animal Facility. The animal care staff should become familiar with this SOP.
- B. **Animal Care:** All animals housed in the facility are to be comfortably housed in safe, secure quarters and have adequate quantities of wholesome feed and clean, fresh water. Sanitation of animal caging and rooms is of primary importance in preserving animal health and comfort. The sanitation measures prescribed will be rigorously followed. The animal care staff observations of animal health and housing conditions are of critical importance and receive a high management priority. Animals will be handled only by prescribed methods, which minimize stress and discomfort, and maximize safety to both the animals and the handler.
- C. **Cage card information:** All cages housing animals MUST display a cage card containing the following information: (1) species; (2) number of animals in cage; (3) Principal Investigator's name; (4) source from which animals were obtained; (5) date of arrival or birth.
- D. **Room card information:** All animal rooms will display a room card on the door. A room card should be present for each research project. This card is to identify the Principal Investigator, protocol number, species of animal, light cycle of room, temperature range, responsible individuals and their phone numbers both at work and at home.
- E. **Shipments:** All shipments for the Small Animal Research Unit are to be checked in with the ASRC Animal Facilities Manager upon arrival. Refer to the section on "Procurement" ( ) for more details regarding how animals and supplies are to be ordered.
- F. **Radioactive & Bio-hazardous Materials:** Use of radioactive or bio-hazardous materials in this facility must be approved by Unit Supervisor and requires each Principal Investigator to prepare a detailed plan (SOP) outlining the proper handling and containment of the materials being used.
  - 1. Environmental Health and Safety authorization is required as is an on-site inspection PRIOR to the introduction of radioactive or bio-hazardous materials into this facility.
  - 2. All personnel that will be entering the Principal Investigator's animal rooms must be familiar with the safety and containment procedures as outlined in their EHS authorization and lab SOP.
  - 3. Rooms in which these materials are used must be properly identified as per EHS guidelines.

### VIII. Procedures for Incoming Animals

- A. The Facility Manager is responsible for placing all animal orders associated with research projects housing animals in this facility.
  - 1. Commercial suppliers with known clean health status may supply animals to investigators without requiring quarantine before they enter Unit B.
  - 2. Before animals from other facilities on campus can be moved into this facility, the Principal Investigator MUST obtain approval from the Attending Veterinarian (via an MU OAR transfer form) and from the Unit Supervisor.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

3. Animals from other, non-approved, sources (e.g., Investigators at other universities) must be approved by both the Attending Veterinarian and the Unit Supervisor. Principal investigators are required to provide the Facility Manager with a copy of the satisfactory health status report BEFORE these animals are brought into this facility.
- B. Health monitoring reports for all rodents from non-approved sources are reviewed by the Attending Veterinarian. Depending on the information provided in these reports, these animals may be quarantined and diagnostic samples submitted as needed until satisfactory animal health reports are obtained. All costs associated with quarantine and screening will be the responsibility of the Principal Investigator.
- C. Rodent shipping boxes MUST be checked by the animal care personnel for damage and dead or sick animals as soon as possible, but no longer than 8 hours after delivery to the facility. Dead and sick animals MUST be removed from shipping boxes and the Facility Manager notified so that the supplier and shipping firm may be notified.
1. Ordinarily, rodents should be housed as soon as possible upon arrival (i.e., within a few hours).
  2. If this is not possible, then they should be placed in the room to which they are assigned while still in their shipping containers.
  3. Water should be provided to the animals in the shipping box. This can be accomplished by placing a clean water bottle on the top and pushing the sipper tube through the screen.
  4. Animals MUST be relocated from their shipping box into clean cages within 24 hours of arrival.
- D. **Acclimation/Conditioning:** It is recommended animals should be allowed a minimum period of one week for conditioning and health status evaluation before assignment to experiments. Incoming animals suspected of being sick should be isolated from other animals. The Facility Manager is to be notified of this condition and the Attending Veterinarian contacted.

### IX. Cage Changing Procedure for Contact Bedding

- A. Bedding in direct contact with animals is to be changed a MINIMUM of once a week. Heavily soiled bedding should be changed more often. Check with the Facility Manager for recommendations for how to resolve frequent occurrences of heavily soiled bedding.
- B. The following procedure is recommended for changing cages with contact bedding:
1. Count number of cages that need to be changed.
  2. Take desired number of cages from the clean storage area and place them on a clean cart.
  3. Fill cages with bedding of a sufficient quantity to insure comfort of the animals and provide adequate absorbance of animal waste. As a guide, it is recommended to cover the entire cage bottom to a minimum depth of 1 cm for cob-type bedding and 2 cm for aspen shavings.
  4. Place an enrichment device in the cage of all singly-housed animals.
  5. Transfer cage cards, food hoppers, water bottles, and animals into clean cage, one at a time and replace cage at the same location on the rack.
  6. Wire lids should be changed every two weeks.
  7. Micro-isolator tops, if used, should be changed when it becomes visibly dirty/dusty.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

8. Racks should be changed every two weeks. If extra racks are not available, the rack in use should be wiped down with a sponge soaked in disinfectant solution while changing cages.
9. **Record what was changed on door sheet.**

- C. Remove dirty cages and equipment to dirty cage wash area. Do NOT leave dirty cages in hallways for more than a short time (e.g., a few hours) or in the animal rooms for more than a day (i.e., 24 hrs).
- D. Dump soiled bedding in a bag-lined dumpster/garbage can. Students and staff are encouraged to use the DUMP STATION in order to reduce the spread of dust, allergens, and infectious agents from the dirty bedding. Any food, bedding, or excreta remaining in the cage MUST be thoroughly scraped free. When finished or when garbage liner is two-thirds to three-quarters full, then close liner tightly and dispose of it in the dumpster located outside the loading dock. Spray and wipe down the DUMP STATION with diluted disinfectant after use. Do NOT forget to turn OFF the dump stations when you are finished.
- E. Pre-wash ALL cages and equipment. Personnel MUST wear eye protections during this process. Be sure to remove all tape and cage cards.
- F. Stack equipment (e.g., cages, lids) GENTLY and neatly in dirty cage wash area along the wall nearest the cage washer.
- G. Sweep floor around area.

### X. Cage Changing Procedure for Non-Contact Bedding

- A. The following procedure is recommended for changing the caging used with indirect bedding:
  1. Count the number of pans that need to be changed.
  2. Take the desired number of pans from the clean storage area and place them on a clean cart.
  3. Fill pan with bedding of a sufficient quantity to provide adequate absorbance of animal waste. Excessive amounts of bedding in pans are unnecessary and costly.  
(NOTE: **Poultry pans** typically are lined only with brown paper, purchased by the Principal Investigator, and located in room N164. Tear paper off of roll using the pan in front as a size guide. Avoid using too much paper, as this will cause the paper to crumple when inserted into the battery.)
  4. Take out the soiled pans and replace them with the clean ones.
  5. Litter and trays under suspended wire caging are to be changed twice a week.
  6. The entire rack and cages are to be changed every two weeks.
  7. Food hoppers should be changed every two weeks.
  8. **Record what was changed on the door sheet.**
- B. Transport soiled pans to the dirty cage area.
  1. Dump bedding in a bag-lined dumpster. Any food, bedding, or excreta remaining in the pan must be thoroughly scraped free. Seal liner and remove to the dumpster located outside the dock.
  2. Pre-wash all pans and equipment **WHILE WEARING EYE PROTECTION**. Use nylon brushes, scrapper, and razor blades to remove all items that do not readily come off with the hose.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

3. Load pre-washed pans onto racks used to wash pans in cage washer or if no appropriate racks are available, then stack pre-washed pans neatly in cage washing area.

### XI. Room Maintenance

#### A. Floor cleaning schedule

1. All floors in animal rooms are to be swept daily with brooms provided.
2. The entire floor in the animal room is to be mopped with a disinfectant solution whenever cages are changed. Make sure the proper dilution is used (e.g., TOO MUCH bleach is bad for the floor finish). Pour mop water down the drain on the floor once a week to keep drain trap from becoming dry, which allows roaches easy access to the room.
3. A clean mop bucket must be used for this. These are available in the clean cage room.
4. Mop buckets are to be returned in a clean condition (i.e., emptied and rinsed out!) in the cage wash area. Mops are to be stored off the floor in the animal room.

#### B. Sink cleaning schedule

1. Sink and counter area are to be cleaned bi-weekly with disinfectant solution and kept free of clutter.
2. Scale remover should be used as needed to remove scale around faucet.
3. Hand soap and paper towels should be placed in dispensers as needed.

#### C. Walls, Ceilings, etc. cleaning schedule

1. Walls, ceilings, doors, light fixtures, etc., will be sanitized once a month with a disinfectant solution.
2. A hand sprayer is available for spraying these areas.
3. When a project is completed, animals are to be removed from the room, the room is to be thoroughly sanitized with disinfectant leaving a clean room for the next project.
4. Rooms that have continuously running projects should thoroughly clean walls, ceiling, fixtures, etc. at least twice a year. The Facilities Manager will periodically designate a time for this during the year.

#### D. Any concerns about the environment (e.g., temperature, humidity, leaking faucet, etc.) in any rooms within the Unit should be reported to the Facility Manager in a timely manner.

1. Timers are set for each animal room to provide automatic light control as per the PI's instructions. Users are to contact the Facility Manager for making any change in the setting of light timers or room temperature.
2. Burned out light bulbs should be reported to the Facility Manager who will arrange to have them replaced by MU maintenance staff.

#### E. **Air filters** should be changed by the PI's animal care staff once a month or more often if deemed necessary by the Facility Manager.

### XII. Cage Pre-Wash Procedure

#### A. Pre-washing equipment

1. DO NOT bang cages on the side of the gray bins. Use scrapers to remove bedding from dirty cages.
2. All equipment needs to be thoroughly pre-washed before being run through the cage washer. The cage washer is built to sanitize but not remove gross soil and dirt. All tape must be removed from equipment during pre-washing procedure.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

3. There will be a charge for any equipment left in dirty cage wash area for over 24 48-hours without being pre-washed. (Cages and pans @ \$5 each; racks, @ \$25). Do not wash large particles of bedding or feathers into the drain of the pre-wash area. This will clog drain and back up onto the floor.

### B. Protective clothing

1. Eye protection, gloves, lab coat, and appropriate shoes (i.e., no sandals, flip-flops, or open-toed shoes) MUST be worn during cleaning activities.
2. Lab coats are available in the break room (N156) and supply storage room (N188).

### C. Sanitization of Equipment

(NOTE: **The cage washer is to be operated by ASRC personnel only unless prior arrangements have been made and training has been provided.**)

1. Loading cages & racks
  - a. Shoebox style cages need to be loaded on cage washing rack so they are positioned for maximum exposure to spray from the water jets in the cage washer.
  - b. Care must be taken to center wash racks in the middle of the cage washer. If anything hits the jet-rack of the cage washer it will throw the unit off track.
  - c. Wire caging need not be removed from the rack for washing; however, pans must be removed from rack.
  - d. Rack-mounted automatic watering systems must be capped prior to sanitation procedures.
2. Cage accessories, bottles, sipper tubes, food containers, etc.,
  - a. All cage accessories must be washed in a manner that prevents small items from falling on floor of cage washer.
  - b. The baskets used for washing water bottles can hold small items.
  - c. Carts and dumpsters should be washed daily.
3. Manual Sanitization
  - a. Caging systems, which cannot be washed in the cage washer, must be manually sanitized.
  - b. Scale from urine and water must be removed with scale remover.
  - c. Equipment should be cleaned with disinfectant and hot water.
4. Use of cage washer
  - a. Turn power switch to on position.
  - b. After holding tank reaches 180°F it may be used for washing equipment.
  - c. When loading equipment makes sure it is centered in middle of cage washer floor.
  - d. Shut doors and push start button.
  - e. Equipment should be taken out on the clean side of cage wash area.
5. Maintenance of cage washer
  - a. Clean loose parts from under washer floor to avoid clogging pump.
  - b. Grease all fittings on a weekly basis.
  - c. Clean jets by poking a wire through them as needed.
  - d. Problems should be reported immediately to the Manager or Maintenance Technician.
  - e. Run temp-tape in one load a week after cage washer has reached recommended temperature (180°F).

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

6. Cage Storage
  - a. Clean cages, racks, and accessory equipment are to be stored in the clean cage area.
  - b. Never store dirty cages, accessories, or room cleaning equipment in the clean cage storage room.
  - c. Do not enter clean cage room with a cart from dirty cage room or animal room.
7. Use of sterilizer (autoclave)
  - a. Do not used without proper instruction
  - b. Follow instruction posted on wall
  - c. Do not leave equipment in sterilizer over night or for extended amount of time. This will cause damage to cages and sterilizer.

### XIII. Environmental Enrichment

#### Rodents

Currently, the majority of rodents at the ASRC are housed on contact bedding, which allows for species specific burrowing and nesting behavior. Singly housed rodents MUST be provided with at least one enrichment device, options include: "Nestlets", paper towel and toilet paper tubes, wood blocks, PVC pipes, Nylabones, plastic chains, and balls. In some cases, these items, as appropriate to each species, are also placed with group-housed animals, in particular male mice, in order to decrease aggression. Caretakers and other staff that work with rodents on a regular basis should monitor the animals' use of the enrichment devices. If animals are not using certain devices these should be changed to a different type of enrichment. Caution should be exercised when using enrichment items that cannot be thoroughly disinfected. Such items should either be discarded upon changing the bedding or at least transferred into the clean cage housing the same animals that had access to the item prior to the cage change.

- Any group or singly housed animals that show signs of self trauma (including chewing at or removing surgical implants or closures) or aggression behavior toward cage mates should be provided with new or more types of enrichment devices in an attempt to alleviate these destructive behaviors.
- Enrichment devices can be changed once a week when cages are cleaned or as needed.
- For nesting purposes, Nestlets are placed in mouse cages of bedding made of recycled fiber products, which cannot be shredded. Used Nestlets can be transferred to new cages in order to have the animals' scent established, particularly important in cages of group-housed male mice or nursing females with litters. Personnel may need to spend considerable time pre-cleaning wire cage tops when using Nestlets.

#### Rabbits

Enrichment is considered important for rabbits for the same reasons as other species. Enrichment helps prevent boredom, reduces behavioral vices and prevents aggression in social groupings. Enrichment also helps reduce adverse responses and stress in response to human contact. In natural environments, rabbits frequently graze and gnaw on items; as such simulating this type of activity is the basis for the environmental enrichment program.

Rabbits can be given alfalfa cubes and mineral blocks as a nutritional form of enrichment. Toys such as stainless steel rattles on chains or bells suspended on a chain within the cage will also be used for enrichment. Radios may also be used in rabbit rooms if the PI has put this in the protocol.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### XIV. Title: Euthanasia and Anesthesia

- A. Animal termination will be carried out in a humane manner by the principal investigator or others designated by the principal investigator.
  1. CO<sub>2</sub> chambers located in Room 126 and N165 are available for euthanasia of animals. Parvo positive animals should be euthanized in Room 126 only. (Note: After euthanasia is performed, death should be assured by bilateral pneumothorax, aortic transection, cervical dislocation, or some other physical means, as appropriate.)
  2. Contact the Facility Manager for instruction for proper operation of the CO<sub>2</sub> chamber.
  3. Euthanized animals are to be placed in a plastic bag and placed in the freezer in Room 126 or walk-in freezer N192. Transgenic and treated animals should be placed in the red biohazard container. Non-treated animals that are suitable for raptor food can be placed in the white Rubbermaid container.
  
- B. The University of Missouri is obligated to follow the NIH guidelines for the care and use of animals and the recommendations of AAALAC. In essence this requires that anesthetics, analgesics, and tranquilizers be used whenever experimentation would otherwise cause pain, discomfort, or distress to laboratory animals. The use of these three classes of drugs must be in accordance with the currently accepted veterinary medical practice and must produce in the animal a high level of anesthesia, analgesia, or tranquilization consistent with the protocol or design of the experiment.
  
- C. Staff veterinarians in the OAR are available to assist in the selection of anesthetic, analgesic, and tranquilizing agents and to assist in the development of protocols for the use of these drugs in experimentation.
  
- D. A good source of information is the Formulary for Laboratory Animals, C. Terrance Hawk and Steven L. Lear, 1995, Ames, Iowa, ISU Press.

## **XV. Summary of Animal Care and Cleaning Schedules (for posting)**

### **A. Daily**

1. Check condition and welfare of animals. Fill out Animal Health observation sheet.
2. Record temperature and humidity.
3. Check all cages for adequate feed.
4. Check watering system to insure animals are receiving water. Depress valve to see if water is being delivered. Water bottles should be clean and contain sufficient potable water.
5. Sweep floor of all debris.
6. Clean sink and counter.
7. Fill out Door Sheet for cleaning completed that day.

### **B. Twice a Week**

1. Change direct contact bedding cages on high density cages (litters).
2. Change pan liners.

### **C. Once a Week**

1. Change and wash water bottles.
2. Change direct contact cages.
3. Empty trashcan.
4. Mop floors with disinfectant.
5. Take dirty cages, lids, old feed, and accessories to the dirty side of the cage wash for processing.

### **D. Twice a Month**

1. Wash racks.
2. Wash hanging wire cages.
3. Change and wash wire-bar cage lid.

### **E. Once a Month**

1. Change air filter.
2. Sanitize food storage containers.
3. Clean walls, ceiling, light fixtures, etc.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### Appendix I: Guidelines for Handling S.O.P. violations in the ASRC

#### Introduction:

Animal facilities in the ASRC are operated independently under faculty supervision. We strive to provide high quality facilities for housing research and teaching animals. In order to maintain a high quality animal care program it is essential that users follow our Standard Operational Procedures (SOP). Individuals who fail to follow the SOPs place at risk the health and well-being of workers and animals, as well as the biosecurity of the facility. Consequences for violating the SOPs may include charges for Unit support staff to provide unmet animal husbandry needs and loss of privilege to use the facility. The following guidelines will be used for handling violations of SOPs.

In an effort to acknowledge that not all failures to follow SOPs are the same, we have established three levels of violations: “A, B and C” separately within the ASRC small and large animal units.

#### ASRC Unit B (Small Animal Facilities)

##### I. Levels of Violations-

**A. Level “A” violations** are generally considered minor violations in procedure that do not significantly impact the health and well-being of animals or personnel.

Examples of such violations include:

1. Failure to sign the health check sheet on the door by 1:00 pm;
2. Failure to record the milling date on the exterior of the feed container;
3. Failure to change racks or cage lids according to the schedule required by the SOP;
4. Not filling out an Animal Health “Blue card” despite doing what is needed to assure that appropriate care has been provided to a sick animal;
5. Not recording cage changing on Daily Activity Sheet

**B. Level “B” violations** are generally considered to have the potential to put the health and well-being of animals or personnel at some increased risk or may compromise the biosecurity of the facility.

Examples of such violations include:

1. Failure to conduct a daily health check;
2. Use of diets beyond the six months after the milling date on the bag;
3. Not changing cages or trays as scheduled;
4. Trafficking into the “clean” room after being in the dirty room; wearing gloves in the clean equipment storage area/room;
5. Leaving dirty cages in the hallways after leaving the facility;
6. Failure to wear a lab coat or some protective outer clothing during any work that involves direct contact with animals or waste-contaminated products;
7. Failure to use the footbaths upon leaving the Dirty room area;
8. Failure to clean a joint-use space following surgery, sample collection or animal kill;
9. Propping doors open during any activity that is likely to generate aerosols of waste contaminated products (e.g., bedding, hair, feathers);
10. Not following the SOP for sentinel animals;
11. Bringing animals into the facility without informing the Unit B manager beforehand

**C. Level “C” violations** are the most serious and are considered those departures from SOPs or ACUC protocols that may clearly result in adverse effects on the health and well-being of animals or personnel in the facility or clearly compromise the biosecurity of the facility.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

Examples of such violations include:

1. Failure to adequately monitor animals following surgery or other invasive procedure;
2. Failure to provide appropriate veterinary care for a sick animal;
3. Failure to provide animals with adequate food and water such that clinical signs of distress are noted, unless such restriction is part of the approved ACUC protocol;
4. Failure to euthanize an animal that is clearly in pain and distress, unless specific exceptions have been described in the ACUC protocol;
5. Bringing an animal into the Unit from a non-approved source without going through a standard OAR quarantine;
6. Bringing animals into the facility without an approved ACUC protocol;
7. Bringing animal into the facility from another location (i.e., animal room) on campus without prior approval of the Attending Veterinarian.

### II. Appeals-

- A. If a PI wishes to appeal a specific violation, then they should do so in writing to the Unit Supervisor or the Unit Operations Committee within 7 days of the violation;
- B. If the Unit Supervisor was directly involved in the incident, then the Operations Committee shall conduct the investigation and appoint an interim chair to oversee the appeals process;
- C. Depending on the nature of the violation, an appeal may require nothing more than interviewing the parties involved to initiating an ACUC investigation into the incident.

### III. Administration of Violations-

- A. Violations can only be given out by the Unit Supervisor or ASRC Coordinator.
- B. Personnel responsible for level A violations will be given a verbal warning initially, followed by a written warning for a second violation with e-mail to the PI and Unit Supervisor.
- C. All level B violations will result in a written warning with PIs and the Unit supervisor receiving copies of all written warnings;
- D. All level C violations must be reported to the ACQA office, the ASRC Coordinator, the Unit Supervisor and the PI as soon as possible.

### IV. Probation

- A. Anyone who receives three or more written warnings for “Level A” violations or two or more “Level B” violations will be placed on probation.
- B. “Level C” violations will be handled on a case-by-case basis by a committee consisting of the appropriate Unit Supervisor, the ASRC Coordinator and the Attending Veterinarian, unless one of those individuals has a conflict-of-interest, in which case that person will be replaced by the Animal Sciences Unit Leader.
- C. Probation will last for **NO LESS THAN A MONTH**; probation means that the individual on probation will not have any privileges associated with working with animals in the Unit, unless directly supervised by someone else qualified & currently authorized to work with the animals.
  1. After the second week of probation the employee may conduct all of their work without supervision, if it is conducted during standard working hours (e.g., 8-5 pm, M-F only).
  2. The probationary period will end at the discretion of the Unit Supervisor after receiving a written request by the PI. A successful petition will clearly describe the additional training that the individual received to assure that every effort has been made to reduce the likelihood that further violations will occur.
  3. Additional level A or B violations by someone who is on probation will extend their probation and may result in the loss of space in the Unit for the Principal Investigator.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### **IV. Service charges:**

- A. PI's will be charged for Unit support staff providing unmet animal husbandry needs (e.g., Health checks; feeding and watering animals).
- B. Charges will be based on time and a half rate for hourly salary for Facility Manager with a minimum billing increment of 30 min.
- C. PI will be informed in writing (e.g., via email) of these charges within 7 days of the incident in question.
- D. These service charges will be added to the regularly scheduled charges for Unit space that go out each quarter of the fiscal year.

### **V. Payment of Unit Fees:**

- A. Unit users who do not pay their Unit bills, risk losing their assigned space.
- B. Unit users will be given 30 days notice prior to revocation of their assigned space for non-payment. In this time, PIs should either make payment in full or make arrangements for relocating their animal.

### **VI. Loss of Assigned Space:**

**PIs may lose their assigned space in the Unit for non-payment as described above or for repeated SOP violations by his/her staff/students.**

- A. A PI whose research program has a track record of multiple individuals being placed on probation or that has received two or more level C violations within a 12 month period will be subject to revocation of their assigned space in the Unit for the remainder of their assignment and will not be assigned space in the Unit for a period of no less than a year.
- B. Decisions regarding revocation of assigned space for non-compliance to SOPs will be made by the Unit supervisor after consultation with the Unit Operations committee;
- C. If a PI wishes to **appeal** a space revocation decision, then they should do so in writing to the ASRC Unit Supervisors Committee within 30 days of the revocation decision.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### Appendix II; OAR Rodent Sentinel Program

Revised 1/04/08

**Policy:** It is OAR policy to meet all federal, state, and local regulations and guidelines, and comply with all institutional policies and procedures as they apply to the performance of rodent health monitoring. Furthermore, it is OAR policy to have one rodent sentinel monitoring program that is standardized across all divisions on the MU campus.

**Applicable:** All faculty, staff, students and employees of the University of Missouri utilizing rodents for research and teaching.

**Purpose:** The purpose of this policy is to ensure that all research facilities on the MU campus have a standardized rodent health monitoring program designed to detect unwanted pathogens and that all personnel involved in animal care and use are adequately trained to perform the routine operations associated with the rodent sentinel program.

**Introduction:** The rodent sentinel program on the MU campus is designed to evaluate the health status of the rodent colonies on campus. Results from the sentinel program are used to determine room classification, to assist investigators in determining variables that may affect their research, and to provide health reports prior to shipment of animals from the University of Missouri to other institutions.

#### **Procedures:**

##### **1. Procurement:**

- Source: Charles River Laboratories
- Specifications:
  - Mice: 3-4 wk old CD-1 females (unless males are requested by PI)
  - Rats: 4 wk old Sprague-Dawley females (unless males are requested by PI).

Order animals to arrive at least 2 weeks before the sentinels are due to be placed in the animal rooms.

##### **2. Placement:**

- Before the sentinel placement, submit two mice and two rats from each new sentinel shipment to RADIL for comprehensive necropsies, microbiology and *Helicobacter* fecal PCR (mice only). House the remaining animals in a barrier room at LSC pending results from the necropsies.
- If the sentinel animals are declared free from infectious disease, contact facility supervisors to arrange placement of sentinels in animal rooms.
- Deliver sentinels to the facilities early in the day so that they can be placed in the animal rooms promptly.
- Sentinels will be supplied to each animal room and housed on the bottom shelf of the racks in the same type of cage as is on the rack. The standard stocking rate for sentinels will be one cage of two sentinel animals for approximately every 60 - 70 cages in each animal room. Rooms with fewer than 60 cages will have 1 cage of sentinels in the room.
- Sentinel cage cards must be filled out completely, indicating the facility, room, and rack that the sentinel cage is located on and the Principal Investigator(s) whose animals are being monitored.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

- In rooms housing immunocompromised animals, immunocompetent animals will be placed in the room as sentinels but only after permission from the investigator has been obtained. Many investigators may not want to risk the possibility of their animals becoming infected with a commensal organism from the sentinels. In these instances, any sick research animals should be submitted to RADIL for an immunocompromised OAR diagnostic necropsy profile.
- Rooms with animals that have been treated with or exposed to biohazardous agents will not be included in the normal sentinel program, unless the Principal Investigator provides sufficient information to the Assistant Director- OAR indicating the sentinel animals will pose no health risk to OAR personnel should the sentinels become infected/contaminated with the agent.
- Normally sentinels remain in the animal rooms for 8-10 weeks before being submitted to RADIL.

### 3. Sentinel Cage Bedding Change Procedure:

- At each bedding change, select a dirty cage from a rack shelf to be sampled. This cage will become the new sentinel cage.
- Into this dirty cage, add a single scoop of dirty bedding from each cage on the shelf(ves) scheduled for sampling that week. A scoop is the amount of bedding that fits in a one (1) oz. disposable plastic cup.
- After all scheduled cages have been sampled; adjust the volume of bedding by adding additional dirty bedding, removing excess dirty bedding, or by adding no more than two (2) scoops of clean bedding.
- Transfer the sentinel mice from their old cage to the new one.
- Track which shelves have been sampled, using the sentinel cage card. Record any deviations from the scheduled cage sampling on the cage card.

### 4. Submission Schedule

Sentinel monitoring is performed on a quarterly basis for all animal rooms.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### Appendix III.

### OAR Animal Room Classification System

#### Standard Operating Procedures

Revised May 30, 2007

**Policy:** It is OAR policy to meet or exceed all federal, state, and local regulations and guidelines and comply with all institutional policies and procedures as they apply to the performance of rodent health monitoring programs. Furthermore, it is OAR policy to have one unified animal room classification system that is standardized across all divisions on the MU campus.

**Applicable:** All faculty, staff, students and employees of the University of Missouri utilizing MU animal facilities to house their research and teaching animals.

**Purpose:** The purpose of this policy is to ensure that all research facilities on the MU campus have a standardized animal room classification system designed to prevent accidental transfer of infectious agents from one room to another. All personnel involved in animal care and use must understand the animal room classification system and follow the predetermined room entry order.

**Introduction:** A classification scheme has been established to reflect the health status of animals in each room. OAR assigns the classification to the animal rooms based on clinical evaluation and health screening information.

#### Procedures:

##### 1. Animal Room Classification System

The housing systems and procedures for entering each facility are based on a graduated scale of three categories (B, C, and D) with the highest level (B) requiring the most stringent procedures. Lower levels have less rigorous operational requirements. As you progress from B to D, procedures to enter the facilities become less rigid. Individual room designations are used to determine traffic flow within and between animal facilities. While it may be necessary for some investigators to maintain their animals at more than one level, this practice is discouraged since it increases the risk of cross-contamination. If you work in multiple facilities, or have multiple rooms, it is vital that you never work with animals from a lower-level category before working with animals from a higher-level category. Laboratories that may temporarily house rodents have the same classification as the animal room from which the rodents came. If you have entered a laboratory that occasionally houses animals from a low-level classification, you should not enter a higher classified animal room on the same day. Likewise, shared procedure rooms should have the same level classification as the least clean animals used in that procedure room.

#### RODENTS:

##### • Level B

Barrier housed animals. Rodents must test negative for all pathogens on the RADIL comprehensive necropsy including bacteriology plus *Helicobacter* PCR. Entry into a B room is prohibited if an individual has had previous contact with animals of a lower health status within the previous 24 hrs unless s/he has showered and changed clothes.

##### • Level C

Barrier or conventionally housed rodents. These animals are negative for endo and ectoparasites and all pathogens tested on RADIL's clinical serology. They may harbor opportunistic pathogens such as *Helicobacter sp.* or *Pasteurella pneumotropica*. Entry is prohibited if an individual has had previous contact with animals of a lower health status within the previous 24 hrs unless s/he has showered and changed clothes.

##### • Level D

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

Barrier or conventionally housed rodents. These animals have tested positive for one or more of the pathogens in the RADIL clinical serology profile (MHV, MMV, MPV, Sendai, *M. pulmonis*, TMEV, and EDIM for mice and RCV, Sendai, PVM, Parvo, *M. pulmonis*, and TMEV for rats). Level D status is also given to any rodent with pinworms or furmites, rodents that are in quarantine, and rodents experimentally infected with most rodent infectious agents.

### **NON RODENTS (rabbits, cats, dogs, swine):**

#### **• Level B**

Purpose bred animals considered to be SPF. This classification is based on vendor health reports, in house testing, and/or veterinary examination. Entry into a B room is prohibited if an individual has had previous contact with animals of the same species with a lower health status within the previous 24 hrs, unless s/he has showered and changed clothes.

#### **• Level C**

Purpose bred animals which are not SPF but whose health status is not considered a threat to other animals of the same species. Entry is prohibited if an individual has had previous contact with animals of the same species with a lower health status within the previous 24 hrs, unless s/he has showered and changed clothes.

#### **• Level D**

Animals which are known to be infected with a pathogen, come from a facility which is not biosecure, or have potential for exposure to pathogens, such as dogs walked outside. Quarantined animals are included in this classification.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### Appendix IV. Species-Specific Animal Space Requirements & Handling Procedures

Minimum space recommendations for Laboratory Animals

(from the Guide for the Care and Use of Laboratory Animals, 1996)

#### MICE

##### A. Caging

Each mouse housed in a primary enclosure must be allotted a specific amount of floor area. This requirement will vary with the cage size and weight of the individual. The following information should be used as a guide to caging mice.

Wt. Range of Individual	Floor Area Required	Height Required
<10 g	6 sq. in./mouse	5 in.
10-15 g	8 sq. in./mouse	5 in.
16-25 g	12 sq. in./mouse	5 in.
>25 g	15 sq. in./mouse	5 in.

Ex. If you have a cage with inside floor dimensions of  $10^{1/2} \times 6^{1/2}$ , the floor space is determined by multiplying the length times the width.

$$10^{1/2} \times 6^{1/2} = 68 \text{ sq. in.}$$

If you are housing mice that weigh between 10-15 g, divide the floor area required for that weight group into the floor area of the cage.

$$68 \text{ sq. in. (cage)} / 8 \text{ sq. in. (space required/animal)} = 8$$

Eight mice can be housed in a cage  $10^{1/2} \times 6^{1/2} \times 5$ .

\*Remember, as animals gain weight, they may have to be redistributed in order to comply with standards.

Cages available at ASRC:

- small plastic cages - - 68 sq. in.
- large plastic cages - - 155 sq. in.
- small wire - - 66.5 sq. in.
- large wire - - 38 sq. in.

Therefore, the housing density for mice in small and large plastic cages is:

Small cage: 4 adult mice

Large cage: 10 adult mice

##### B. Environment

Light cycle: 12 hours on / 12 hours off (14 / 10 is also acceptable)

Humidity: 30-70%

Temperature: 64-79 degrees F (70-74 ideal)

##### C. Cage Cards

Each cage must be identified with a cage card. Cage cards are to be attached to the front of each cage with a cardholder. The following information is recommended to be on each card:

1. Principal Investigator's name
2. Strain
3. Protocol #
4. Number of animals in cage
5. Source

##### D. Handling

Mice may be easily transferred from cage to cage by gently lifting them by the base of their tails.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

Mice can be restrained by gently but firmly pinching the skin on the back of their necks and wrapping the tail around a finger.

### E. Breeding

1. Mice reach breeding age at approximately 35-40 days of age.
2. Females may be bred either intensively (i.e., immediately after parturition) or non-intensively. If successive mating is desired, the mated pair should be left together, as the female has a postpartum estrus. Gestation period of the mouse is ~20 days and the young are ready to be weaned at 21 days. **Polygamous mating** can also be used in reproducing mice as a means to economize the number of available males. If breeding is to be polygamous, only one male in each breeding cage should be utilized. Adult male mice will fight and may kill one another if housed together with females.

### F. Health Problems

**It is the responsibility of the animal care personnel to watch for symptoms of illness or disease.**

Common symptoms of health problems in mice include:

1. Weight loss/loss of appetite
2. Unthrifty coat
3. Inactivity
4. Diarrhea-especially in infant mice
5. Labored breathing
6. Scabby wounds or abscesses

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### G. Obtaining Biological Samples

1. Blood may be obtained from anesthetized mice by heart puncture or by infraorbital puncture using heparinized capillary tubes. Mice bled by heart puncture should be euthanized after blood collection.
2. Small amounts can be obtained by nicking the tail veins.
3. Intravenous injections may be given through tail veins.

## HAMSTERS

### A. Caging

Each hamster housed in a primary enclosure must be allotted a specific amount of floor area. The following chart should be used as a guide in caging hamsters. This requirement will vary with cage size and weight of the individual. Food pellets for hamsters should be placed on the floor of the cage and not in wire bar lid. Hamsters are covered under the Animal Welfare Act and are subject to USDA inspection.

Wt. Range of Individual	Floor Area Required	Height Required
<60 g	10 sq. in./hamster	6 in.
60-80 g	13 sq. in./hamster	6 in.
80-100 g	16 sq. in./hamster	6 in.
>100 g	19 sq. in./hamster	6 in.

Ex. If you have a cage with inside floor dimensions of 17 X 8 <sup>1/2</sup>, the floor space is determined by multiplying the length times the width.

$$17 \times 8^{1/2} = 144.5 \text{ sq. in.}$$

If you wish to house hamsters weighing over 100 g, divide the floor area required for that weight group into the floor area of the cage.

$$144.5 \text{ (cage)}/19 \text{ (space required/animal)} = 7$$

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

Seven hamsters can be housed in a cage 17 X 8 <sup>1/2</sup> X 6 if they weigh >100 g.

\*Remember, as animals gain weight, they may have to be re-distributed in order to comply with standards. Cage changing frequency may need to be readjusted for animals housed with large numbers.

### A. Environment

Light cycle: 12 hours on / 12 hours off (14 / 10 is also acceptable)

Humidity: 30-70%

Temperature: 64-79 degrees F (70-74 ideal)

### B. Cage Cards

Each cage must be identified with a cage card. Cage cards are to be attached to the front of each cage with a cardholder. The following information is recommended to be on each card:

1. Principal Investigator's name
2. Strain
3. Protocol #
4. Number of Animals in cage
5. Source

### C. Handling

Some strains of hamsters are often difficult to handle and will bite readily. It is best not to pick up a hamster while it is sleeping. First, wake it up and make sure it sees you approaching it before picking it up by the scruff of the neck.

### D. Breeding

1. Hamsters reach breeding age at approximately 30 days.
2. Hamsters breed best when mated non-intensively and monogamously. The females are very aggressive and will often harm or kill their mates. Close observation is necessary to insure the welfare of the male. Gestation period of the hamster is 16 days. Young can be weaned at 21 days.

### E. Health Problems

**It is the responsibility for the animal care personnel to watch for symptoms of illness or disease.**

Common symptoms of disease in hamsters include:

1. Weight loss/loss of appetite
2. Unthrifty coat
3. Inactivity
4. Diarrhea-soiling of the entire hind end is an indication of "wet tail"
5. Discharge from eyes or nose
6. Bite wounds

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### F. Obtaining Biological Samples

1. Blood samples may be obtained from anesthetized hamsters by heart puncture or by the saphenous vein located on the rear leg. Hamsters should be euthanized after blood collection by heart puncture.
2. Intravenous injections may be given through the saphenous vein.

## RATS

### A. Caging

Each rat housed in a primary enclosure must be allotted a specific amount of floor area. This requirement will vary with the cage size and weight of the individual. The following information should be used as a guide to caging rats.

Wt. Range of Individual	Floor Area Required	Height Required
<100 g	17 sq. in./rat	7 in.

*(adopted with revisions on 9/2/2010)*

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

100-200 g	23 sq. in./rat	7 in.
200-300 g	29 sq. in./rat	7 in.
300-400 g	40 sq. in./rat	7 in.
400-500 g	60 sq. in./rat	7 in.
>500 g	70 sq. in./rat	7 in.

Ex. If you have a cage with inside floor dimensions of  $25 \times 9^{1/2}$ , the floor space is determined by multiplying the length times the width.

$$25^{1/2} \times 9^{1/2} = 242 \text{ sq. in.}$$

If you are housing rats that weight between 200-300 g, divide the floor area required for that weight group into the floor area of the cage.

$$242 \text{ sq. in. (cage)}/29 \text{ sq. in. (space required/animal)}= 8$$

Eight rats can be housed in a cage  $25^{1/2} \times 9^{1/2} \times 7$ .

Remember, as animals gain weight, they may have to be redistributed in order to comply with standards.

Cages available at ASRC:

- Plastic cages - - 142 sq. in.
- Slide-on plastic cages - - 200 sq. in.
- Single wire - - 66 sq. in.
- Double wire - -154 sq. in.
- Triple wire - - 242 sq. in.

### B. Handling

1. Never pick rats up by the tail. The skin on the tail is not strong enough to support the weight of the rat and it can strip off if the animal struggles. Rats are best handled if they are picked up and held firmly with the thumb and forefinger behind the front legs.
2. Rats kept in rooms where the temperature is between 65-85°F, and the humidity is under 20%, have a high incidence of ringtail. Therefore, it is important to monitor the environment of rat rooms to maintain optimum conditions.

### C. Environment

Light cycle: 12 hours on / 12 hours off

Humidity: 30-70%

Temperature: 64-79 degrees F (70-74 ideal)

D.

### E. Cage Cards

Each cage must be identified with a cage card. Cage cards are to be attached to the front of each cage with a cardholder. The following information is recommended to be on each card:

1. Principal Investigator's name
2. Strain
3. Protocol #
4. Number of Animals in cage
5. Source

### E. Breeding

1. Rats reach breeding age at approximately 70 days.
2. Rats are best bred non-intensively using one male for every five females.
3. The gestation period runs an average of 22 days and the young can be weaned at 21 days.

### F. Health Problems

It is responsibility of all animal care personnel to watch for symptoms of illness of disease. Common symptoms of diseases in rats include:

1. Weight loss/loss of appetite
2. Unthrifty coat

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3. Inactivity
4. Raised, inflamed, concentric ridges on tail
5. Respiratory infection

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### G. Obtaining Biological Samples

1. Blood may be obtained from anesthetized rats by heart puncture and should be euthanized after the procedure. Small amounts of blood may be collected from the lateral orbital vein and by nicking the tail vein.
2. Intravenous injections may be administered via the tail vein.

## GUINEA PIGS

### A. Caging

Each guinea pig housed in a primary enclosure must be allotted a specific amount of floor area. This requirement will vary with the cage size and weight of the individual. The following chart should be used as a guide in caging guinea pigs. Guinea pigs are covered under the Animal Welfare Act and are subject to USDA inspection.

Wt. Range of Individual	Floor Area Required	Height Required
Weaning - 350 g	60 sq. in./animal	7 in.
>350 g	101 sq. in. /animal	7 in

Ex. If you have a cage with inside floor dimensions of 20 X 10 1/2 X 7, the floor space is determined by multiplying the length times the width.

$$20 \times 10 \frac{1}{2} = 210 \text{ sq. in.}$$

If you are housing guinea pigs weighing above 350 g, divide the floor area required for that weight group into the floor area you have available.

$$210 (\text{your cage}) / 101 (\text{space required/animal}) = 2$$

Two guinea pigs can be housed in a cage 20 X 10 1/2 X 7.

Remember, as animals gain weight they may have to be re-distributed in order to comply with standards.

### B.Environment

- Light cycle: 12 hours on / 12 hours off (14 / 10 is also acceptable)
- Humidity: 30-70%
- Temperature: 64-79 degrees F (70-74 ideal)

### C.Cage Cards

Each cage must be identified with a cage card. Cage cards are to be attached to the front of each cage with a cardholder. The following information is recommended to be on each card:

- 1.Principal Investigator's name
2. Strain
- 3.Protocol #
4. Number of Animals in cage
5. Source

### D. Handling, Feeding and Watering

- 1.Guinea pigs should be picked up by grasping gently around the thoracic area or cupping both hands around the animal at the sides.
2. All guinea pigs require a daily supply of vitamin C. It is important to make sure the feed is not used 90 days passed the milling date.
3. Provide water in bottles or automatic watering system to prevent spilling of water and contamination with urine and feces

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

4. Feed guinea pigs with hoppers that have a large opening.

### E. Breeding

1. Guinea pigs reach breeding age at approximately 55 days.
2. Guinea pigs do best when harem bred in an enclosure that houses one male and four sows. The female has a postpartum estrus and is ready to be bred again shortly after parturition. It is best to avoid mating older females as the joints in the pelvis fuse together and presents a narrowing of the birth canal and difficult labor. The optimum age for the first mating is approximately 85 days. If the mating of the female is delayed much beyond this age there is a danger that she will not be able to deliver. Gestation period of the guinea pig is 60-72 days and the young are weaned between 7-21 days or when they reach a weight of 250 g.

### F. Health Problems

It is the responsibility of the animal care personnel to watch for symptoms of illness or disease. Common symptoms of disease in guinea pigs include:

1. Weight loss/loss of appetite
2. Unthrifty coat
3. Inactivity
4. Diarrhea

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### G. Obtaining Biological Samples

1. Blood may be obtained from jugular vein or anterior vena cava while animals are under anesthesia.
2. It is difficult to find a good site for an intravenous injection but the jugular vein is accessible as well as the penile vein in males. Generally, injections by these routes are performed in anesthetized animals.

## POULTRY

### A. Caging

Each bird housed in a primary enclosure must be allotted a specific amount of floor area. this requirement will vary with the cage size and weight of the individual. The following information should be used as a guide to caging birds.

Wt. Range of Individual	Floor Area Required	Height Required	
Pigeons ----	.8 ft <sup>2</sup>	Sufficient to	
Quail ----	.25 ft <sup>2</sup>	stand erect	
Chickens <0.25 kg	.25 ft <sup>2</sup>		
<0.5 kg	.50 ft <sup>2</sup>		0.5-1.5
kg 1 ft <sup>2</sup>		with feet on wire	
& 1.5-3.0 kg	2 ft <sup>2</sup>		
Turkeys >3.0 kg	3 ft <sup>2</sup>		

Ex. If you have a cage  $16^{1/2} \times 9^{1/2} \times 7$ , the floor space is determined by multiplying the length times the width.

$$16^{1/2} \times 9^{1/2} = 154 \text{ sq. in.}$$

If you need to house pigeons they need a floor space of 118 sq. in. so this size cage would meet the requirements.

\*Remember, as animals gain weight, they may have to be redistributed in order to comply with standards.

### B. Handling

1. Gloves and a mask should be worn at all times. Poultry are frequent carriers of Salmonella enteritidis var. typhimurium, which is capable of causing disease in humans and animals by direct infection.
2. Birds may be picked up by gently holding them around the mid-section with their wings held securely against their body.
3. The handler should take precaution not to be scratched by the animal.

### C. Health Problems

*(adopted with revisions on 9/2/2010)*

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

It is the responsibility of the animal care personnel to watch for symptoms of illness or disease. Common symptoms of disease in poultry include:

1. Weight loss/loss of appetite
2. Loss of feathers
3. Inactivity
4. Labored Breathing
6. Pecking wounds

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### D. Obtaining Biological Samples

Blood may be collected from the brachial artery.

## RABBITS

### A. Caging

Each rabbit housed in a primary enclosure must be allotted a specific amount of floor area. This requirement will vary with cage size and weight of the individual. The following chart should be used as guide in caging rabbits. Rabbits are covered under the Animal Welfare Act and are subject to USDA inspection.

Wt. Range of Individual	Floor Area Required	Height Required
<2 kg or <4.4 lbs.	1.5 ft. <sup>2</sup> or 180 sq. in./rabbit	14 in.
2-4 kg or 4.4-8.8 lbs.	3 ft. <sup>2</sup> or 432 sq. in./rabbit	14 in.
4-5.4 kg or 8.8-11.9 lbs.	4 ft. <sup>2</sup> or 576 sq. in./rabbit	14 in.
5.4 kg or 11.9 lbs.	5 ft. <sup>2</sup> or 720 sq/ in./rabbit	14 in.

Rabbit cages measuring 25"x24"x16" are 4 sq. ft.

\*Remember, as animals gain weight the divider may need to be removed from the center of the cae creating a larger cage for individual rabbits.

### B. Environment

Light cycle: 12 hours on / 12 hours off (14 / 10 is also acceptable)

Humidity: 30-70%

Temperature: 61-72 F (68 ideal)

### C. Cage Cards

Each cage must be identified with a cage card. Cage cards are to be attached to the front of each cage with a cardholder. The following information is recommended to be on each card:

1. Principal Investigator's name
2. Strain
3. Protocol #
4. Number of Animals in cage
5. Source

### D. Handling

1. Never pick a rabbit up by the ears. The rabbit can suffer damage to the cartilage of the ear or injury to the spine resulting from lack of support. Rabbits have strong muscles and a long, weak spine, which causes them to break their backs very easily
2. The safest way to handle a rabbit is to cradle it close to the body with one hand grasping the scruff of the neck and other hand supporting the hind quarters.
3. When handling any female rabbit, take special care to avoid contact with the genital region. Females can be induced into a pseudopregnancy that can invalidate research if they are handled incorrectly.

### E. Health Problems

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

It is the responsibility of the animal care personnel to watch for symptoms of illness or disease. Common symptoms of disease in rabbits include:

1. Weight loss/loss of appetite
2. Unthrifty coat
3. Inactivity
4. Diarrhea
5. Wet fur on the front paws
6. Discharge or scabby sores in the ear
7. Sores on the hind limbs (hocks)
8. Nasal discharge

Record any evidence of health problems on the animal health observation sheet and report it to the Facility Manager immediately.

### F. Obtaining Biological Samples

1. Large volumes of blood may be collected by nicking the marginal ear vein or central ear arterial or jugular venapuncture.
2. Intravenous injections may be given via the marginal ear veins.

## STANDARD OPERATING PROCEDURES (SOP) – UNIT B

### Appendix V. Monitoring of Sanitizing and Sterilizing Procedures

#### A. Monitoring Effectiveness of Equipment and Room Sanitization

This SOP describes procedures used to monitor the effectiveness of the cage washer and the steam sterilizer. The following procedures are those that evaluate the sanitation program for cages, racks, feeders, water bottles, and stoppers/sipper tubes. This program is coordinated by the OAR Animal Health Technician and administered in part by the ASRC Manager and staff.

#### B. Areas to Monitor/Frequency

Each of the following will be monitored at least semiannually, and whenever there's a change in equipment, chemicals, or procedures.

1. Interior surface of polycarbonate cages (mouse and rat)
2. Interior surface of rabbit cages
3. Interior surface of drop pans
4. Cage rack surface
5. Water samples from each of the following-tap water, distilled water, and water from inside of clean bottle

C. Room sanitization procedures are monitored when rooms are broken down, following a complete sanitizing procedure. One room should be monitored every six months.

#### D. Monitoring Procedures

1. A RODAC plate (Remel Microbiological Products) containing Dey-Engley neutralizing agar will be used for each sampling site specified above.
2. Plates are labeled with appropriate information; room number, location of sampling and date.
3. Plates are applied directly to the specified areas following completion of the applicable sanitization procedure. The equipment must be dry and cool. It's important to use careful technique to avoid contamination of the plate with hands, clothes or any object.
4. RODAC plates are incubated at 37°C for 24 hours.
5. After the 24 hour incubation period, if colonies are present, they are counted and gram stained.

#### E. Interpretation of Results

Proper sanitization should eliminate agents that may affect animal health. Growth of small numbers of gram positive cocci may require improvement of sanitization methods but probably disinfection is sufficient enough to prevent a health risk to the animals. Presence of gram negative bacteria may require additional monitoring and evaluation of sanitization procedures.

#### F. Recording of Results

Results from RODAC plate monitoring are recorded by the OAR Animal Health Technician.

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#### G. Monitoring Sterilizing Procedures in Steam Autoclave

Individual Principal Investigators chose to sterilize bedding and cages in the Unit B sterilizer for precautionary reasons. The animals are conventionally housed and are not changed under a hood. The animals are not immuno-compromised. A test strip is included in one load of cages that is run in the sterilizer once a week.

#### G. Monitoring Cage Washer Temperature

The Steris 9500 Cage and Rack Washer is programmed to run at a set wash and rinse cycle. The rinse cycle is set to run for 3 minutes at 180° F and will go into alarm if this temperature is not met within a set amount of time. The doors on the clean side of the cage room will not open if the cycle is not complete. Wahl temp-plate @150-180° F tapes are run in the cage washer a minimum of once a week.